RESEARCH ARTICLE



Sphaerodoridae (Annelida) of the deep Northwestern Atlantic, including remarkable new species of Euritmia and Sphaerephesia

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Abstract

Sphaerodoridae (Annelida) is a seeming uncommon and minimally diverse group of polychaetes in the northwestern Atlantic, with only seven species reported from the United States, and none from the eastern coast of Canada, before the present study. Review of the large Smithsonian collection (National Museum of Natural History, Washington) revealed the presence of two morphologically extraordinary undescribed species and added a new record to the north-western Atlantic region. *Euritmia carolensis* sp. n. is characterised by bearing approximately 20 sessile spherical papillae arranged in three transverse rows per segment, ventrum with 4–6 larger papillae near the parapodial bases and parapodia without papillae; bearing 4–5 simple chaetae that are enlarged subdistally. *Sphaerephesia amphorata* **sp. n.** is distinguished from other congeners in the presence of four longitudinal rows of sessile, bottle-shaped macrotubercles with exceptionally long digitiform terminal papilla, and parapodia with four rounded and small papillae, bearing 4–7 compound chaetae, with blades 7–11 times as long as wide. Other encountered species are also herein re-described, including intraspecific variation and updated iconography. Comparison of material also allowed some systematic changes in the group, including the synonymisation of the genus *Amacrodorum* with *Euritmia*, and the transfer of *Ephesiopsis shivae* to *Ephesiella*. A key to the species reported from the Northwestern Atlantic is provided.

Keywords

Generic synonymy, Amacrodorum, Ephesiopsis, epithelial tubercles, NW Atlantic, shelf, slope

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Introduction

The northwest Atlantic (considered herein as the continental shelf and slope areas off Atlantic Canada and New England) is a relatively well-studied area in terms of its benthic polychaete fauna (e.g. Pettibone 1952, Hartman 1965, Blake 1971, Hartman and Fauchald 1971, Maciolek 1981, 2000, Hilbig and Blake 1991, Frame 1992, Schaff et al. 1992, Blake and Grassle 1994, Hilbig 1994). The dominant taxonomic group in these environments, representing nearly half of the species of the infauna, is the Annelida (Hilbig and Blake 1991, Grassle and Maciolek 1992, Schaff et al. 1992, Blake and Grassle 1994). Sphaerodoridae, a polychaete family typically known to inhabit deep sediments, were neither particularly abundant nor diverse (Hilbig and Blake 1991, Schaff et al. 1992, Blake and Grassle 1994). Seven nominal species have been reported from the area (Table 1), together with probably another eight undescribed species (Hilbig and Blake 1991). Surprisingly, all the sphaerodorids known to the northwest Atlantic have been reported from the United States waters, and none has yet been recorded from Canada (Pocklington and Tremblay 1987, Carr 2012).

Revision of the sphaerodorids deposited at the Smithsonian collection, National Museum of Natural History, Washington, revealed undescribed species fitting the definition of *Euritmia* Sardá-Borroy, 1987 and *Sphaerephesia* Fauchald, 1972, but presenting noteworthy attributes never described in any member of either of these two groups. *Euritmia hamulisetosa* Sardá-Borroy, 1987, type species of the genus, was described from shallow subtidal environments in southern Spain. *Euritmia capense* (Day, 1963) is so far considered the only congener and reported from South Africa (Day 1963, Sardá-Borroy 1987). Members of this genus are characterised by the absence of macrotubercles (i.e. large tubercles arranged in longitudinal rows) on the dorsum, absence of microtubercles (i.e. tubercles provided with a terminal papillae and a basal collar) on the dorsum, presence of epithelial papillae (smaller than macrotubercles) and arranged in various transversal rows per segment, and the presence of only simple chaetae on every parapodia (Sardá-Borroy 1987, Fauchald 1974). These attributes are all shared by *Amacrodorum* Kudenov, 1987a (Capa et al. 2014), which gives reasons to investigate the validity of both genera.

Members of *Sphaerephesia*, a genus with nine nominal species described to date, are recognised by the presence of macrotubercles with a terminal papilla and compound chaetae (Fauchald 1972, 1974; Capa and Bakken 2015). However, the terminal papillae are not often evident and instead, pear shaped tubercles have been observed in some species (see Capa et al. 2014, Capa and Bakken 2015). This is not the case for the new species of *Sphaerephesia* described herein, which is provided with an exceptional digitiform papilla on all dorsal macrotubercles.

For this study, specimens of previously described species from the area were also examined, allowing re-description of *Ephesiopsis guayanae* Hartman & Fauchald, 1971, *Sphaerodoridium minutum* (Webster & Benedict, 1887) and *Sphaerodoropsis corrugata* Hartman & Fauchald, 1971. Each of these species is also peculiar and inhabit the northwestern Atlantic. In the process of re-describing these species we comment on the morphological intraspecific variability observed.

Species	Type locality
<i>Clavodorum atlanticum</i> Hartman & Fauchald, 1971	Northwest of Bermuda, 37°59.2'N, 69°26.2'W, 3834 m depth
<i>Ephesiella macrocirris</i> Hartman & Fauchald, 1971	Off New England, 39°46.5'N, 70°43.3'W, 1470–1330 m depth.
<i>E. mixta</i> Hartman and Fauchald, 1971	Off New England, 38°33'N, 68°32'W, 3753 m depth.
Sphaerororidium minutum (Webster & Benedict, 1887)	Off New England, mainly shelf depths
Sphaerodoropsis corrugata Hartman & Fauchald, 1971	Off New England, 39°56'30"N, 70°39'54"W, 400 m depth.
<i>S. elegans</i> Hartman & Fauchald, 1971	Off Brazil, 00°03.0'S, 27°48.0'W, 3730–3783 m depth.
<i>S. longipalpa</i> Hartman & Fauchald, 1971	Bermuda Slope, 32°16'36"N, 64°36'18"W, 1700 m depth.

Table 1. Species recorded in northwestern Atlantic shelf, slope and abyssal depths.

This study does not intend to represent an exhaustive taxonomic account of the sphaerodorid fauna of the northwest Atlantic. Instead, the aim is to highlight the need of further benthic surveys and taxonomic revisions of the material housed in museum collections in order to increase the knowledge of the biodiversity inhabiting the northwestern Atlantic Ocean floor, including the description of new species with remarkable morphological characteristics that may assist in understanding the morphological variation of the family.

Methods

Specimens deposited in the collections of the Smithsonian Institution, National Museum of Natural History, Washington (USNM) mainly collected in the Atlantic Slope and Rise Program (ASLAR) were examined. Holotypes and comparative material from other institutions (Australian Museum (AM), Sydney; Zoologisches Museum (ZMH), Hamburg; Natural History Museum of Los Angeles County (LACM-AHF), Los Angeles, and Museu de Zoologia Universidade de São Paulo (MZSP), São Paulo) were also studied. The material was fixed in formalin and preserved in 70–80% ethanol. Specimens were examined under dissecting and compound microscopes.

Methylene-blue staining was used to highlight glandular areas and papillae by immersing selected specimens in 70–80% ethanol with some dissolved crystals of the compound for several minutes. Micrographs were taken with a Leica DFC 420 camera attached to a Leica MZ 16A stereo microscope and a Leica DM 6000B compound microscopes (Leica Microsystems, Wetzlar, Germany). Stacks of multi-focus shots were merged into a single photograph to improve resolution with Leica Application Suite v3.7 software (Leica Microsystems, Wetzlar, Germany). Some parapodia were mounted on microscope slides with glycerine.

Scanning electron micrographs were taken on specimens after dehydrating them in a series of 70, 80, and 90% ethanol and series of mixtures of absolute ethanol and Hexamethyldisilazane (HMDS) with the following ratios 2:1, 1:1, 1:2, and then into pure

HMDS. The prepared samples were mounted on holders, sputter-coated with gold (10 nm thickness). The micromorphology and topography were determined using a Philips FEI INSPECT (Hillsboro, Oregon, USA) scanning electron microscope (SEM) at the Museo Nacional Ciencias Naturales (Madrid, Spain) and a JEOL-JSM-6480 SEM at the Cellular and Molecular Imaging Core Facility (CMIC) of the Faculty of Medicine of the Norwegian University of Science and Technology (NTNU). The samples were observed with the Back Scattering Electron Detector (BSED) with a resolution at high vacuum of 4.0 nm at 30 kV. The accelerating voltage was 30 kV and working distance of 10 mm to the detector.

A key for species identification was generated after consideration of the species reported from the northwestern Atlantic (with an asterisk) and other from adjacent geographic regions such as the Gulf of Mexico and the Caribbean (considering recent reviews e.g. Kudenov 1987b, 1994, Salazar-Vallejo 1996, Fauchald et al. 2009) in view that some of these species, especially those typical from deeper environments, could be present in the area. The features used in this key correspond mostly to original descriptions except for the material reviewed in the present study. Species originally described in distant disjunct geographic areas and whose identifications are dubious have been left out of the key (e.g. *Sphaerodoridium claparedii* (Greeff, 1866) and *Sphaerodorum ophiurophoretos* Martin & Alvà, 1988 or those currently not considered belonging to the family (e.g members of *Levidorum* Perkins, 1987 or *Sphaerodoridium guilbaulti* Rullier, 1974).

Abbreviations used on the figures: 1st, first chaetiger; al, acicular lobe; ap, antenniform papillae; eg, egg; la, lateral antenna; ma, median antenna; mc, macrotubercle; mi, microtubercle; mo, mouth; no, nuchal organ; pa, palp; tc, tentacular cirrus; vc, ventral cirrus.

Systematics

Ephesiopsis Hartman & Fauchald, 1971

Ephesiopsis Hartman & Fauchald, 1971: 68; Fauchald 1974: 270; Rizzo 2009: 62–63 (in part); Capa et al. 2014.

Type species. Ephesiopsis guayanae Hartman & Fauchald, 1971.

Diagnosis. Body long and slender. Two longitudinal rows of macrotubercles, one pair per segment, absent on first chaetiger. Macrotubercles sessile, with terminal papillae. Two longitudinal rows of microtubercles, one pair per segment, running parallel between macrotubercles. Additionally, papillae arranged in 4–5 transverse rows on dorsum and ventrum. Prostomial and peristomial appendages short, spherical or digitiform. Parapodia from chaetiger 2 with both simple and compound chaetae; hooks on first chaetiger absent or present.

Remarks. The genus was originally erected and justified by the presence of both simple and compound chaetae in every chaetiger (Hartman and Fauchald 1971, Fauchald 1974), a condition different to that found in members of *Ephesiella* Chamberlin, 1919, with only compound or pseudo-compound chaetae (Moore 1909, Fauchald 1974, Capa and Bakken 2015, Capa et al. 2014, 2016), or Sphaerodorum Örsted, 1843, with typically only simple chaetae (Fauchald 1974, Capa et al. 2014, 2016). Otherwise, these three genera are very similar and they all share the general body shape, with slender bodies, the number and arrangement of epithelial tubercles, with two longitudinal rows of macrotubercles with a terminal papilla, two longitudinal rows of microtubercles, and about 3-4 transverse rows of smaller papillae on each segment. The re-examination of the types of the type species, *Ephesiopsis guayanae* Hartman & Fauchald, 1971, confirms the presence of sub-distally widened simple chaetae, with tapering tips and straight edges, and with apparently no sign of being compound under the light and compound microscopes (Fig. 1A, B); while an additional chaeta in same parapodia is compound, with thinner blades and with more rounded edges. It is unclear at this point, if the presence of both compound and apparently simple chaetae justifies the validity of the genus and further analyses should be performed to elucidate this issue.

An examination of the types of the recently described *Ephesiopsis shivae* Rizzo, 2009 from Brazil revealed that the specimens do not show the typical generic attributes (Rizzo 2009, Fig. 3A–C, E–F). The chaetae considered in the original description as simple seem to be compound chaetae that have lost the blades, as it has been observed in many specimens of *Ephesiella* spp. Moreover, *E. shivae* was described as having a pygidium with four macrotubercles (Rizzo 2009), but the examined Brazilian specimens showed the typical two dorsal macrotubercles and the ventral digitiform pygidial cirrus. Based on this observation, we propose that *E. shivae* is transferred to *Ephesiella* and conclude that *Ephesiopsis* is monotypic.

Ephesiopsis guayanae Hartman & Fauchald, 1971

Figs 1, 2

Sphaerodorum sp. C.– Hartman 1965: 96, pl. 14, Figs A–B. Ephesiopsis guayanae Hartman & Fauchald, 1971: 68–69, Pl 33, figs A–G.

Material examined. Holotype: LACM-AHF POLY TYPE 942, off Dutch Guayana Surinam, 07°52'N, 54°31.5'W, 520–550 m, coll. Woods Hole Oceanographic Institution, 25 Apr 1963. **Paratype:** same collection information LACM-AHF POLY TYPE 943 (1 ind.).

Additional material. USNM 1001772 (2 ind.), Georges Bank, 40°57.21'N, 066°13.68'W, coll. MMS Collections, Atlantic Slope and Rise Program, ASLAR, 25 Jul 1986; USNM 1001773 (1 ind.) off New Jersey, 38°35.98'N, 072°52.86'W, 2195 m, coll. MMS Collections;



Figure 1. *Ephesiopsis guayanae* paratypes LACM-AHF POLY TYPE 943, micrographs. A Mid-body chaetigers and chaetae (apparently simple), ventral view **B** Mid-body chaetigers with compound and simple chaetae, dorsal view.

USNM 1001777 (1 ind.), off New Jersey, United States, 2150 m, 1 Dec 1984, coll. MMS Collections, Atlantic Slope and Rise Program, ASLAR; USNM 1001713 (1 ind.), off Cape Lookout, North Carolina, 34°11.16'N, 75°38.98'W, 2006 m, coll. MMS Collections, Atlantic Slope and Rise Program, ASLAR, 14 Jul 1984; USNM 1001780 (1 ind.), off Delmarva, 37°51.58'N, 73°19.914'W, 2100 m, coll. MMS Collections, Atlantic Slope and Rise Program, ASLAR, 30 Nov 1984; USNM 1001781 (1 ind.), off New Jersey, 38°29.28'N, 72°42.11'W, 2507 m, coll. MMS Collections, Atlantic Slope and Rise Program, ASLAR, 4 Dec 1984; USNM 1001782 (1 ind.), off New Jersey, 38°29.23'N, 72°42.19'W, 2505 m, coll. MMS Collections, Atlantic Slope and Rise Program, ASLAR, 18 May 1985; USNM 1001783 (1 ind.), off New Jersey, 38°29.23'N, 72°42.19'W, 2505 m, coll. MMS Collections, Atlantic Slope and Rise Program, ASLAR, 18 May 1985; USNM 1001789 (1 ind.), Baltimore Canyon, Maryland, 37°53.76'N, 73°44.76'W, 1499 m, coll. MMS Collections, Atlantic Slope and Rise Program, ASLAR, 15 Nov 1985.

Comparative material. *Ephesiopsis shivae*, holotype MZSP883 24°07.637'S 45°51.895'W, 09 Jan 1998, Sta. 6661, 147 m, Santos/São Paulo to Ilha Grande Bay/Rio de Janeiro; paratypes MZSP1031 (2 ind.), 24°07.637'S, 45°51.895'W, 09 Jan 1998, Sta. 6661, 147 m, Santos/São Paulo to Ilha Grande Bay/Rio de Janeiro.

Diagnosis. Palps and lateral antennae digitiform, median antenna shorter. Tentacular cirri ellipsoid. Parapodia with 4–6 parapodial papillae; compound chaetae with blades 1.5–2.5 times as long as maximum width on mid-body chaetigers, simple chaetae wider and with angular silhouette; hooks present on first chaetiger.

Re-description. *Measurements and general morphology.* Holotype 2.2 mm long, 0.2 mm wide, with 26 chaetigers, divided in two. Body elongated, sub-quadrangular in section, with slightly convex dorsum. Anterior end bluntly rounded, slightly narrowing along posterior segments. Segmentation inconspicuous, tegument with transverse wrinkles. Preserved specimen lacking pigmentation.

Head. Prostomium with five short appendages, including a pair of digitiform palps in ventral-most position, a pair of lateral antennae, similar in shape and size to palps,



Figure 2. *Ephesiopsis guayanae*, USNM 1001777, SEM. **A** Whole specimen, side view **B** Anterior end, side view **C** Chaetiger 3, with parapodia, macrotubercle and microtubercle, side view **D** Mid-body segments, side view **E** Mid-body parapodium showing detail of parapodial papillae, acicular lobe and ventral cirrus **F** Posterior chaetigers and pygidium with paired dorsal cirri and ventral digitiform anal cirrus, side view **G** Chaetae chaetiger 4 **H** Chaetae mid-body fascicle **I–L** Chaetae mid-body chaetigers. Abbreviations: 1st mc, macrotubercle from first chaetiger; al, acicular lobe; la, lateral antenna; ma, median antenna; mc, macrotubercle; mi, microtubercle; mo, mouth; no, nuchal organ; pa, palp; tc, tentacular cirrus; vc, ventral cirrus.

and a median antenna, shorter (one third) and thinner than lateral antennae (Fig. 2A, B). A pair of tentacular cirri shorter than lateral antennae and palps. A few rounded (about six) small papillae confined by head appendages.

Tubercles. First chaetiger with two dorsal macrotubercles; microtubercles absent (Fig. 2B). Following chaetigers each with two dorsal macrotubercles arranged in two dorso-lateral longitudinal rows, and two microtubercles forming two longitudinal rows between the macrotubercles (Fig. 2B–D, F). Macrotubercles sessile and spherical each provided with a digitiform terminal papilla (Fig. 2B–D, F); with groups of pores around terminal papilla. All macrotubercles similar in shape and size. Microtubercles with digitiform terminal papilla generally longer than collar (Fig. 2C). Spherical or ellipsoid papillae over dorsum, arranged in 3–4 transversal rows per chaetiger, with around 15–20 papillae on each mid-body chaetigers. Ventral surface with spherical papillae, arranged in four more or less regular transversal rows, with about 20 per segment, in mid-body; numbers decreasing towards posterior end (Fig. 2F). Body epithelium with ellipsoid granules.

Parapodia. Parapodia sub-conical, increasing in size towards chaetiger 3 (Fig. 2B), around 1–3 times longer than wide (Fig. 2C–E). Acicular lobe projecting distally anterior to chaetae, resembling other parapodial papillae or slightly longer (Fig. 2E). Ventral cirri digitiform projecting as long as acicular lobe on anterior segments or shorter in mid-body and posterior ones (Fig. 2C–E). Mid-body parapodia with 4–5 ellipsoid to digitiform papillae, all similar in size, in addition to the acicular lobe: 2–3 anterior, 1–2 posterior-ventral (Fig. 2C–E).

Chaetae. First chaetiger with two pairs of hooks, one pair on each parapodia together with elongate simple chaetae. One compound and 2–4 simple chaetae in all chaetigers, arranged in a curved transverse row around acicular lobe (Figs 1A, B, 2G– L). First and second chaetigers with slightly serrated long blades, 4–5 times longer than wide. Chaetae from chaetiger 3 with shafts widened sub-distally, a larger distal tooth and fine spinulation; blades twice as long as wide (Fig. 2G–L).

Pygidium. Pygidium terminal, with one mid-ventral digitiform anal cirrus and a pair of dorsal anal cirri, similar in shape but slightly smaller than macrotubercles (Fig. 2F).

Internal features. A pair of eyes anterior to first chaetiger.

Reproductive features. Copulatory organs or eggs not seen in holotype. Paratype with eggs in coelomic cavity.

Variation. The paratype, an incomplete gravid female, is larger than holotype, 3 mm long and 0.25 wide, with 44 chaetigers. The specimen from New Jersey is 2 mm long and 0.25 wide. The number and morphology of chaetae showed variation among the material examined. The holotype possessed two pair of hooks on first chaetiger, absent or not seen in the paratype while the specimen from New Jersey had one pair (Fig. 2B). Additionally, an extra set of thin and simple chaetae, resembling those present in rest of chaetigers were observed only on the holotype. Following chaetigers had fascicles with 3–5 chaetae, one of them clearly compound in the holotype, two in the paratype and up to three in the specimen from New Jersey. Chaetae appearing compound under the light microscope (Fig. 1B) did not clearly seem so under the electron

microscope (Fig. 2G, I, J), but at least a faint edge between the shaft and the blade could be noticed. The simple chaetae, probably a result of the fusion of shaft and the blade, show in some cases a different outline, with an angular edge, and a less curved tip (Fig. 2H, L).

Remarks. The most remarkable attribute of this species is the presence of simple chaetae, in all parapodia, that are flat and sub-distally widened and have an angular contour. This differs from other members of the long bodied sphaerodorids (*Ephesiella* and *Sphaerodorum*), where chaetae, simple or compound respectively, present more rounded edges. In this respect, the holotype and paratypes of *E. guayanae* show some morphological differences, which, if considered as part of the intraspecific variation, may open a discussion of the legitimacy for the genus. These special, simple, sub-distally widened and flat chaetae were not conspicuous in the paratype. The apparently simple chaetae present in both holo- and paratype do not differ much from those pseudocompound chaetae present in other *Ephesiella* (e.g. *Ephesiella brevicapitis* Moore, 1909). With so little material in hand, only two types, it is difficult to conclude on the status of the genus and the identity of the paratype. The species seems not to have been found again, until now.

The chaetae of the specimens collected from sediments of deep New Jersey waters resemble those observed in the paratype of *E. guayanae*. In every parapodium a group of simple chaetae can be observed, they are not so wide and angular as those present in the holotype, but still they appear to be simple chaetae. Examination of these specimens under SEM revealed a faint oblique mark in the position where an articulation between shaft and blade is expected, and some chaetae seem to be bent at this point. Together these observations suggest that they may be pseudocompound chaetae (Fig. 2G–L). This cannot be addressed in the type material, which needs to be left intact.

Type locality. Surinam, Dutch Guayana, 520-550 m.

Distribution. From Dutch Guayana to New Jersey, 520–2507 m.

Euritmia Sardá-Borroy, 1987

Euritmia Sardá-Borroy, 1987: 48; Capa et al. 2014. *Amacrododrum* Kudenov, 1987a: 917–918.

Type species. Euritmia hamulisetosa Sardá-Borroy, 1987.

Diagnosis. Body short and ellipsoid. Macro- and microtubercles absent; papillae all over body surface and parapodia. Prostomial and peristomial appendages short, spherical or digitiform. Parapodia with simple chaetae, enlarged sub-distally, with serrated cutting edges; hooks absent.

Remarks. This genus was erected to accommodate a small and atypical species with the body covered by numerous papillae and bearing simple chaetae, *Euritmia hamuli-setosa* Sardá-Borroy, 1987, from southern Spain, and a species previously described as *Sphaerodorum, Euritmia capense* (Day, 1963), from South Africa (Sardá-Borroy 1987).

Euritmia capense is distinguished from E. hamulisetosa in the number, arrangement and type of dorsal epithelial papillae, having two transverse rows of each large and small papillae per segment instead of four rows of similar papillae; and the chaetal morphology with a typical distal spine in *E. hamulisetosa*, absent in *E. capense* (Day 1963, 1967; Sardá-Borroy 1987). A few months later, another genus was erected to accommodate a similar species, Amacrodorum bipapillatum Kudenov, 1987a. In addition to the absence of large tubercles and the presence of simple chaetae, this species was characterised by the presence of two kinds of epithelial papillae, spherical and ellipsoid. The latter author was probably not aware of the description of Euritmia and also did not notice the diagnostic features of the South African species, similar to A. bipapillatum. Amacrodorum *bipapillatum* is distinguished from the two previously described species in the number of transverse rows of dorsal papillae and the arrangement of the ellipsoid and hemispherical papillae. The new species described herein shares with the other three species the absence of macro- and microtubercles, the presence of several rows of papillae over the body surface and the presence of distally hooked simple chaetae. For these reasons, all these species are considered as members of the genus Euritmia and united under this name. Being an older name than *Amacrodorum* and applying the Principle of Priority (Article 23, International Code of Zoological Nomenclature), *Euritmia* takes priority.

The genus currently gathers four species, distinguished mainly by the shape and arrangement of dorsal and ventral papillae, the number and arrangement of parapodial papillae and the shape of the chaetae (Table 2). This latter attribute has only been studied under SEM in *Euritmia hamulisetosa*, and therefore is the best known in the group; it is probable that the reported smooth chaetae in the other species would show some thin spinulation if observed under high magnification.

Moreover, a group of species to date considered belonging to *Sphaerodoropsis* (Group 4, according to Borowski 1994) also show these features, although the chaetae, instead of being simple, could be considered as pseudocompound. These are *Sphaero-doropsis multipapillata* (Hartmann-Schröder, 1974), *Sphaerodoropsis heteropapillata* Hartmann-Schröder, 1987, and *Sphaerodoropsis plurituberculata* Capa & Rouse, 2015. *Commensodorum commensalis* (Lützen, 1961) shares the type of chaetae with *Euritmia*, being typically simple. Nevertheless, *C. commensalis* has few and small macrotubercles, consisting of four macrotubercles arranged in a simple transverse row per segment. It is still unclear if these taxa are closely related and if the potential fusion of shafts and blades to form simple chaetae (like in *Sphaerodorum, Commensodorum*, and *Euritmia*) occurred more than once within the group.

Euritmia carolensis sp. n.

http://zoobank.org/BCB8304D-75F9-4566-A619-6BA3606B6469 Figs 3, 4

Material examined. Holotype: USNM 1001792, Off Charleston Bump South Carolina United States 32.3944N, 77.0181W, 799 m, coll. Battelle/Woods Hole Oceano-

Table 2. Comparative features (of members of <i>Euritmia</i> , from or	iginal descriptions.		
	Euritmia hamulisetosa	Euritmia capense	Euritmia bipapillatum	Euritmia carolensis sp. n.
Source	Sardá-Borroy (1987)	Day (1963)	Kudenov (1987a)	this study
Type locality	Cadiz, Spain	Cape Town, South Africa	Alaska, US	South Carolina, US
Depth	Intertidal	۸.	59 m	799 m
Size (length x width)	0.6×0.125 mm	2.5×0.8 mm	2.1×0.5 mm	0.5×0.2 mm
Number of chaetigers	14	16	16	12
Prostomial appendages	3 pairs + median antenna	not distinguishable	two pairs (longer) + median antenna. Lat ant. with spurs	two pairs (longer) + median antenna
Eyes	two pairs	one pair	one pair	not observed
Shape of dorsal papillae in mid-body	spherical, all similar in size	spherical, two sizes	two kinds and sizes, hemispherical and ellipsoid	two kinds, hemispherical and ellipsoid
Number of dorsal papillae in mid-body	four transversal rows	two transverse rows	three transversal rows	three transversal rows
Ventral papillae	four transversal rows	۰.	three transversal rows, elliptical and spherical	two transversal rows
Parapodial papillae	one dorsal, one ventral	one dorsal, one ventral and three smaller ones on anterior and posterior surfaces	one on anterior surface	none
Chaetae mophology	serrated, hooked and with distal spine	smooth, hooked	smooth, hooked	smooth or finally serrated, hooked
Number of chaetae per parapodium	six	ten	four or five	five

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Figure 3. *Euritmia carolensis* sp. n., paratype, USNM 1001791, micrographs. **A** Whole body, side view **B** Dorsal papillae of different sizes, anterior segments **C** Detail of a large dorsal papillae **D** Mid-body simple chaetae **E** Same. Abbreviations: la, lateral antenna; pa, palp.

graphic Institute for BLM/ MMS, Atlantic Slope and Rise Program, ASLAR, 18 Nov 1985. **Paratypes:** USNM 1001791 (1 ind.), off Cape Fear, North Carolina, 33.0806N, 76.4194W, 896 m, 23 Sep 1985; USNM 1001793 (1 ind.), off New Jersey, 38.5944N, 72.8953W, 2024 m, 13 Nov 1985.

Type locality. Off Charleston Bump, South Carolina, 799 m.

Diagnosis. Body short and ellipsoid. Dorsum with approximately 20 sessile spherical papillae arranged in three transverse rows per segment. Ventrum with 4–6 larger papillae near the parapodial bases. Prostomial and peristomial appendages short and ellipsoidal. Parapodia without papillae; with 4–5 simple chaetae with serrated cutting edges, enlarged sub-distally.

Description. *Measurements and general morphology.* Holotype 0.5 mm long, 0.2 mm wide, with 12 chaetigers; gravid female. Body ellipsoid, with strongly convex dorsum and flattened ventrum. Epithelium with transversal wrinkles, segmentation not noticeable (Fig. 4A, B). Pigmentation absent on preserved material.

Head. Head fused to first chaetiger (Figs 3A, 4A, B). Prostomial appendages ellipsoid, slightly longer than wide (Figs 3A, 4A). Pair of palps and pair of lateral antennae similar in size, median antenna smaller (Fig. 4B). Dorsal antenniform papillae absent or not conspicuous. Few small hemispherical papillae scattered on head surface, approximately four arranged among palps and antennae. Tentacular cirri, ellipsoid, smaller than palps, similar in size and shape to median antenna (Fig. 4A). Eyes not observed.



Figure 4. *Euritmia carolensis* sp. n., holotype, USNM 1001792, drawings. **A** Anterior end, dorsal view **B** Same, lateroventral view **C** Pygidium, side view **D** Mid-body parapodium, dorsal view **E** Detail of chaeta.

Tubercles. Dorsum with three transverse rows of papillae per segment (Figs 3A, 4A, B). Papillae sessile, spherical but some larger and ellipsoid papillae over dorsum (Figs 3C–E, 4A), over 20 per segment on mid-body chaetigers. Ventrum with fewer and larger spherical papillae, arranged near parapodial bases in two transverse rows, about 4–6 papillae per segment (Fig. 4B).

Parapodia. Parapodia conical, as long as wide in all chaetigers (Fig. 4D), small in anterior chaetigers. Acicular lobe hemispherical; ventral cirri ellipsoid longer than acicular lobe (Fig. 4D). No additional papillae or parapodial appendages.

Chaetae. Large, recurved hooks in first chaetiger absent. All parapodia with 4–5 simple chaetae; blades seemingly smooth on cutting edge and slight curved distal tip (Figs 3D, E, 4D, E). One straight acicula per parapodium.

Pygidium. Pygidium with two dorsolateral pear-shaped tubercles and single midventral digitiform cirrus, longer than lateral cirri (Fig. 4C).

Internal features. Muscular pharynx visible though body wall from chaetiger 2–5.

Reproductive features. Copulatory organs not observed. Ovoid eggs measuring 100 µm diameter occupy most of the coelomic cavity.

Variation. The two paratypes lack eggs, other features are very similar.

Remarks. *Euritmia carolensis* sp. n. is distinguished from other congeners by the presence of dorsal papillae arranged in three transverse rows per segment. Most of these are spherical and similar in size but some in a dorsal-most position are elliptical and larger, without a clear distribution pattern. *Euritmia hamulisetosa* is provided with

spherical papillae, all similar in size, in four transverse rows per segment; *E. capense* bears two different sizes of spherical papillae each on a single transversal row per segment; and *E. bipapillatum* bears both hemispherical and elliptical papillae in three transversal rows per segment and with a particular zig-zag arrangement (Day 1963, 1967, Sardá-Borroy 1987, Kudenov 1987a) (Table 2). The number and arrangement of ventral papillae are also distinct in the new species, with two transverse rows of 4–6 papillae, mainly arranged near the parapodial bases, while the other species (unknown for *E. capense*) bear numerous papillae with similar arrangement to the dorsum. The parapodia are in all cases conical and small, but only *Euritmia carolensis* sp. n. lacks parapodial papillae; the only appendices present on parapodia being acicular lobe and the ventral cirrus.

Etymology. The epithet of this species refers to the type locality North Carolina. **Distribution.** North Carolina to New Jersey (US), from 799 to 2014 m.

Sphaerephesia Fauchald, 1972

Sphaerephesia Fauchald, 1972: 97; Magalhães et al. 2011: 40; Capa et al. 2014.

Type species. Sphaerephesia longisetis Fauchald, 1972.

Diagnosis. Body short and ellipsoid, some species slender. Four or more longitudinal rows of sessile macrotubercles with terminal papillae. Microtubercles absent (?). Papillae over body surface and parapodia. Prostomial and peristomial appendages short, spherical or digitiform. Parapodia with compound chaetae; hooks absent.

Remarks. There are two species in the genus described as presenting microtubercles (tubercles consisting of a basal collar and a terminal papillae) on the lateral or dorsolateral side of the body, but these differ from those typically present in the long-bodied sphaerodorids (i.e. *Ephesiella, Ephesiopsis* and *Sphaerodoridium*).

Sphaerephesia amphorata sp. n.

http://zoobank.org/9FC1C233-CBF8-4669-BD62-BF4B59CAD091 Figs 5, 6

Material examined. Holotype: USNM 1001815, East of Cape Lookout, North Carolina, United States, north Atlantic, 34°16.32'N, 75°45.498'W, 640 m, 11 Nov 1983, MMS Collections, Atlantic Slope and Rise Program, ASLAR (CRSAP 1 st.1 rep. 2 core 6 sec 0.2). **Paratypes:** USNM USNM 1407168, same sample, different cores (7 ind.). **Additional material.** USNM 1001801 (4 ind.), East of Cape Lookout, North Carolina, United States, North Atlantic, 34°16.002'N, 75°45.967'W, 580 m, 11 Nov 1983; USNM 1001802 (5 ind.), East of Cape Lookout, North Carolina, 34°15.936'N, 75°46.164'W, 583 m, 26 Mar 1984; USNM 1002041 (9 ind., 3 for SEM), East of Cape Lookout, North Carolina, 34°15.816'N, 75°45.786'W, 593 m,



Figure 5. *Sphaerephesia amphorata* sp. n., paratypes, USNM 1002041, SEM. **A** Whole specimen, dorsal view **B** Same, ventral view **C** Anterior end, showing head appendages, dorsal view **D** Same, with head papillae visible **E** Same, lateroventral view; mouth and nuchal organ pits, visible **F** Anterior end, ventral view **G** Posterior chaetigers, dorsal view, showing the characteristic dorsal macrotubercles with long terminal papillae **H** Mid-body chaetigers, ventral view. Abbreviations: lateral antenna; ma, median antenna; mo, mouth; no, nuchal organ; pa, palp; tc, tentacular cirrus.

27 Mar 1984; USNM 1002196 (1 ind.), East of Cape Lookout, North Carolina, 34°49.8'N, 75°13.5'W, 2003 m, 20 Jul 1985.

Comparative material. *Sphaerephesia fauchaldi* Kudenov, 1987b, holotype NMNH 102785; *Sphaerephesia longisetis* Fauchald, 1972, holotype AHF POLY 0964; *Sphaerephesia regularis* Böggemann, 2009, holotype ZMH P25498, paratypes

ZMH P25497 (4 ind.), ZMH P25499 (4 ind.); *Sphaerephesia similisetis* Fauchald, 1972, paratype AHF POLY 0967 (2 ind.). *Sphaerephesia hutchingsae* Capa and Bakken, 2015, holotype: AM W.42748, East of Malabar, Sydney, New South Wales, Australia, 33°58.717S', 151°18.00'E, 82 m, 22 Aug 1995; paratypes: east of Malabar, Sydney, New South Wales, 80–28 m. AM W.42717 (1 ind.), AM W.42721 (1 ind.), AM W.42731 (2 ind.), AM W.42749 (1 ind.), AM W.42751 (1 ind.), AM W.42752 (2 ind.), AM W.42758 (1 ind.).

Diagnosis. Four longitudinal rows of sessile, bottle-shaped macrotubercles with long digitiform terminal papilla and 3–4 transverse rows of small spherical papillae per segment. Microtubercles absent. Distance between dorsal-most macrotubercles similar to distance between those and lateral ones. Parapodia with ventral cirri nor surpassing length of acicular lobe and four rounded and small papillae. Parapodia with 4–7 compound chaetae, with thin shafts and blades 7–11 times as long as wide.

Description. *Measurements and general morphology.* Gravid female, 1.5 mm long, 0.2 mm wide, with17 chaetigers. Body elongated, tapering at both ends, slightly flattened dorso-ventrally (wider than high). Dorsum convex and ventrum flattened (Fig. 5A, B). Tegument with transverse wrinkles and segmentation not obvious (Fig. 5A). Preserved specimen lacking pigmentation.

Head. Anterior end bluntly rounded (Fig. 5A, C–F). Prostomium with seven longer appendages including a pair of palps, in ventral-most position near the mouth, a pair of lateral antennae and a median antenna; and a pair of antenniform papillae behind lateral antennae, all digitiform, slightly wrinkled and similar in size except for the antenniform papillae, slightly shorter than lateral antennae (Fig. 5C–F). Approximately 10 digitiform papillae confined by prostomial appendages and mouth, in frontal view (Fig. 5D, F). A pair of tentacular cirri, similar in shape and size to lateral antennae and palps, and several scattered papillae similar to prostomial in head surroundings. Nuchal organ pits located between lateral antennae and tentacular cirri (Fig. 5E).

Tubercles. First and last chaetigers with two macrotubercles, sessile, bottle-shaped and provided with a long terminal papilla (Figs 5A, C, D, 6E). Rest of chaetigers with four macrotubercles each, arranged in four longitudinal rows along dorsum (Figs 5C, 6A, B). Distance between mid rows and lateral rows similar (Fig. 5A–B). Shape and size of all macrotubercles similar, slightly decreasing in size in last chaetigers (Fig. 5A). Spherical or ellipsoid papillae present over dorsum, arranged in 3–4 transversal rows per segment (Fig. 5A, D, G), around 10 papillae present between mid-macrotubercles and five between these and lateral ones in mid-body segments (Fig. 5A, G). Microtubercles absent. Ventral surface with small ellipsoid papillae, arranged in 3–4 irregular transversal rows (Fig. 5H), with a total of around 12 papillae per segment in mid-body; numbers decreasing towards posterior end (Fig. 6D). Body epithelium with ellipsoid granules (e.g. Fig. 6A).

Parapodia. Parapodia elongated, sub-conical, increasing in size towards chaetiger 4 and around 2–3 times longer than wide (Figs 5B, H, 6A–D). Acicular lobe projecting distally anterior to chaetae (Fig. 6A–D). Ventral cirri sub-conical, pear-shaped, as long as acicular lobe in posterior chaetigers, and shorter in anterior (Figs 5B, H, 6A–E).



Figure 6. *Sphaerephesia amphorata* sp. n., paratypes, USNM 1002041, SEM. **A** Anterior parapodium, dorsal view **B** Mid-body parapodium, dorsal view **C** Mid-body chaetigers, lateroventral view **D** Posterior end, side view **E** Posterior end, dorsal view **F** Chaetae anterior parapodium **G**, **H** Detail of chaetae, mid-body chaetiger. Abbreviations: al, acicular lobe; mc, macrotubercle; vc, ventral cirrus.

Mid-body parapodia with about four small spherical papillae, all similar in size: one distal, on dorsal surface, one anterio-dorsal and one anterio-ventral and another ventral, all in proximal half of parapodia (Figs 5H, 6A–E).

Chaetae. Compound chaetae present in all chaetigers, arranged in a curved transverse row (C-shaped) behind acicular lobe and numbering 4–7 per fascicle (Fig. 6A–D). Shaft with slightly widened distal end with delicate, almost inconspicuous spinulation (Fig. 6G, H). Blades decreasing in size from mid-fascicle to dorsal and ventral

ends (7–11 times longer than maximum width), with fine and short spinulation along cutting edge and a slightly curved tip (Fig. 6G, H).

Pygidium. Pygidium terminal, with mid-ventral digitiform anal cirrus and a pair of dorsal anal cirri, similar in shape to macrotubercles (Fig. 6E).

Internal features. Eyes not observed in any specimen. Muscular pharynx runs along three anterior chaetigers.

Reproductive features. Some paratypes and additional specimens examined are gravid females carrying large discoid eggs, 200 μ m in diameter that occupy most of the body coelom, from the anterior to the posterior segments; other specimens seem to be filled with sperm. However, 'copulatory organs' were not observed in either females or males.

Variation. Paratypes varying in size from 0.8 to 1.5 mm and seven to 17 chaetigers. Most features are conserved in this species and all specimens examined regardless the size bear the dorsal macrotubercles with the unusual elongated terminal papillae as long as the macrotubercle. Length of blades vary within fascicles and also along the chaetigers, generally between 7–11 times longer than maximum width.

Remarks. The most conspicuous and distinct morphological attribute of *Sphaerephesia amphorata* sp. n. is the presence of dorsal macrotubercles with elongated papillae providing them the characteristic amphora shape, while other described species in the genus have a rounded terminal papilla. Six of the nine nominal *Sphaerephesia* species share with *S. amphorata* sp. n. the presence of four rows of macrotubercles with a terminal rounded papilla, several additional papillae on dorsal surface, and falcigers with long blades. These are *Sphaerephesia similisetis* Fauchald, 1972, *Sphaerephesia chilensis* Fauchald, 1974, *Sphaerephesia fauchaldi* Kudenov, 1987b, *Sphaerephesia regularis* Böggemann, 2009 and *Sphaerephesia hutchingsae* Capa & Bakken, 2015. None of the species mentioned share with the new species the number of parapodial papillae; *S. fauchaldi, S. hutchingsae, S. longisetis* and *S. similisetis* bear over six parapodial papillae while *S. regularis* and *S. chilensis* are provided with one or two parapodial papillae (Fauchald 1972, 1974, Kudenov 1987b, Capa and Bakken 2015). Other differences between *S. longisetis, S. fauchaldi* and *S. amphorata* sp. n. are the presence of microtubercles in the former two species, absent in the latter.

Distribution. Only known from type locality, East of Cape Lookout, North Carolina, United States, North Atlantic, ranging from 580 to 2003 m.

Etymology. Amphora, is a Greek word that refers to the pottery vases used since the Bronze Age by the Greco-Romans to transport liquids. The shape of these containers resembles the characteristic shape of the dorsal macrotubercles of this species.

Sphaerodoridium Lützen, 1961

Sphaerodoridium Lützen, 1961: 409–410 (in part), Fauchald 1974: 270, Capa et al. 2014.

Type species. Sphaerodorum claparedii Greeff, 1866

Diagnosis. Body short and ellipsoid. Six or more longitudinal rows of macrotubercles on dorsum, in one transversal row per segment. Macrotubercles stalked and smooth, without terminal papilla. Smaller, stalked tubercles on ventrum. Microtubercles absent. Papillae over body surface and parapodia. Prostomial and peristomial appendages digitiform. Parapodia with only compound chaetae; hooks absent.

Remarks. Capa et al. 2016 recovered *Clavodorum* nested within *Sphaerodoridium*, indicating the circumscription of these two genera according to the relative length of the head appendages should be revised.

Sphaerodoridium minutum (Webster & Benedict, 1887)

Fig. 7

Ephesia minuta Webster & Benedict, 1887: 728–729, pl. IV, figs 64–66.
Sphaerodoropsis minuta.– Imajima 1969: 153–154, fig. 2; Imajima 2009: 77.– Hartmann-Schröder 1996: 237.– Moreira 2012: 39–41, fig. 13.
Sphaerodoum minutum.– Berkeley and Berkeley 1948: 27–28, fig. 34.
Sphaerodoridium minutum.– Lützen 1961: 415.– Capa et al. 2016: 12.

Material examined. Lectotype: USNM 393, Eastport, Maine, United States, North Atlantic Ocean, coll. Webster, H. E. **Paralectotypes:** USNM 1407984 (11 ind. and 4 slides), Eastport, Maine, United States, North Atlantic Ocean, coll. Webster, H. E. **Paratypes:** USNM 22873 (29 ind., 3 for SEM) Eastport, Maine, United States, North Atlantic Ocean, coll. Webster, H. E.

Diagnosis. Palps and lateral antennae digitiform, median antenna shorter and digitiform. Tentacular cirri digitiform. Eyes not observed. Parapodia with three (or four) parapodial papillae; compound chaetae with blades 4–5 times as long as maximum width on mid-body chaetigers.

Re-description. *Measurements and general morphology.* Holotype 1.1 mm long, 0.6 mm wide and with 22 chaetigers. Body ellipsoid, ovoid in cross-section, with slightly flattened ventrum and convex dorsum (Fig. 7A–C). Anterior end blunt, mid-body broad, slightly narrowing along posterior segments (Fig. 7A). Segmentation inconspicuous, tegument with transverse wrinkles (Fig. 7A–C). Preserved specimen lacking pigmentation.

Head. Prostomium with five short appendages, including a pair of digitiform palps in ventral-most position, a pair of digitiform lateral antennae, similar in size and shape as palps, and a digitiform median antenna, shorter than lateral antennae and palps (Fig. 7C, F). A pair of tentacular cirri shorter than lateral antennae and palps, close to lateral antennae (Fig. 7F). A few rounded small papillae scattered around head appendages (Fig. 7F).

Tubercles. First chaetiger with eight dorsal macrotubercles; following chaetigers each with one transversal row of dorsal macrotubercles increasing to 10–12 tubercles per segment from chaetiger 5 (Fig. 7B–C). Macrotubercles spherical to club-shaped



Figure 7. Sphaerodoridium minutum, paratypes USNM 22873, SEM. A Whole specimen, side view
B Whole specimen, ventro-lateral view C Same, ventral view D Detail of stalked, dorsal macrotubercle
E Dorsal macrotubercle and papilla F Anterior end, frontal view, showing head appendages and tubercles of anterior chaetigers G Mid-body parapodium, side view H Mid-body parapodia, anterior view I Posterior end and pygidium, ventral view J Chaetae anterior chaetigers K Chaetae mid-body parapodium
L Detail of Mid-body chaeta. Abbreviations: al, acicular lobe; la, lateral antenna; ma, median antenna; mc, macrotubercle; mi, microtubercle; pa, palp; tc, tentacular cirrus; vc, ventral cirrus.

with a short and smooth stalk (Fig. 7D–E); all macrotubercles similar in shape and size. Additional spherical and sessile papillae in different sizes over dorsum, arranged in 2–3 irregular transversal rows per chaetiger; 20–30 papillae on each mid-body chaetiger (Fig. 7A–C). Ventral surface with spherical papillae in different sizes, arranged in 2–3 transversal rows in a zig-zag pattern, with about 20 per segment in mid-body; numbers decreasing towards posterior end (Fig. 7A, C).

Parapodia. Parapodia sub-conical, increasing in size towards chaetiger 3 (Fig. 7C), around 2 times longer than wide (Fig. 7G, H). Acicular lobe anterior to chaetae, digitiform to clavate, longer than parapodial papillae and projecting distally (Fig. 7C, G, H). Ventral cirri digitiform projecting 1/2 to 2/3 as long as acicular lobe on anterior and mid-body segments, almost as long as in posterior segments (Fig. 7C, G, H). Parapodia with three spherical to clavate papillae: one on anterio-dorsal surface, one on anterio-basal position (Fig. 7F–H), and one on the posterior surface (Fig. 7F–G).

Chaetae. All parapodia with 4–7 compound chaetae, arranged in a curved transverse row around acicular lobe (Fig. 7F–I). Serrated, long blades, 4–5 times longer than maximum width, with a curved tip (Fig. 7J–L), similar throughout.

Pygidium. Pygidium terminal, with one mid-ventral digitiform anal cirrus projecting beyond parapodia, and one pair of clavate anal cirri, at base on median cirrus (Fig. 7I).

Internal features. Specimens are all opaque after fixation and preservation and internal features not observable.

Reproductive features. Copulatory organs or eggs not seen in type specimens.

Variation. Most paratypes show small, digitiform anterior appendages but the anterior end is often retracted and these are not easily discernible. This species seems very homogenous regarding the number and arrangement of epithelial tubercles. The exact number of macrotubercles and papillae is, however, difficult to assess in the type material because some of these are detached or because when macrotubercles are inflated and the stalk allows some reorganization, the exact number of tubercles per segment is imprecise. Parapodial papillae in mid-body segments vary between three and four as in some specimens a second papillae can be observed at the base of the posterior parapodial surface (Fig. 7G). Sexual dimorphism not observed.

Remarks. The original description is detailed and accurate for most characters, including those more difficult to observe. In most type specimens the dorsal macrotubercles are lost or they fall off when being handled and consequently the total number of tubercles vary from the 10–12 macrotubercles in each transverse row as stated in the original description. Macrotubercles are described as being attached to the body by a "short neck" (Webster and Benedict 1887), here interpreted as a short stalk. This feature justifies this species to be considered as a *Sphaerodoridium* and must have been overlooked in later studies dealing with this species, explaining why it was placed in *Sphaerodoropsis* for a long time (Berkeley and Berkeley 1948, Imajima 1969, Hartmann-Schröder 1996, Imajima 2009, Moreira 2012). Dorsal cirri were described as present (Webster and Benedict 1887), most likely referring to dorsal macrotubercles closest to parapodia.

Sphaerodoridium minutum is most similar to *S. guerritai* Moreira & Parapar, 2015, described from the north eastern Atlantic, having as many dorsal macrotubercles, but

S. minutum lacks the characteristic papillae on the macrotubercle's stalk that S. guerritai possesses, and the latter species has long stalks compared to the short ones in S. minutum. Further, there are clear differences between the two species in number and composition of parapodial papillae. Sphaerodoridium minutum is also similar to Sphaerodoridium evgenovi Gagaev, 2015, Sphaerodoridium kolchaki Gagaev, 2015, and Sphaerodoridium kupetskii Gagaev, 2015, all recently described from the Arctic Ocean, as they all share a similar arrangement and number of macrotubercles (Gagaev 2015). The two latter species, S. kolchaki and S. kupetskii, have long macrotubercle stalks, hence they are different from S. minutum. The seemingly short macrotubercle stalks in S. evgenovi are rather similar to those in S. minutum. However, S. evgenovi has chaetae with somewhat shorter blades, lack parapodial papillae on the posterior surface, and also has parapodia much longer than wide compared to S. minutum (Gagaev 2015). The three species described by Gagaev (2015) and the one described by Moreira and Parapar (2015) are all similar to each other and show resemblance to S. minutum. These two papers were published the same year and the authors were most likely not aware of each other's work. These species should be compared in detail in order to assess the species delimitation (Gagaev 2015).

Sphaerodoridium minutum has similar number of stalked macrotubercles to *Clavodorum polypapillata* (Hartmann-Schröder & Rosenfeldt, 1988) described from Antarctica, and *C. andamanense* Bakken, 2002 described from Thailand. In *C. polypapillata* the original description overlooked stalked macrotubercles (Moreira and Parapar 2011), and has been re-described to have a larger number of macrotubercles (12–17 per transverse row, compared to 10–12 present in *S. minutum*) and a large number of ventral papillae (>40 per segment compared to ca. 20 in *S. minutum*). *Clavodorum polypapillata* further has longer stalks than in *S. minutum* (Moreira and Parapar 2011). *Clavodorum andamanense* lacks papillae on the dorsum, unlike *S. minutum*.

Sphaerodoridium minutum has been reported with a wide geographic distribution (Berkeley and Berkeley 1948, Lützen 1961, Imajima 1969, 2009, Hartmann-Schröder 1996). In order to clarify the geographic distribution of this species careful examination of specimens reported under this name from other geographic areas is needed.

The validity and clear delimitation of *Sphaerodoridium* and *Clavodorum* Hartman & Fauchald, 1971 has been regarded as doubtful. Several authors pointed out the insubstantial generic differences between the two genera (Bakken 2002, Moreira and Parapar 2011, 2015) and only recently reciprocal monophyly was not assessed in the first phylogenic hypothesis of the family (Capa et al. in 2016). There are currently 10 species of *Sphaerodoridium* and another 10 of *Clavodorum* described from world-wide areas and from shelf to abyssal depths (Capa et al. 2014, Moreira and Parapar 2015). As the delimitation of the two genera is not clear, and species representing the two genera are phylogenetically nested in a group (Capa et al. 2016), it will be necessary to assess all described species in both genera when comparing similarities.

Distribution. This species is known from the New England region of the US. It has been reported from the North Atlantic (e.g. Hartmann-Schröder 1996), North Pacific (e.g. Berkeley and Berkeley 1948) and Japan (Imajima 1969, 2009). The emer-

gence of descriptions of new species similar to *S. minutum* suggests a review of the true identity of reported specimens is needed.

Sphaerodoropsis Hartman & Fauchald, 1971

Sphaerodoropsis Hartman & Fauchald, 1971: 69; Fauchald 1974: 69.

Type species. *Sphaerodorum sphaerulifer* Moore, 1909.

Diagnosis. Body generally short and ovoid, some forms slender. Four or more longitudinal rows of macrotubercles, in one or several transverse rows per segment. Macrotubercles sessile and smooth, without terminal papillae. Microtubercles absent. Papillae over body surface and parapodia. Prostomial and peristomial appendages short, spherical or digitiform. Parapodia with compound chaetae; hooks absent.

Sphaerodoropsis corrugata Hartman & Fauchald, 1971

Figs 8, 9

Sphaerodoropsis Hartman & Fauchald, 1971: 69–71, pl 34, figs a, b. *Sphaerodoridium* sp. A. – Hartman 1965: 94, Pl. 14, fig. f.

Material examined. Holotype: LACM-AHF POLY 950, west of Atlantis Canyon, New England continental slope, North Atlantic, 39°56.5'N, 70°39.9'W, 400 m, 28 Aug 1962. Additional material. USNM 1002203 (1 ind.), off Massachusetts, United States, North Atlantic Ocean, 40°01.284'N, 70°55.032'W, 255 m, 8 Dec 1984; USNM 1002207 (6 ind., 3 for SEM) Lydonia Canyon, Georges Bank, United States, 40°21.114'N, 67°32.232'W, 590 m, 28 Apr 1985; USNM 1002209 (4 ind.) ff Massachusetts, United States, 40°01.248'N, 70°55.086'W, 250 m, 4 May 1985; USNM 1002193 (2 ind., 1 for SEM), Off Cape Hatteras, North Carolina, United States, 35°26.268'N, 74°41.436'W, 2003 m, 24 May 1985; USNM 1002212 (1 ind.) off Massachusetts, United States, 39°50.382'N, 70°01.65'W, 1239 m, 27 Nov 1985; USNM 1001989 (2 juvenile ind.) off New Jersey, United States, 38°40.068'N, -072°56.418'W, 1519 m, 13 Nov 1985; USNM 1001830 (1 ind.), Baltimore Canyon, United States, 37°53.286'N, 73°45.264'W, 1619 m, 7 Aug 1984; USNM 1002032 (1 ind.), off Massachusetts, 39°48.36'N, 70°54.93'W, 1249 m, coll. Battelle-New England Marine Research Lab For BLM/ MMS, 30 Nov 1985; USNM 1002033 (1 ind.), Georges Bank, 41°01.35'N, 66°20.23'W, 1345 m, coll. Battelle-New England Marine Lab For BLM/ MMS, 25 Jul 1986; USNM 1002035 (1 ind.), Georges Bank, 41°01.54'N, 66°20.11'W, 1345 m, coll. Battelle-New England Marine Lab For BLM/ MMS, 25 Jul 1986.

Comparative material. Sphaerephesia fauchaldi Kudenov, 1987b, holotype NMNH 102785; Sphaerephesia longisetis Fauchald, 1972, holotype AHF POLY

0964; Sphaerephesia regularis Böggemann, 2009, holotype ZMH P25498, paratypes ZMH P25497 (4 ind.), ZMH P25499 (4 ind.); Sphaerephesia similisetis Fauchald, 1972, paratype AHF POLY 0967 (2 ind.). Sphaerephesia hutchingsae Capa & Bakken, 2015, holotype East of Malabar, Sydney, New South Wales, Australia, AM W.42748, 33°58.71667'S, 70°39.9'E, 82 m, 22 Aug 1995; paratypes from nearby collecting sites (see Capa and Bakken 2015).

Diagnosis. Body ellipsoid, with four longitudinal rows of sessile, rounded to pear-shaped macrotubercles without terminal papillae, per segment. Distance between dorsal-most macrotubercles exceeds distance between those and lateral ones. Parapodia with ventral cirri as long as acicular lobe or slightly shorter and 4–6 rounded, small papillae, sometimes a dorsal one slightly larger. Parapodia with 10–16 compound chaetae, with thin shafts and blades 12–16 times as long as wide.

Re-description. *Measurements and general morphology.* Gravid female, 2 mm long, 0.5 mm wide, with 17 chaetigers. Body ellipsoid, slightly flattened dorso-ventrally (wider than high). Tegument with transverse wrinkles and segmentation only barely discernible. Preserved specimen lacking pigmentation.

Head. Anterior end bluntly rounded (Fig. 8A–F). Prostomium with seven longer appendages, including a pair of palps, in ventral-most position near the mouth, conical, wrinkled and about 4–5 times longer than wide at base; a pair of lateral antennae, similar in shape and size to palps; a median antenna, shorter (two thirds) than lateral antennae and with a rounded distal end; and a pair of antenniform papillae behind lateral antennae, similar in shape and size to median antenna, or smaller (Fig. 8C–F). Around 20 digitiform smaller papillae confined by prostomial appendages and mouth, in frontal view (Fig. 8F). One pair of tentacular cirri similar in shape and size to lateral antennae and palps.

Tubercles. First chaetiger with two macrotubercles; rest of chaetigers with four macrotubercles, each arranged in four longitudinal rows along dorsum (Fig. 8A–D, G). Distance between mid-rows larger than between these and lateral rows of macrotubercles (Fig. 8G). Size of all macrotubercles similar, with base as large as base of parapodia or smaller, or slightly increasing in size in first four chaetigers (Fig. 8A–B), also slightly reducing in size in posterior chaetigers towards pygidium. Macrotubercles spherical or pear shaped, with some pores (Fig. 8G–J). Spherical papillae present over dorsum and ventrum, with arrangement hard to determine in holotype.

Parapodia. Parapodia sub-conical, 1–2 times longer than wide (shorter in anterior and posterior most chaetigers), wrinkled (Fig. 9A–E). Acicular lobe projecting distally anterior to chaetae (Fig. 9A–E). Ventral cirri sub-conical, similar in length to acicular lobe but not projecting over the tip of acicular lobe (Fig. 9A). Mid-body parapodia with 4–5 spherical papillae: 1–2 on dorsal surface, 1 on anterior surface, 2 on ventral surface and 0–1 on posterior surface (Fig. 9A–F); all similar in size or a dorsal slightly larger.

Chaetae. Compound chaetae present in all chaetigers, arranged in a curved transverse row around acicular lobe and numbering 8–12 per fascicle in mid-body chaetigers (Fig. 9A–E). Blades similar in length along fascicles (12–16 times longer than maximum width), only slightly longer than those from mid-fascicle, with fine and short spinulation along superior edge, slightly curved (Fig. 9G).



Figure 8. *Sphaerodoropsis corrugata*, USNM 1002193 and 1002207. **A** Whole specimen, dorsal view **B** Juvenile, dorsal view **C**, **D** Anterior end, dorsal view **E** Same, side view **F** Head, frontal view **G** Midbody segments, dorsal view **H**, **I** Mid-body dorsal macrotubercles **J** Posterior segments and pygidium, dorsal view. Abbreviations: la, lateral antenna; ma, median antenna; pa, palp; tc, tentacular cirrus.



Figure 9. *Sphaerodoropsis corrugata*, USNM 1002193 and 1002207. **A** Parapodium first chaetiger, anterior view **B** Parapodia chaetiger 6 and 7, anterior view **C** Posterior parapodium, anterior view **D**, **E** Midbody parapodia, dorsal view **F** Midbody segments, side view **G** Midbody chaetae. Abbreviations: al, acicular lobe; vc, ventral cirrus.

Pygidium. Pygidium terminal, with mid-ventral digitiform anal cirrus and a pair of dorsal anal cirri, similar in shape to macrotubercles but slightly smaller.

Internal features. Eyes not observed in any specimen; holotype with a pair of reddish anterior spots that may not be eyes, but the nuchal organs. Muscular pharynx runs along chaetigers 3–6.

Reproductive features. Holotype and a few of the additional specimens examined are gravid females carrying large discoid eggs, 200 μ m in diameter that occupy most of the body coelom, from the anterior to the posterior segments. "Copulatory organs" not observed.

Variation. Specimens measured from 0.6 mm to 3.5 mm. Specimens assigned to this species show some variation in the number and arrangement of the epithelial papillae, probably because there are not easily ascertained in the holotype and were not described in detail in the original description. Larger specimens with approximately 30 papillae present between mid-macrotubercles and 10 between these and the lateral ones in mid-segments (Fig. 8G). Smaller specimens with less epithelial papillae (Fig. 8B). About five papillae between lateral macrotubercles and parapodia. Ventral surface with small papillae, arranged in about five transversal rows in mid-body. Body epithelium with ellipsoid granules (Fig. 8H). Chaetae numbering up to 16 per fascicle in mid-body chaetigers. Shaft with slightly widened distal end with delicate, almost inconspicuous spinulation (Fig. 9G), length of blades up to 15–16 times longer than wide.

Remarks. Sphaerodoropsis corrugata has not been reported since its original description. Records of this species include off New York state, from 400 to 1500 m (Hartman and Fauchald 1971). The type is not in excellent conditions to examine the number and arrangement of papilla. Nevertheless, some features observed on this specimen do not match the original description or are somehow imprecise. Antenniform papillae are present, unlike indicated in original and later descriptions by Hartman and Fauchald (1971) and Fauchald (1974). The parapodial papillae are all spherical or hemispherical, there is no truncate forms (Hartman and Fauchald 1971: Fig. 34B, Fauchald 1974) and if they were observed with that shape it could have been due to a collapse of the structure that has recovered its original shape after years of preservation (see Capa and Bakken 2015 for other examples). The chaetae drawn (Hartman 1965) may be ones with shorter blades in the fascicle, and some of the additional chaetae observed in the type and additional material are provided with longer and slender blades. As originally described, the acicular lobe is well developed and there is no additional parapodial lobes, only epithelial papillae, what seems to be the common pattern from all members in the family (Capa et al. 2014), and not characteristic of this particular species. The different terminology for papillae used by different authors creates confusion.

Sphaerodoropsis elegans Hartman & Fauchald, 1971 originally described from Brazil but also reported from New England, resembles *S. corrugata*. It also belongs to the *Sphaerodoropsis* Group 1 *sensu* Borowski (1994) and is provided with long blade chaetae. Review of the types of this species reveal that the macrotubercles are pear-shaped, almost possessing a terminal papilla, indicating they could formally be considered as *Sphaerephesia*. Other *Sphaerephesia* species with ellipsoid body, four rows of macrotubercles with a terminal rounded papilla, several additional papillae on dorsal surface, and falcigers with long blades are *Sphaerephesia similisetis* Fauchald, 1972, *Sphaerephesia longisetis* Fauchald, 1972, *Sphaerephesia chilensis* Fauchald, 1974, *Sphaerephesia fauchaldi* Kudenov, 1987b, and *Sphaerephesia hutchingsae* Capa & Bakken, 2015. *Sphaerodopsis corrugata* differs from all of these in the number of parapodial papillae since the other have one or two (*S. chilensis*) or more than seven (Fauchald 1972, 1974, Kudenov 1987b, Capa and Bakken 2015) while *S. corrugata* has four or five in mid-body chaetigers.

Distribution. New England, United States, 250–2000 m.

Key to sphaerodorids reported from shelf and continental slope environments in the northwest Atlantic between the Artic and the equator

The species reported from the northwestern Atlantic (considered herein as the continental shelf and slope areas off Atlantic Canada and New England) are marked with *.

1 Long-bodied individual, with two longitudinal rows of spherical macrotubercles (large tubercles) on dorsum with a terminal papilla, and two additional rows of microtubercles (small tubercles with a collar and a terminal papillae) Body ellipsoid with blunt anterior and posterior ends, dorsal tubercles in 2 All chaetae simple, robust, with hooked distal ends and rounded edgesSphaerodorum "flavum" Ørsted, 1843* 3 All parapodia, except in first or second anterior segment, bearing compound and simple chaetae, the latter with angled edges..... Ephesiopsis guayanae Hartman & Fauchald, 1971 * All chaetae, except in first or second anterior segment, compound or semicompound, often with lost blades4 4 Parapodia with 5-6 papillae each, all arranged at distal end, no erect papilla on dorsal surface of parapodia..... Parapodia with two papillae each, not at distal end, one erect papilla on dorsal surface and one on anterior surface of each parapodium Ephesiella mixta Hartman & Fauchald, 1971* Parapodia with about 11 papillae each, two on superior margin of parapodia, in addition to others (about nine) distributed randomly over the parapodia Ephesiella bipapillatum Kudenov, 1987 Dorsal macrotubercles, stalked, arranged in six or more longitudinal rows....6 5 Dorsal macrotubercles sessile, arranged in four or more longitudinal rows.....9 6 Lateral antennae without spurs at their bases, ventrum with 10 or more longitudinal rows of papillae in mid-body segments......7 Lateral antenna with spurs at their bases, ventrum with up to six longitudinal 7 Antennae over three times longer than wide, middle antennae often longer than lateral antennae. Ventrum with 10 longitudinal rows of papillae in middle segments, parapodia with 3-6 papillae Antennae short, up to three times longer than wide, middle antennae often shorter than lateral. Ventrum with up to 15 longitudinal rows of papillae in

	middle segments, parapodia with 2–3 papillae
8)	Ventrum with papillae arranged in two longitudinal rows, parapodia with a
	single papilla on the ventral surface
	Clavodorum atlanticum Hartman & Fauchald, 1971*
_	Ventrum with papillae arranged in six longitudinal rows, parapodia with four
	papillae Sphaerodoridium lutzeni Kudenov, 1987
9	Dorsal macrotubercles arranged in four longitudinal rows, and one transverse
	line per segment; chaetae compound10
_	Dorsal tubercles, considered papillae because of the smaller size compared
	to macrotubercles of other species, arranged in over 10 longitudinal rows,
	and approximately two transverse rows per segment; chaetae simple, widened
	subdistally <i>Euritmia carolensis</i> sp. n. *
10	Dorsal macrotubercles smooth, mainly spherical but some, especially in pos-
	terior segments can be pear-shaped11
_	Dorsal macrotubercles with a terminal papilla12
11	Parapodia with 4–6 rounded, small papillae, sometimes a dorsal one slightly
	larger, 10–16 compound chaetae with blades 12–16 times as long as wide
	<i>Sphaerodoropsis corrugata</i> Hartman & Fauchald, 1971*
_	Parapodia with about 20 papillae and up to 10 chaetae about eight times as
	long as wide
12	Parapodia with a single papilla near de base of the superior edge; chaetae are
	about eight times as long as wide
	Sphaerodoropsis elegans Hartman & Fauchald, 1971*
_	Parapodia with four papillae; 7–9 chaetae with blades 10–12 times as long as
	wide Sphaerodoropsis longipalpa Hartman & Fauchald, 1971*

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RESEARCH ARTICLE



Borniopsis mortoni sp. n. (Heterodonta, Galeommatoidea, Galeommatidae sensu lato), a new bivalve commensal with a synaptid sea cucumber from Japan

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Abstract

The Galeommatoidea is a bivalve superfamily that exhibits high species diversity in shallow waters. Many members of this superfamily are associated commensally with burrowing marine invertebrates in benthic sediments. The genus *Borniopsis* is known only from eastern Asia and exhibits high host diversity (e.g., mantis shrimps, crabs, holothurians, sipunculans and echiurans). A new species, *Borniopsis mortoni* **sp. n.**, is described from mud flats at the mouth of the Souzu River, southwestern Shikoku Island, Japan. This species has elongate-ovate shells covered by a tan to dark brown periostracum, and lives attached by both its foot and byssal threads to the body surface of the synaptid sea cucumber *Patinapta ooplax*. Several individuals of *B. mortoni* are often found on the same host, but sometimes more than 10 individuals can occur together. *Borniopsis mortoni* is one of the smallest species in this genus. Probably, its small body size is an adaptation to the mode of life in a narrow host burrow. Until now, only two other *Borniopsis* species were known to have commensal associations with synaptids. Thus, this is the third example of a synaptid-associated species from this genus. In addition, we briefly review the galeommatoideans commensal with apodid sea cucumbers.

Keywords

Apodida, Bivalvia, *Borniopsis, Byssobornia*, commensalism, Galeommatoidea, host shift, Holothuroidea, *Pseudopythina*, symbiosis, Synaptidae

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Introduction

The Galeommatoidea is a superfamily of small bivalves that exhibits tremendous diversity in the intertidal zone (Bouchet et al. 2002, Paulay 2003, Lützen and Nielsen 2005). Many members of this superfamily are commensals associated in highly specific relationships with benthic invertebrates that burrow in soft sediments (Boss 1965, Morton and Scott 1989, Li et al. 2012, Goto et al. 2012). Most commensal galeommatoideans live attached directly onto the host's body surface or the walls of its burrow (Boss 1965, Morton and Scott 1989, Goto et al. 2012).

The genus Borniopsis was established for B. tsurumaru Habe, 1959 as the type species with a second species, B. ariakensis Habe, 1959, from Japan (Habe 1959). Morton and Scott (1989) described four Pseudopythina (Borniopsis) species from Hong Kong. After this publication, Borniopsis was frequently synonymized with Pseudopythina (e.g., Lützen et al. 2004, Goto et al. 2012). However, Huber (2015) noticed that these East Asian Pseudopythina species are distinct from the type species P. macandrewi (P. Fischer, 1867), which occurs only in the eastern Atlantic Ocean, in dentition, seminal receptacles, and demibranchs and thus he transferred them to Borniopsis together with some species of Byssobornia, Squillaconcha and Kellia (Huber, 2015), although B. fujitaniana (Yokoyama, 1927) was recently reassigned to the genus *Tellimya* (Goto et al., 2016). As a result, at least nine species currently belong to this genus – B. tsurumaru, B. ariakensis, B. macrophthalmensis (Morton & Scott, 1989), B. maipoensis (Morton & Scott, 1989), B. nodosa (Morton & Scott, 1989), B. ochetostomae (Morton & Scott, 1989), B. subsinuata (Lischke, 1871), B. yamakawai (Yokoyama, 1922) and B. sagamiensis (Habe, 1961) (Huber 2015). All of these species are known only from eastern Asia. Those Borniopsis species, for which the biology is known, are host-specific commensals associated with burrowing invertebrates (mantis shrimps, crabs, holothurians, sipunculans, echiurans and probably tanaids) (Kuroda 1937, Morton 1972, 1988, Morton and Scott 1989, Goto and Kato 2012, Goto et al. 2012). Host animals are different among the Borniopsis species, except for one species pair (B. tsurumaru and B. ariakensis), suggesting that this group diversified by repeated host shifts among various invertebrates in eastern Asia. This assumption is partially confirmed by molecular phylogeny (Goto et al. 2012).

In this study, we describe a new species of *Borniopsis*, which was collected from the synaptid sea cucumber *Patinapta ooplax* (von Marenzeller, 1881) on mud flats at the mouth of Souzu River, southwestern Shikoku Island, Japan. *Patinapta ooplax* is a small earthworm-like holothurian that burrows in muddy sediments in the intertidal zone. We compared its morphology and host associations with the other *Borniopsis* species. In addition, we reviewed galeommatoideans associated with apodid sea cucumbers.

Materials and methods

An undescribed species of *Borniopsis* was found attached to the synaptid sea cucumber *Patinapta ooplax* in the mud flats at the mouth of the Souzu River, Ainan-cho, Ehime Prefecture, southwestern Shikoku Island, Japan (32°57'N, 132°33'E) on 20 May 2000. We collected the specimens of this bivalve species during spring low tides in 2000, 2001, 2003, 2006, 2007, 2009, 2012, and 2013. Seven specimens collected on 11 March 2012 and two specimens collected on 15 March 2013 were preserved in 100% ethanol and brought back to the laboratory and observed under a binocular dissecting microscope for description. One specimen collected on 12 April 2009 was bleached to remove the periostracum for observation of shell surface sculpture. We deposited the holotype and two paratypes in the Museum of Zoology, University of Michigan (UMMZ), and two paratypes in National Museum of Nature and Science, Tokyo (NSMT). We also observed specimens of *B. tsurumaru* (SBMNH 149526), *B. ariakensis* (SBMNH35056), *B. ochetostomae* (SBMNH 149525) and *B. maipoensis* (SBMNH 35126) loaned from the Santa Barbara Natural History Museum. All the loaned specimens were collected from the mud flats of Hong Kong.

Systematics

Superfamily Galeonmatoidea J.E. Gray, 1840 Family Galeonmatidae *sensu lato* (Ponder, 1998) Genus *Borniopsis* Habe, 1959

Borniopsis mortoni sp. n.

http://zoobank.org/1782E002-A7F1-42D1-A1EC-0824A89B5A3E New Japanese name: Himoikarinamako-yadorigai Figs 1–4

Material examined. Holotype (Figs 1, 2): UMMZ 305035 (SL 4.1 mm, SH 2.8 mm). Paratype 1 (Fig. 3): UMMZ 305036-1 (SL 3.9 mm, SH 2.7 mm), paratype 2: UMMZ 30536-2 (SL 2.4 mm, SH 1.8 mm), paratype 3: NSMT-Mo 78968 (SL 3.7 mm, SH 2.7 mm) and paratype 4 (Fig. 1B): NSMT-Mo 78969 (SL 3.3 mm, SH 2.3 mm). Non-type specimens: four individuals of *B. mortoni* (SL 2.4, 3.3, 3.8, 3.3 mm). All specimens were collected in the mud flats at the mouth of the Souzu River, Ainancho, Ehime Prefecture, southwestern Shikoku Island, Japan (32°57'N, 132°33'E). Comparative species (Fig. 4): *B. tsurumaru*, SBMNH 149526; *B. ariakensis*, SBMNH 35056; *B. ochetostomae*, SBMNH149525; and *B. maipoensis*, SBMNH 35126.

Type locality and habitat. Mud flats at the mouth of the Souzu River, Ainan-cho, Ehime, southwestern Shikoku Island, Japan (32°57'N, 132°33'E).

Description. Shell (Figs 1–3): Shell small (up to 4.1 mm), thin, slightly inflated, shape elongate-ovate and equivalve; inequilateral, longer anteriorly. Anterior and posterior margins rounded, ventral margins slightly rounded. Umbo small. Beak small, prosogyrate, situated 2/3 of way toward posterior. Each valve covered by tan to dark brown periostracum with black deposits, often eroded around beaks (Figs 2, 3). Shell surface underneath periostracum smooth and whitish with pearly luster (Fig. 1B).



Figure 1. *Borniopsis mortoni* and its host. **A** A crawling individual of *B. mortoni*. **B** Right shell valve of Paratype 4 (NSMT Mo 78969) bleached to remove the periostracum for observation of shell surface sculpture **C**, **D** *Patinapta ooplax* with multiple individuals of *B. mortoni* attached **E**, **F** *P. ooplax in situ* in mud flats with *B. mortoni* attached (arrowed). Photo credits: H. Ishikawa: **A–C, E, F**; Y. Hamamura: **D**. Scale bar: 1 mm.

Sculpture consisting of fine, dense and a few strong, widely spaced commarginal growth striae visible even on periostracum and very faint dense radial striae only visible underneath periostracum (Fig. 1B). Hinge of each valve consisting of a single stout cardinal tooth in front of umbo and well-developed oblique internal ligament posterior to umbo (Figs 2F, G; 3E, F). Soft parts (Figs 1, 2): Mantle not reflected,


Figure 2. Holotype of *Borniopsis mortoni* (UMMZ 305035) (SL 4.1 mm). **A, C, E** Right valve **B, D** Left valve **F, G** Hinge structure of left and right valves. Abbreviations: aa, anterior adductor muscle; ac, anterior cardinal tooth; f, foot; gn, gonad; il, internal ligament; lid, inner demibranch of left side; pa, posterior adductor muscle; rid, inner demibranch of right side; st, stomach. Scale bars 1 mm. Photo credits: R. Goto: **A–G**.

without prominent tentacles, edges narrowly extend beyond margin of shell, with numerous short papillae regularly arranged. Both anterior and posterior adductor muscles elongate-ovate, subequal and situated in the middle of dorsal and ventral margin. Ctenidia with gill axis nearly vertical, flat, consisting of single demibranch with both ascending and descending lamellae, joined anteriorly to inner and outer



Figure 3. Paratype 1 of *Borniopsis mortoni* (UMMZ 305035) (SL 2.4 mm). **A, C** Right valve **B, D** Left valve **E, F** Hinge structure of right and left valves. Abbreviations: ac, anterior cardinal tooth; il, internal ligament. Scale bars 0.5 mm. Photo credits: R. Goto: **A–F**.

labial palps. Labial palps leaf-shaped. Foot slender, of moderate size, with small heel; byssal glands located just in front of heel. Gonads situated from middle to posterior in visceral mass just below umbo. Stomach and digestive gland large, occupying anterior part of visceral mass.

Distribution. Only known from the type locality.

Host. *Patinapta ooplax* (Echinodermata: Holothuroidea: Apodida: Synaptidae) (Fig. 1C–F).

Host association. *Borniopsis mortoni* attaches to the body surface of *P. ooplax* by both its foot and byssal threads (Fig. 1C–F). Individuals were patchily distributed in the mud flats. Within the particular patches we sampled, approximately 70% of synaptids served as hosts for *B. mortoni*. Several *B. mortoni* often occured on one the same host. At the maximum, more than 10 individuals were attached to a single host. Two

P. ooplax infested by *B. mortoni* also harbored the endoparasitic eulimid gastropod *Hypermastus lacteus* (A. Adams, 1864).

Etymology. The specific name is dedicated to Dr. Brian Morton who has made great contributions to marine biology, marine ecology and malacology. He discovered many interesting commensal galeonmatoidean species from Hong Kong, some of which now belong to the genus *Borniopsis*.

Remarks. The genus *Borniopsis* has been variously assigned to the Kelliidae (Morton and Scott 1989), Lasaeidae (Bieler et al. 2010), and the subfamily Montacutinae within Galeonmatidae *sensu lato* (Huber 2015). However, these family- or subfamily-level groupings are ill-defined when a range of characters and taxa are considered (Ponder 1998). Indeed, the molecular analyses conducted by Goto et al. (2012) showed that each of these groups is actually polyphyletic. In this study, we assigned the genus *Borniopsis* tentatively to Galeonmatidae *sensu lato*, which Ponder (1998) defined by the same diagnosis that was applied to the superfamily Galeonmatoidea, as did Huber (2015). Further taxonomic assignment of this genus within Galeonmatoidea (or Galeonmatidae *sensu lato*) should be delayed until its family-level (or subfamily-level) classification is revised.

As with *B. mortoni*, both *B. tsurumaru* and *B. ariakensis* have a symbiotic relationship with synaptid sea cucumbers (Morton 1988, Morton and Scott 1989, Lützen et al. 2004, Kai and Henmi 2008). However, the particular host species differ between them – *Patinapta ooplax* (host solely for *B. mortoni*) and *Protankyra bidentata* (Woodward & Barrett, 1858) (host for both *B. tsurumaru* and *B. ariakensis*) (Morton and Scott 1989, Lützen et al. 2004). *Borniopsis mortoni* always attaches directly onto the body surface of the host (this study), whereas *B. tsurumaru* can attach to the body surface of the host, or the wall of the host's burrow, or to the carapace of commensal crabs living in the same burrows (Morton 1988, Morton and Scott 1989, Lützen et al. 2004, Kai and Henmi 2008, Goto et al. 2012). Furthermore, the number of bivalves per host is much higher in *B. mortoni* (several to more than 10) than *B. tsurumaru* and *B. ariakensis* (usually one) (Lützen et al. 2004, Goto, Ishikawa, and Hamamura, personal observations).

The shells of *B. tsurumaru* and *B. ariakensis* are much larger (up to 11–12 mm in SL) than those of *B. mortoni* (up to 4.1 mm) (Morton and Scott 1989) (Fig. 4A, B). Probably, this corresponds with the size of the host because *P. bidentata* is much larger than *P. ooplax*. The shells of *B. tsurumaru* and *B. ariakensis* are thicker and more inflated than those of *B. mortoni* (Lützen et al. 2004; this study). In addition, the shells of *B. mortoni* are always covered by a dark brown periostracum, whereas those of *B. tsurumaru* and *B. ariakensis* are often whitish, although some are dark brown. The umbones of *B. tsurumaru* and *B. ariakensis* are more protruding than those of *B. mortoni* (Fig. 4A, B). A molecular analysis is needed to understand whether these three synaptid-associated species are monophyletic or not. In addition, morphological variation of *B. tsurumaru* and *B. ariakensis* is apparently continuous (Goto, Ishikawa and Hamamura, pers. obs.) so molecular testing should be employed to investigate whether they can be distinguished genetically or not.



Figure 4. A Borniopsis tsurumaru (SBMNH 149526) B B. ariakensis (SBMNH 35056) C B. ochetostomae (SBMNH 149525) and D B. maipoensis (SBMNH 35126). Photo credits: R. Goto: A–C; P. Valentich-Scott: D. Scale bars 1 mm.

The present new species also closely resembles *B. ochetostomae* and *B. maipoensis* in having an elongate ovate shell covered by a brownish periostracum (Fig. 4C, D). However, *B. ochetostomae* is much larger (up to 11 mm) than *B. mortoni* and its beak is located more centrally than that of *B. mortoni* (Morton and Scott 1989, Jespersen et al. 2002, this study) (Fig. 4C). On the other hand, *B. maipoensis* is rather smaller (up to 3 mm) and more rounded than *B. mortoni* (Morton and Scott 1989, this study) (Fig. 4D). In addition, *B. maipoensis* has two distinct papillae on the dorsal surface of the foot (Morton and Scott 1989), whereas we did not observe such papillae on *B. mortoni* (holothurian hosts), *B. ochetostomae* (echiuran hosts) and *B. maipoensis* (probably tanaid hosts) (Morton and Scott 1989, this study).

Key to Borniopsis

1	Beaks subcentral	2
_	Beaks near posterior end	3
2	Shell outline elliptical to subelliptical	4

5	Shell outline trigonal to subtrigonal	_
nner surface of anterior valve	Shell outline subovate, without nodules on i	3
6	margin	
surface of anterior valve mar- \mathbf{B} and \mathbf{A}	Shell outline quadrate, with nodules on inner	-
B. noaosa	g_{11}	,
B. ochetostomae	SH/SL 0.60–0.66 (Morton and Scott 1989)	4
B. yamakawai	SH/SL 0.73 (this study)	—
B. macrophthalmensis	Periostracum brownish, max. SL < 4 mm	5
7	Periostracum whitish, max. SL > 4 mm	_
8	Umbo slightly protruding	6
B. mortoni	Umbo not protruding, max. SL < 5 mm	_
B. subsinuata	SH/SL 0.80 (Morton, 1972)	7
B. sagamiensis	SH/SL 0.59–0.64 (Habe, 1961)	_
c of foot, max. SL < 3 mm	With two distinctive papillae on dorsal surface	8
B. maipoensis		
ce of foot, max. SL > $3 \text{ mm} \dots 9$	Without two distinctive papillae on dorsal surface	_
B. tsurumaru	SH/SL 0.73 (this study)	9
B. ariakensis	SH/SL 0.66 (this study)	_

Discussion

Apodid sea cucumbers, including members of the families Synaptidae and Chiridotidae, are one of the major hosts for galeommatoideans (Boss 1965, Morton and Scott 1989, Kato 1998, Middelfart and Craig 2004) (Table 1). Including *B. mortoni*, at least 13 species are known to have commensal associations with apodid holothurians (Table 1). They can be separated into the following four groups: the first group includes *Anisodevonia*, *Austrodevonia*, *Devonia* and *Entovalva*, which have reduced shells covered by well-developed mantle lobes (Kawahara 1942, Kato 1998, Middelfart and Craig 2004); the second group includes *Borniopsis* covered in this study (Morton 1988, Morton and Scott 1989); the third groups includes *Montacuta* (Bateson 1923, Fox 1979); and the forth group includes *Scintillona* (Morton 1957, Ó Foighil and Gibson 1984). All of these four groups live attached to the apodid's body surface, except for *Entovalva*, which lives inside the host's oesophagus (Spärk 1931, Kato 1998). Molecular phylogenies suggested that associations with apodids have evolved repeatedly in the Galeommatoidea (Goto et al. 2012).

Borniopsis mortoni lives commensally with the apodid sea cucumber *P. ooplax* on the temperate coast of western Shikoku Island. On the other hand, another galeommatoidean species, *Anisodevonia ohshimai*, lives attached to *P. ooplax* on the subtropical coast of the Ryukyu Islands, southwestern Japan (Kawahara 1942, Kato 1998). Extensive sampling for *A. ohshimai* has been undertaken in the Ryukyu Islands (Kawahara 1942, Kato 1998, Kosuge 2001). However, *B. mortoni* has never been found there. Thus, *B. mortoni* probably does not occur on the subtropical coast of the Ryukyu Islands and may be restricted to more temperate coasts. An alternative hypothesis is that *P. ooplax* used by

Species	Host	Distribution	References
Anisodevonia ohshimai (Kawahara, 1942)	Patinapta ooplax	NW Pacific	Kawahara 1942; Kato 1998
Austrodevonia sharnae Middelfart & Craig, 2004	Taeniogyrus australianus	NW Pacific	Middelfart and Craig 2004
Devonia perrieri (Malard, 1903)	Leptosynapta inhaerens	NE Atlantic	Malard 1903
Devonia semperi (Oshima, 1930)	Protankyra bidentata	NW Pacific	Oshima 1930
Devonia sp.	Protankyra similis	NW Pacific	Semper 1868
Entovalva amboinensis (Spärck, 1931)	Patinapta laevis	NW Pacific	Spärck 1931
Borniopsis tsurumaru Habe, 1959	Protankyra bidentata	NW Pacific	Morton and Scott 1989
Borniopsis ariakensis Habe, 1959	Protankyra bidentata	NW Pacific	Morton and Scott 1989
Borniopsis mortoni sp. n.	Patinapta ooplax	NW Pacific	this study
Montacuta donacina (Wood, 1848)	Leptosynapta inhaerens	NE Atlantic	Bateson 1923
Montacuta percompressa Dall, 1899	Leptosynapta tenuis	NE Atlantic	Fox 1979
Scintillona bellerophon Ó Foighil & Gibson, 1984	Leptosynapta clarki	NE Pacific	Ó Foighil and Gibson 1984
Scintillona zelandica (Odhner 1924)	Taeniogyrus dendyi	New Zealand	Finlay 1927

Table 1. Galeommatoidean bivalves commensal with apodid sea cucumbers.

A. ohshimai in the Ryukyu Islands is a different species to that used by *B. mortoni* in eastern Shikoku Island. The taxonomy of this group of synaptid holothurians remains poorly understood and thus it is highly probable that *P. ooplax* is a species complex.

A new species of the genus Borniopsis is described herein. This genus is restricted to eastern Asia, and each species apparently utilizes a different invertebrate host (Table 2), suggesting that its diversification was caused by repeated host shifting. The shell size varies among species (Table 2), which is probably a specialization to each particular commensal lifestyle. Borniopsis macrophthalmensis, B. maipoensis and B. mortoni are the three smallest species in this genus (Table 2). Borniopsis macrophthalmensis is attached to the body surface of highly-mobile intertidal crabs only by fine byssal threads (Morton and Scott 1989). If B. macrophthalmensis had a large, heavy shell, it would easily fall off the host crab when it scurries quickly on the surface of the mudflat. Thus, such a small shell is probably an adaptation for life on a highly-mobile crab. Similarly, another galeomatoidean species, Arthritica japonica Lützen & Takahashi, 2003, which is also attached to the body surface of crabs, has very small shells (up to 2.05 mm) (Lützen and Takahashi 2003). This may represent a morphological convergence to a similar commensal lifestyle and supports our hypothesis of reduction in shell size associated with fast-moving hosts mentioned above. On the other hand, the second species B. maipoensis lives commensally with the tanaid Discapseudes sp. (Morton and Scott 1989), whereas the third species B. mortoni lives with the synaptid sea cucumber P. ooplax. The diameter of these host burrows is very small. Thus, the small-sized shells of B. maipoensis and B. mortoni are probably an adaptation to the mode of life in narrow host burrows.

The mudflats of eastern Asia evidently possess one of the richest burrowing invertebrate faunas in the world. However, burrow associates remain poorly understood in many of them. Thus, further investigation in this area could reveal increased diversity of this distinctive bivalve genus *Borniopsis*.

Species	Host	Max. SL (mm)	References
Borniopsis ariakensis Habe, 1959	holothurian (<i>Protankyra bidentata</i>)	12.3	Habe 1959
Borniopsis macrophtalmensis (Morton & Scott 1989)	crab (<i>Macrophthalmus</i>)	3.2	Morton and Scott 1989
Borniopsis maipoensis (Morton & Scott 1989) probably tanaid (Discapseudes sp.)		3.0	Morton and Scott 1989
Borniopsis mortoni sp. n.	<i>prniopsis mortoni</i> sp. n. holothurian (<i>Patinapta ooplax</i>)		this study
Borniopsis nodosa (Morton & Scott 1989)	sipunculan (<i>Sipunculus nudus</i>)	6.1	Morton and Scott 1989
Borniopsis ochetostomae (Morton & Scott 1989)	echiuran (<i>Listriolobus sorbillans</i>)	10.1	Morton and Scott 1989
Borniopsis sagamiensis (Habe 1961)	unknown	19.4	Habe 1961
Borniopsis subsinuata (Lischke, 1871)	mantis shrimp (<i>Squilla, Oratosquilla</i>)	12.0	Morton 1972
Borniopsis tsurumaru Habe, 1959	holothurian (<i>Protankyra bidentata</i>)	10.7	Habe 1959
Borniopsis yamakawai (Yokoyama, 1922)	echiuran (Ochetostoma erythrogrammon)	11.0	Goto and Kato 2012; this study

Table 2. Borniopsis, its host and maximum size (shell length).

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RESEARCH ARTICLE



The complete mitochondrial genome of the Chinese Daphnia pulex (Cladocera, Daphniidae)

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Abstract

Daphnia pulex has played an important role in fresh-water ecosystems. In this study, the complete mitochondrial genome of *Daphnia pulex* from Chaohu, China was sequenced for the first time. It was accomplished using long-PCR methods and a primer-walking sequencing strategy with genus-specific primers. The mitogenome was found to be 15,306 bp in length. It contained 13 protein-coding genes, two rRNA genes, 22 tRNA genes and a typical control region. This research revealed an overall A+T content of 64.50%. All of the 22 typical animal tRNA genes had a classical clover-leaf structure except for *trnS1*, in which its DHU arm simply formed a loop. The lengths of small and large rRNA were 744 bp and 1,313 bp, respectively. The A+T-rich region was 723 bp in length, which is longer than that from the North American species (689 bp). In terms of structure and composition, many similarities were found between the Chinese and North American *Daphnia pulex*.

Keywords

Daphnia pulex, gene order, mitochondrial genome, secondary structure

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Introduction

Cladocerans ("water fleas") are an important component of the microcrustacean zooplankton. Their habitats are mostly continental fresh and saline waters (Forró et al. 2008). *Daphnia pulex* has become a well-known model species for studying evolutionary biology, environmental biology and ecology (Miner et al. 2013, Geng et al. 2016). Although other related research has been done (Roland et al. 2011, Geng et al. 2014), there are still some difficulties with species identification. In this study, meaningful data to assist in the taxonomy of different species of *Daphnia* is provided, and variations in similar morphological groups using molecular tools are analysed (Petrusek et al. 2012).

The sequence and structure of mitochondrial genomes has been frequently used to study phylogenetic relationships of animal taxa. More specifically, the unusual characters of mitochondrial genome DNA, for instance its small size, fast evolutionary rate, simple structure, maternal inheritance and high informational content, have been widely regarded as a molecular marker for phylogenetic analysis (Wilson et al. 2000, Chao et al. 2014, Ma et al. 2015).

All metazoan animals contain their own circular mitochondrial genome with two strands (a J-strand and an N-strand) (Simon et al. 2014), which range from 14 kb to 42 kb in length (Wolstenholme 1992). These typically encoded 37 genes, namely: 2 rRNA genes (*16S rRNA* and *12S rRNA*), 22 tRNA genes, and 13 proteincoding genes (*COI, COII, COIII, Cytb, ATP6, ATP8, ND1, ND2, ND3, ND4, ND4L, ND5, ND6*) (Boore 1999). Moreover, the non-coding region (also called the control region or D-loop), which with significant functions in the regulation and initiation of mitochondrial DNA transcription and replication (Brown et al. 1979, Shadel and Clayton 1993, Zhang and Hewitt 1997). Complete mitochondrial genome sequences are more informative than shorter sequences of individual genes but also provide a set of genomic characters. This led to the recognition of relative positions of different genes, RNA secondary structures and modes of control of replication and transcription (Masta and Boore 2008). However, the complete mitochondrial genome sequences data on *Daphnia* released in Genbank is far from enough.

The main purpose of this study was to disclose the complete mitochondrial genome sequence of the Chinese *Daphnia pulex* for the first time, and to compare its features with other available cladoceran mitochondrial genomes.

This study also served as a useful source of information for both nuclear and mitochondrial markers in comparative analyses of the evolution of mitochondrial genomes in Cladocerans.

Materials and methods

Samples and DNA extraction

Total DNA was extracted from individual specimens using a TIANamp Micro DNA Kit (TIANGEN BIOTECH (BEIJING) CO., LTD) following manufacturer protocols. DNA samples were stored at -20 °C until further use.

PCR amplifications and sequencing

The *Daphnia pulex* mitochondrial genome was amplified using five pairs of primers (Table 1). To obtain the complete sequences of Chinese *Daphnia pulex*, short-PCR and long-PCR methods were used. The primers employed in this study were designed based on the mitochondrial genomes of the North American *Daphnia pulex* (Gen-Bank accession number AF117817) (Crease 1999) by using an NCBI primer-BLAST (http://www.ncbi.nlm.nih.gov/tools/primer-blast/).

The PCRs were performed by using an Eppendorf Thermal Cycler (5331AH760577, Eppendorf, Germany) with a 25 μ L volume reaction mixture containing 2.5 μ L 10×LA-Taq Buffer II(Mg²⁺ plus), 4 μ L dNTP Mixture (2.5 mM), 2 μ L DMSO, 1 μ L genomic DNA, 1 μ L 10 μ M of each primer, 0.5 μ L MgCl₂ (25 mM) and 0.25 μ L 2.5 units of LA Taq polymerase (TaKaRa Biomedical, Japan), and 12.25 μ L distilled water.

The reaction conditions were one cycle of denaturation at 95 °C 5 min, 35 cycles of denaturation at 95 °C 30 s, annealing at 50 °C 30 s, extension at 72 °C for 2 to 8 min and a final extension at 72 °C for 10 min. Each amplicon (5 μ L) was examined with agarose gel electrophoresis to validate amplification efficiency. PCR products were sequenced directly by primer walking from both directions after purification.

Primer pair	Size (bp)	Primer sequence(5'-3')
F1		AGAAGGGAATTTGAGCTCTTTTWGT
R1	5450	TTACCCTAGGGATAACAGCGTAA
F2		TCGTCTCGTCATTCATACCAGC
R2	2221	GTGCCAGCAGYYGCGGTTANAC
F3		ATAAYAGGGTATCTAATCCTRGT
R3	3122	ACTTCCWGATTGTCCYAAYTC
F4		ACTACCCGCAAACGATCTGG
R4	4000	TGGGATGGGTTGGGGCTAAT
F5		AGCCCCAAAAATTGGATTTCCC
R5	750	TGGCTTCGGCAACGGATAG

Table 1. Details of the primers used to amplify the mitogenome of Chinese Daphnia pulex.

Analysis and annotation

The raw sequences of mitochondrial genome were edited and assembled by using the program Seqman (DNAStar, Inc.) and then adjusting them manually. Protein-coding genes and rRNA genes were identified by the MITOS WebServer (http://mitos.bioinf. uni-leipzig.de/index.py) and the similarity between *Daphnia pulex* and that published in NCBI database were distinguished by BLAST search function (http://www.ncbi. nlm.nih.gov/BLAST/). Nucleotide sequences of PCGs were translated using the invertebrate mitochondrial genetic code. The tRNA genes were initially identified by the MITOS WebServer (http://mitos.bioinf.uni-leipzig.de/index.py) and their secondary structures were predicted and modified based on other metazoan's secondary structure of tRNA genes.

The exact initiation and termination codons were identified by using Clustal X version 2.0 (Larkin et al. 2007) and relied on reference sequences from other invertebrates. Nucleotide composition and codon usage were calculated with MEGA 6.0 software (Tamura et al. 2013). The sequence data has been deposited into GenBank database under the accession number KT003819.

Results and discussion

Genome organization and base composition

The mitochondrial genomes of the Chinese *Daphnia pulex* used in this study were similar to that of the *Daphnia pulex* in North America (Crease 1999). The complete mitochondrial genome of Chinese *Daphnia pulex* was a circular molecule 15,306 bp in size, containing 13 protein-coding genes, 22 tRNA genes, 2 rRNA genes for both the small and large subunits (*rrnS* and *rrnL*) and a putative control region (Fig. 1). Among all the 37 genes, 23 genes were encoded on the J-strand. The remaining genes were encoded on the N-strand. 8 overlaps were found between adjacent genes (29 bp in total), among which the longest was 10 bp located at *trnS2* and *ND1*. This included 15 intergenic spacers that ranged from 1 to 31 bp (84 bp in total), of which only one spacer was longer than 10 bp. That occurred between *ND4L* and *trnT*.

The mitochondrial genome of the Chinese *Daphnia pulex* has an A+T content of 64.50%, which is a little higher than that of the North American species (62.26%). Furthermore, it was determined that the AT skew was 0.006, and the GC skew was -0.107. AT skew and GC skew for a given strand were calculated as (G-C)/(G+C) and (A-T)/(A+T), respectively, with negative values in skewness meaning the coding strand is enriched for T or C. In contrast, positive values infer more As and Gs. On the whole, AT skew was slightly negative, or positive in the third codon position of vestimetiferans, and GC skew was more negative than AT skew (Table 2). Nucleotide bias can also be reflected by codon usage. We found that the RSCU (Relative Synonymous Codon Usage) value of NNA and NNU codons were greater than 1,



Figure 1. Structure of Chinese *Daphnia pulex* mitochondrial genome. *COI, COII, COIII*, refer to the cytochrome oxidase subunits, *Cytb* refers to cytochrome b, *ND1 - ND6* refer to NADH dehydrogenase components, and *rrL* and *rrnS* refer to rRNAs. tRNA genes are denoted by one letter symbol according to the IPUC-IUB single-letter amino acid codes. L1, L2, S1 and S2 denote tRNALeu(CUN), tRNALeu(UUR), tRNASer(AGN) and tRNASer(UCN), respectively. D-loop indicates A+T-rich region. Gene names outside the ring are coded on the majority strand while those inside are on the minority strand.

which indicates that codons were biased in favor of codons with A or T in the third position (Table 3).

Amino acids are denoted as one-letter symbol according to the IUPAC-IUB single letter amino acid codes.

Protein-coding genes

The complete mitochondrial DNA of Chinese *Daphnia pulex* from Chaohu had 13 protein-coding genes. Nine of these genes were located on the J-strand while the others

	areas	length (bp)	A(%)	T(%)	G(%)	C(%)	A+T(%)	G+C(%)	AT skew	GC skew
The whole	Ch	15306	32.45	32.04	15.84	19.66	64.49	35.50	0.006	-0.107
genome	Na	15333	31.47	30.79	16.69	21.05	62.26	37.74	0.011	-0.116
Protein-coding	Ch	11026	24.73	38.64	18.31	18.32	63.37	36.63	-0.219	-0.0002
genes	Na	11074	23.39	37.04	19.40	20.17	60.43	39.57	-0.226	-0.019
1.	Ch	3665	26.63	29.66	25.21	18.50	56.29	43.71	-0.054	0.154
lst	Na	3681	25.94	29.34	25.42	19.29	55.28	44.71	-0.061	0.137
2.1	Ch	3665	17.11	45.70	16.92	20.27	62.81	37.19	-0.455	-0.090
2nd	Na	3681	17.33	45.18	16.54	20.95	62.51	37.49	-0.445	-0.117
2.1	Ch	3665	30.23	40.52	12.91	16.34	70.75	29.25	-0.145	-0.118
3rd	Na	3681	26.68	36.57	16.30	20.46	63.25	36.76	-0.156	-0.113
DNIA	Ch	1448	33.22	33.01	18.99	14.78	66.23	33.77	0.003	0.124
tKINA	Na	1452	32.78	32.99	19.42	14.81	65.77	34.23	-0.003	0.134
	Ch	2057	34.03	34.71	16.67	14.58	68.74	31.25	-0.010	0.067
rKNA	Na	2067	35.41	32.41	15.19	16.98	67.82	32.17	0.044	-0.056
D-loop	Ch	723	32.09	33.33	16.04	18.53	65.42	34.57	-0.019	-0.072
D-100p	Na	689	32.37	34.69	15.38	17.56	67.06	32.94	-0.035	-0.066

Table 2. Nucleptide composition in different regions of the *Daphnia pulex* mitochondrial from different areas.

AT skew = (A-T)/(A+T), GC skew = (G-C)/(G+C).

Table 3. Codon usage of the Chinese Daphnia pulex mitogenome.

Codon	Count	RSCU									
UUU(F)	20.1	1.39	UCU(S)	8	2.04	UAU(Y)	7.9	1.27	UGU(C)	3.5	1.3
UUC(F)	8.8	0.61	UCC(S)	3	0.76	UAC(Y)	4.5	0.73	UGC(C)	1.9	0.7
UUA(L)	12.3	1.76	UCA(S)	3.7	0.94	UAA(*)	5.9	1.01	UGA(*)	4.5	0.77
UUG(L)	6.3	0.9	UCG(S)	1.8	0.45	UAG(*)	7.2	1.22	UGG(W)	3.8	1
CUU(L)	9.2	1.32	CCU(P)	5	1.71	CAU(H)	3.3	1.23	CGU(R)	1.2	0.63
CUC(L)	5.2	0.74	CCC(P)	3.2	1.08	CAC(H)	2.1	0.77	CGC(R)	1.1	0.59
CUA(L)	5.3	0.76	CCA(P)	1.5	0.5	CAA(Q)	3.5	1.08	CGA(R)	1.9	1.05
CUG(L)	3.6	0.52	CCG(P)	2.1	0.71	CAG(Q)	2.9	0.92	CGG(R)	1	0.55
AUU(I)	12.2	1.57	ACU(T)	6	1.88	AAU(N)	5.6	1.4	AGU(S)	4.6	1.18
AUC(I)	5	0.64	ACC(T)	2.4	0.75	AAC(N)	2.4	0.6	AGC(S)	2.5	0.63
AUA(I)	6.2	0.79	ACA(T)	2.9	0.92	AAA(K)	4.4	1.1	AGA(R)	3.8	2.06
AUG(M)	5	1	ACG(T)	1.5	0.46	AAG(K)	3.6	0.9	AGG(R)	2.1	1.13
GUU(V)	5.2	1.41	GCU(A)	4	1.81	GAU(D)	4.3	1.23	GGU(G)	2.4	0.62
GUC(V)	2.3	0.62	GCC(A)	1.8	0.83	GAC(D)	2.7	0.77	GGC(G)	2.1	0.54
GUA(V)	4.6	1.24	GCA(A)	2.2	0.97	GAA(E)	1.8	0.69	GGA(G)	4.4	1.13
GUG(V)	2.7	0.73	GCG(A)	0.8	0.38	GAG(E)	3.4	1.31	GGG(G)	6.6	1.71

RSCU: Relative Synonymous Codon Usage.



Figure 2. Nucleotide compositions of the two *Daphnia pulex* from Chinese Chaohu (Ch) and North America (Na). CDS: protein-coding genes; 1st: first codon position; 2nd: second codon position; 3rd: third codon position; tRNA: tRNA genes; rRNA: rRNA genes; D-loop: A+T-rich region. In addition, stop codons were excluded.

were found on the N-strand; the same as the *Daphnia pulex* in North America (Table 3). Ten out of these 13 protein-coding genes initiated with typical ATN codons. *ND2*, *COII*, *ATP6*, *COIII*, *ND4*, *Cytb* and *ND1* started with ATG, *COI* initiated with ATA, and moreover *ND3* and *ND6* used ATC as the initiating codon. The *ATP8* and *ND5* genes used GTG. The *ND4L* gene used none of these as initiating codon, but GCT.

As is the case with some other arthropod species, the initiation functions of the *COI*codon has not been fully investigated. Atypical initiating codons for the *COI* gene in mitochondrial genomes have been reported in many studies, examples of these genes are: CGA (Gong et al. 2012), GTG (He et al. 2011), TTG (Hu et al. 2010, Li et al. 2012), ACG (Wilson et al. 2000), CCG (Fenn et al. 2007), ACC (Yamauchi et al. 2004), and TTA (Yamauchi et al. 2002). In *Drosophila, Locusta* and *Daphnia*, there are occasionally some uncommon quadruplets like, ATAA or ATTA, that may serve as an initiation codon (Wilson et al. 2000). One example of this is the *COI* gene in the North American *Daphnia pulex* initiating with ATTA (Crease 1999). However, the *COI* gene of Chinese *Daphnia pulex* started with classical ATA.

Nine of the 13 protein-coding genes used the typical termination codon TAN. *ND2* and *ATP8* terminated with TAG. *COIII*, *ND3*, *Cytb*, *ATP6*, *ND4L*, *ND6* and *ND1* all terminated with TAA. *COI*, *COII*, *ND4* and *ND5* used the incomplete termination codon T. Both of the complete termination codons TAG and TAA and two additional abbreviated termination codons T and TA were found in the North American Daphnia pulex (Table 4).

The use of incomplete termination codons on these genes might serve the purpose of avoiding overlapping nucleotides between adjacent genes (He et al. 2012). The incomplete termination codons would become functional termination codons after polycistronic transcript cleavage and polyadenylation processes have occured (Ojala et

Constant	posi	length		Start/stop codon		
Gene/strand	Ch	Na	Ch	Na	Start codon (Ch/Na)	Stop codon (Ch/Na)
trnI/J	1–64	1–64	64	64		
trnQ/N	66–133	66–133	68	68		
trnM/J	134–197	134–197	64	64		
ND2/J	198–1139	198–1185	942	988	ATG/ATG	TAG/T
trnW/J	1138–1202	1186–1251	65	66		
trnC/N	1206–1268	1253–1316	63	64		
trnY/N	1278–1340	1328–1391	63	64		
COI/J	1350-2886	1397–2934	1537	1538	ATA/(A)TTA	T_/T_
trnL2/J	2887–2954	2935-3002	68	68		
COII/J	2956-3634	3004–3682	679	679	ATG/ATG	T_/T_
trnK/J	3635-3704	3683–3752	70	70		
trnD/J	3709–3773	3757–3821	65	65		
ATP8/J	3774–3935	3821-3982	162	162	GTG/GTG	TAG/TAG
ATP6/J	3929-4603	3976-4649	675	674	ATG/ATG	TAA/TA_
COIII/J	4603-5391	4650–5438	786	789	ATG/ATG	TAA/TAA
trnG/J	5393–5456	5439–5499	64	61		
ND3/J	5457-5810	5500–5852	354	353	ATC/ATT	TAA/TA_
trnA/J	5811-5874	5853-5918	64	66		
trnR/J	5876-5940	5920-5984	65	65		
trnN/J	5943-6010	5985–6051	68	67		
trnS1/J	6011–6075	6052–6116	65	65		
trnE/J	6076–6141	6117–6184	66	68		
trnF/N	6141–6205	6184–6249	65	66		
ND5/N	6207–7913	6250–7957	1707	1708	GTG/ATG	T_/T_
trnH/N	7908–7971	7952-8015	64	64		
ND4/N	7972–9292	8016–9336	1321	1321	ATG/ATG	T_/T_
ND4L/N	9295–9570	9339–9614	276	276	GCT/ATT	TAA/TAA
trnT/J	9602–9664	9646–9710	63	65		
trnP/N	9665–9730	9711–9775	66	65		
ND6/J	9733-10245	9778-10290	513	513	ATC/ATT	TAA/TAA
Cytb/J	10245-11378	10298-11431	1134	1134	ATG/ATG	TAA/TAA
trnS2/J	11379–11447	11432-11500	69	69		
ND1/N	11438–12373	11494–12426	936	936	ATG/ATG	TAA/TAA
trnL1/N	12377-12443	12430-12496	67	67		
rrnL/N	12454-13766	12506-13819	1313	1314		
trnV/N	13769–13840	13821-13892	72	72		
rrnS/N	13840-14583	13892–14644	744	753		
D-loop/J	14584–15306	14645–15333	723	689		

Table 4. Organization of the mitochondrial genomes of *Daphnia pulex* from Chinese Chaohu (Ch) and that from North America (Na).

Note: J and N refer to the majority and minority strand, respectively. Position numbers refer to positions on the majority strand.



Figure 3. Inferred secondary structure of 22 tRNA genes in Chinese Daphnia pulex mtDNA genome.

al. 1981). These incomplete codons and this mechanism has been commonly found in metazoan mitochondrial genomes (Wei et al. 2009, Liao et al. 2010). The total length of the 13 protein-coding genes was found to be 11,026 bp for the Chinese *Daphnia pulex*, which accounts for 63.37% of the total mitogenome length.

Many composition similarities were noted between the two different species compared in this study (Fig. 2).

tRNA genes

All of the 22 typical arthropod tRNAs were found in the Chinese *Daphnia pulex* mitochondrial genome. They ranged from 63 to 72 bp in size. A schematic drawing of their respective secondary structures is shown in Figure 3. All tRNA genes had a clover-leaf structure except for *trnS1*, in which its DHU arm simply formed a loop. This loop in *trnS* is not uncommon in metazoan mitochondrial genomes (Crease and Little 1997). Whether or not the aberrant tRNAs lose their respective functions is still unknown. However, it's possible this anomaly may be rectified by subsequent RNA-editing mechanisms (Lavrov et al. 2000, Masta and Boore 2004, Li et al. 2012).

Non-canonical pairs, which possessed non Watson-Crick matches, commonly manifest in mitochondrial tRNA gene secondary structures. There are 30 base pair mismatches present in the tRNA secondary structures of Chinese *Daphnia pulex* mtD-NA, including 15 wobble G-U pairs, 13 U-G pairs, two U-U pairs, one A-A pair and one U-C pair mismatch (Fig. 3). Nevertheless, the post-transcriptional RNA-editing mechanism can rectify these mismatches to maintain tRNA functions (Tomita et al. 2001, Wang et al. 2014).

rRNA genes

Both the *rrnL* and *rrnS* genes were present in Chinese *Daphnia pulex* mitochondrial genome. They were located between *trnL1* and the non-coding putative control region and separated by *trnV*, as similarly found in vertebrate mitochondrial genomes (Delisle and Strobeck 2002, Hwang et al. 2008, Chao et al. 2014).

Large and small ribosomal RNA genes (*rrnL* and *rrnS*) in Chinese *Daphnia pulex* were 1,313 bp and 744 bp long, respectively. The lengths of the two rRNAs were almost similar to that of the *Daphnia pulex* in North America (1,314 bp and 753 bp, respectively).

Non-coding sequence

There are 15 non-coding regions ranging from 1 to 31 bp except for the A+T-rich region in the Chinese *Daphnia pulex* mitochondrial genome.

A 31 bp intergenic sequence was present between ND4L and trnT, which is also found in the North American *Daphnia pulex* mitochondrial DNA. The longest inter-



Figure 4. Phylogenetic tree obtained by the maximum-likelihood (ML) method and bootstrap values (1000 repetitions) of the branches were indicated. *D.magna* and *D. carinata* were used as outgroups.

genic region in Chinese *Daphnia pulex* was the A+T-rich region. It was between *rrnS* and *trnI* with the length of 723 bp. It has an A+T content of 65.42%. It was a little longer than that of the North American *Daphnia pulex* mitochondrial DNA (689 bp), but lower in A+T content. This region usually contains replication and transcription areas in both vertebrates and invertebrates (Zhang and Hewitt 1997, Boore 1999). The stem-loop structure and the quantity of multiple repeats of AT sequences are notable features of the control region, ranging from 200 bp to 1,300 bp, and determine the difference in arthropod mitochondrial DNA size (Boore 1999).

Phylogenetic analyses

The phylogenetic relationships among the *Daphnia pulex* from different areas were reconstructed based on nucleotide sequences of the *COI* gene by using the maximum likelihood (ML) mothod (Fig. 4). The phylogenetic analyses show that the Chinese and North American *Daphnia pulex* are recovered as two monophyletic clades with strong bootstrap support values (bs=100). They maybe evolved into two different species.

Conclusion

The mapping of the mitochondrial genome of the Chinese *Daphnia pulex* was completed in this study. It was found to be 15,306 bp in length and had a similar composition in size and structure to the *Daphnia pulex* mitochondrial DNA in North America published in GenBank AF117817 (Crease 1999). However, the phylogenetic analysis showed that the Chinese and North American *Daphnia pulex* maybe evolved into two different species (Fig. 4). The complete mitogenome of the Chinese *Daphnia pulex* reported here is expected to supply more molecular information for further studies of the *Daphnia* phylogeny and for analyses on the taxonomic status of the Cladocera.

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RESEARCH ARTICLE



Yuebeipotamon calciatile, a new genus and new species of freshwater crab from southern China (Crustacea, Decapoda, Brachyura, Potamidae)

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Abstract

A new genus and species of freshwater crab, *Yuebeipotamon calciatile* gen. n., sp. n., is described from southern China. While the carapace features are superficially similar to species of *Sinopotamon* Bott, 1967, *Longpotamon* Shih, Huang & Ng, 2016, and *Tenuilapotamon* Dai, Song, Li, Chen, Wang & Hu, 1984, the new genus possesses a distinctive combination of carapace, ambulatory leg, male thoracic sternal, male abdominal, and gonopodal characters that distinguish it from these and other genera. Molecular evidence derived from the mitochondrial 16S rDNA supports the establishment of a new genus.

Keywords

China, freshwater crabs, new genus, new species, Potamidae, systematics, *Yuebeipotamon calciatile*, 16S rDNA

Introduction

The South China region is diverse in freshwater crabs from the family Potamidae Ortmann, 1893. Despite its large land mass, Guangdong has a relatively low diversity when compared to other South Chinese provinces (Dai 1999, Shih and Ng 2011), which is probably the result of insufficient surveys conducted in this region.

In the past few years, there has been a growing trend in the aquarium trade for colorful freshwater crabs from South China, with species from the genera Nanhaipotamon Bott, 1968, Hainanpotamon Dai, 1995, Neilupotamon Dai & Türkay, 1997, and Heterochelamon Dai & Türkay, 1997, sometimes showing up in pet shops and even exported to other countries. We initially obtained one such species from the trade that has relatively long ambulatory legs with reddish to purplish coloration, which was interesting as it possessed a unique male first gonopod structure. The native ornamental fish dealer who sold these crabs to us eventually agreed with the first author's request to conduct a survey at his collection site, which was in northern Guangdong. This new species was compared to all known genera from around the region and while superficially similar to Sinopotamon Bott, 1967, Longpotamon Shih, Huang & Ng, 2016, and Tenuilapotamon Dai, Song, Li, Chen, Wang & Hu, 1984, in general carapace morphology (Dai 1999, Shih et al. 2016), it can immediately be differentiated by distinctive combinations of carapace, ambulatory leg, male thoracic sternal, male abdominal and gonopod characters. A molecular analysis conducted using the mitochondrial 16S rRNA marker also suggests that it does not belong to any known genera. Therefore, a new genus is established in this paper for this new species.

Material and methods

Specimens were collected from Yingde City of northern Guangdong, preserved in 75% ethanol and have been deposited in the Sun Yat-sen Museum of Biology, Sun Yat-sen University (SYSBM), Guangzhou, China, and the National Zoological Museum of China, Institute of Zoology, Chinese Academy of Sciences (IZCAS), Beijing, China. Measurements, in millimeters, are for the carapace width and carapace length. The following abbreviations are used: G1 – male first gonopod; G2 – male second gonopod.

Genomic DNA was isolated from the muscle tissue of ambulatory legs by using the Tiangen universal DNA purification kit (Beijing, China) and GeneMark tissue and cell genomic DNA purification kit (Taichung, Taiwan). A region of ~550 basepairs (= bp) of the 5'-end of the 16S gene was selected for amplification with polymerase chain reaction (PCR) using the primers 1471 and 1472 (Crandall and Fitzpatrick 1996). The PCR conditions were denaturation for 45 s at 94 °C, annealing for 40 s at 45 °C, and extension for 120 s at 72 °C (35 cycles), followed by extension for 10 min at 72 °C. Sequences were obtained by automated sequencing (Applied Biosystems 3730) and were aligned with the aid of ClustalW (vers. 1.4, Thompson et al. 1994), after verification with the complementary strand. To confirm the systematic position

of this species, the 16S sequences of genera from the eastern Asian continent in Shih et al. (2009), as well as the more recently described genus *Minutomon* Huang, Mao & Huang, 2014, were included for comparison. Sequences of the haplotypes have been deposited in a DNA Data Bank of Japan (DDBJ). We followed Shih et al. (2009) to exclude the variable regions in loop regions of the 16S which could not be aligned adequately for phylogenetic analyses.

The best-fitting model for sequence evolution of the 16S dataset was determined by MrModeltest (vers. 2.2, Nylander 2005), selected by the Akaike information criterion (AIC). The best model obtained was HKY+I+G, and was subsequently applied for Bayesian inference (BI) and maximum likelihood (ML) analyses. The BI analysis was performed with MrBayes (vers. 3.2.2, Ronquist et al. 2012) and the search was run with four chains for 10 million generations, with trees sampled every 1000 generations. The convergence of chains was determined by the effective sample size (ESS) (>200 as recommended) in Tracer (vers. 1.5, Rambaut and Drummond 2009) and the first 1000 trees were discarded as the burnin (determined by the average standard deviation of split frequency values below the recommended 0.01; Ronquist et al. 2005). ML analysis was conducted in GARLI (vers. 2.0, Zwickl 2006), with 10 replicate searches (searchreps = 10) and 100 bootstraps (bootstrapreps = 100) and the consensus tree from the GARLI output was computed using the program PAUP* (vers. 4.0b10, Swofford 2003) to assess node supports.

Systematic account

Family Potamidae Ortmann, 1896

Yuebeipotamon gen. n.

http://zoobank.org/BF003144-6BF4-43AD-8772-DDF577DD6F22

Diagnosis. Carapace subquadrate, with dorsal surface slightly convex, surface generally smooth, rugose on anterolateral regions (Fig. 2A); postorbital and epigastric cristae distinct, not confluent (Fig. 2A); external orbital angle sharply triangular, separated from anterolateral margin by a narrow gap (Fig. 2A, B); median lobe of posterior margin of epistome sharply triangular (Fig. 2B); third maxilliped with relatively broad ischium, exopod of third maxilliped reaches beyond anterior edge of ischium, with short flagellum (Fig. 3D); male abdomen triangular, with short triangular telson (Fig. 2C); G1 generally slender, terminal segment large, elongated, with subbasal flap (Figs 2D, 3B, C); basal segment of G2 subquadrate (Fig. 3A).

Type species. *Yuebeipotamon calciatile* sp. n., by monotypy.

Etymology. The genus name is derived from the Chinese spelling system "Yue Bei", which means northern Guangdong, for the locality of this genus. The suffix "Potamon" refers to the type genus of the family Potamidae, *Potamon*. Gender of genus neuter.

Character	Yuebeipotamon	Tenuilapotamon	Sinopotamon	Longpotamon
Enibern shial saash	distinct, sharp	indistinct, granular (cf.	varied (cf. Dai 1999:	varied (cf. Zou et. al.
Epibranchiai teeth	(Fig. 2A)	Dai 1999: pl. 29)	pl. 17)	2008: fig. 1)
A	1	slender (cf. Dai 1999:	stout (cf. Dai	stout (cf. Zou et. al.
Ambulatory legs	siender (Fig. 2A)	pl. 29)	1999: pl. 17)	2008: fig. 1)
Size of triangular	rolativoly largo	rolativoly opall	relatively small (cf.	relatively small (cf.
structure of male	(Fig. (C)	(Shih et al. 2016:	Shih et al. 2016:
thoracic sternites 1, 2	(Fig. 4C)	(unpublished data)	fig. 3C)	fig. 6C)
	relatively short,	relatively long,	relatively long, nipple	relatively long,
Male telson	triangular	subtriangular (cf. Dai	shaped (cf. Dai 1999:	subtriangular (cf. Zou
	(Fig. 2C)	1999: fig. 225)	fig. 139)	et. al. 2008: fig. 2)
	long, reaching	short, not reaching	long, reaching	short, not reaching
C1	beyond tubercle	tubercle of abdominal	beyond tubercle of	tubercle of abdominal
GI	of abdominal lock	lock (cf. Dai 1999:	abdominal lock (cf.	lock (cf. Dai 1999: fig.
	(Fig. 2D)	fig. 225)	Dai 1999: fig. 139)	137)
T	lo006Ecxcg, with	short, without	short, without	short, without subbasal
reminal segment	subbasal flap	subbasal flap (cf. Dai	subbasal flap (cf. Dai	flap (cf. Zou et. al.
10101	(Fig. 3C)	1999: fig. 225)	1999: fig. 139)	2008: fig. 2)

Table I. Morphological differences among *Yuebeipotamon* gen. n., *Tenuilapotamon* Dai, Song, Li, Chen, Wang & Hu, 1984, *Sinopotamon* Bott, 1967, and *Longpotamon* Shih, Huang & Ng, 2016.

Remarks. Although *Sinopotamon, Longpotamon, Tenuilapotamon, and Yuebeipotamon* are superficially similar in carapace features, *Yuebeipotamon* can easily be distinguished from by a number of characters (Table 1).

Comparative material from China. Sinopotamon kwanhsiense Tai & Sung, 1975: 1 \bigcirc (45.3 × 36.0 mm), IZCAS CB7659, Chengdu, Sichuan, 1984. Sinopotamon pingshanense Dai & Liu, 1994: 1 \bigcirc (45.2 × 34.3 mm), IZCAS CB8278, Muchuan, Sichuan, Oct. 1986. Longpotamon anyuanense (Dai, Zhou & Peng, 1995): 1 \bigcirc (45.7 × 39.1 mm) (SYSBM 001080), Shaoguan City, Guangdong, Apr. 2013; Longpotamon chekiangense (Tai & Sung, 1975): 1 \bigcirc (32.0 × 26.1 mm) (SYSBM 001079), Lucheng District, Wenzhou City, Zhejiang, Mar. 2013; Longpotamon fukienense (Dai & Chen, 1979): 1 \bigcirc (45.7 × 39.1 mm) (SYSBM 001054), Fuqing City, Fujian, May 2013. Tenuilapotamon joshuiense (Dai, Song, He, Cao, Xu & Zhong, 1975): 1 \bigcirc (23.6 × 19.3 mm) (SYSBM 001270), Lianyuan City, Hunan, Sep. 2013; 1 \bigcirc (23.7 × 19.4 mm) (SYSBM 001271), same data as above.

Yuebeipotamon calciatile sp. n.

http://zoobank.org/7DF2B1CA-5E8A-42D1-837D-B35EDA68B10C Figs 1–4

Material examined. Holotype: \bigcirc (32.4 × 27.0 mm) (SYSBM 001294), Yingde, Guangdong, China, karstic hillstream, coll. C. Huang, Jun. 2014. Paratypes: 1 \bigcirc (allotype) (33.0 × 27.9 mm) (SYSBM 001295), same data as holotype; 2 $\bigcirc \bigcirc$ (40.6 × 32.5



Figure 1. *Yuebeipotamon calciatile* gen. n., sp. n., color in life. **A** male, specimen not collected **B** a limestone hill stream at the type locality.

mm, 41.0 × 32.9 mm) (SYSBM 001296, 001297), same data as holotype; $1 \circ (37.9 \times 31.8 \text{ mm})$ (IZCAS), same data as holotype. Others: $3 \circ \circ (38.3 \times 31.7 \text{ mm}, 36.5 \times 29.9 \text{ mm}, 20.7 \times 17.4 \text{ mm})$ (SYSBM 001298, 001299, 001300), Yingde, Guangdong,



Figure 2. *Yuebeipotamon calciatile* gen. n., sp. n., male holotype (32.4 × 27.0 mm) (SYSBM 001294). **A** dorsal view **B** frontal view of carapace **C** ventral view showing anterior thoracic sternum and abdomen **D** ventral view showing sterno-abdominal cavity with right G1 in situ (left G1 removed).

China, karstic hill stream, coll. G.-H. Yuan, May 2014; $2 \bigcirc \bigcirc (17.1 \times 14.2 \text{ mm}, 18.2 \times 14.8 \text{ mm})$ (SYSBM 001301, 001302), same data as above.

Diagnosis. As for genus.

Description. Carapace subquadrate; dorsal surface slightly convex transversely, longitudinally; surface with rugose on anterolateral region (Fig. 2A). Front slightly deflexed, margin almost straight on dorsal view (Fig. 2A). Epigastric cristae low, separated by narrow gap (Fig. 2A, B). Postorbital cristae blunt, laterally expanded, not fused with epigastric cristae or reach the anterolateral margin (Fig. 2A, B). Branchial regions slightly convex (Fig. 2A). Cervical groove shallow, inconspicuous (Fig. 2A). Mesogastric region slightly convex (Fig. 2A). External orbital angle sharply triangular (Fig. 2A). Epibranchial tooth pointed, distinct (Fig. 2A, B). Anterolateral margin distinctly cristate, lined with approximately 17-19 granules; lateral part bent inwards (Fig. 2A). Posterolateral margin comparatively smooth, lined with multiple oblique striae, converging towards posterior carapace margin (Fig. 2A). Orbits large; supraorbital and infraorbital margins cristate, lined with numerous inconspicuous granules (Fig. 2B). Suborbital, subhepatic and upper parts of pterygostomial regions covered with rounded granules (Fig. 2B). Third maxilliped with merus about 1.1 times as broad as long; ischium about 1.5 times as long as broad; merus trapezoidal, with median depression; ischium trapezoidal, with distinct median sulcus; exopod reaching to proximal third of merus, with short flagellum reaching proximal three-fifths width of



Figure 3. *Yuebeipotamon calciatile* gen. n., sp. n., male holotype (32.4 × 27.0 mm) (SYSBM 001294). **A** left G2 **B** left G1 (ventral view) **C** terminal segment of G1 (ventral view) **D** left third maxilliped. Scale bar 1.0 mm.



Figure 4. *Yuebeipotamon calciatile* gen. n., sp. n., male holotype (32.4 × 27.0 mm) (SYSBM 001294). **A** left G1 (ventral view) **B** terminal segment of G1 (ventral view) **C** anterior thoracic sternum.

merus; upper-inner margin of ischium forming subauriculiform structure (Figs 2B, 3D). Posterior margin of epistome narrow; median lobe sharply triangular, lateral margins almost straight (Fig. 2B).

Chelipeds unequal (Fig. 2A). Merus cross-section trigonal; margins crenulated, dorsal-outer surface granulated (Fig. 2B). Carpus with sharp spine at inner-distal angle, spinule at base, dorsal surface with curved striae (Fig. 2A). Palm of larger chela about 1.6 times as long as high. Movable finger equal to fixed finger (Fig. 2A). Inner margin of fingers with rounded, blunt teeth; with small gap when fingers closed.

Ambulatory legs relatively slender, surfaces generally smooth (Fig. 2A). Last leg with propodus about 2.5 times as long as board, approximately same length as dactylus (Fig. 2A).

Male thoracic sternum generally smooth, weakly pitted; sternites 1, 2 completely fused to form triangular structure; sternites 2, 3 separated by continuous suture; sternites 3, 4 fused without obvious median suture; male sterno-abdominal cavity reaching to imaginary line joining median part of coxae of cheliped; median longitudinal groove between sternites 7, 8 deep (Figs 2C, 4C).

Male abdomen narrowly triangular; somites 3–6 progressively broader longitudinally; somite 6 about 1.9 times as board as long; telson about 1.5 times as board as long with a rounded tip, lateral margins of telson slightly concave (Fig. 2C). G1 generally slender; terminal segment large, elongated, inner margin with subbasal flap; tip of terminal segment reaches beyond tubercle of abdominal lock in situ; distal part of subterminal segment relatively narrow; subterminal segment about 1.3 times as long as terminal segment (Fig. 2D, 3B, C). G2 basal segment about 2.8 times length of flagelliform distal segment (Fig. 3A).

Variation. Adult specimens are usually much more brightly colored than juveniles. The terminal segment of the G1 may vary in proportionate length, while the angle at which it points varies from around 45–60 degrees.

Etymology. The species name, "*calciatile*", means living on limestone, relating to its natural habitat.

Color. Carapace is usually maroon to dark brown, while chelipeds and ambulatory legs are reddish to purplish in life (Fig. 1A).

Ecology. This primarily aquatic species is found in the pools of limestone hill streams where they hide in crevices. Almost each pool was occupied by at least one crab at the type locality, which is a relatively high density of distribution. Its slender legs indicate that this species has good climbing abilities and mobility on land. These abilities are assumed to be advantageous in the volatile and short-lived nature of limestone hill streams, which may force them to intermittently find new water sources. No other potamids were observed at the type locality.

DNA analyses and discussion

In total, 51 species from 44 genera of potamids were included in the phylogenetic analyses. A 503 bp segment, excluding the variable regions, of the 16S rDNA was amplified and aligned. The accession numbers of the 16S sequences of Yuebeipotamon calciatile sp. n. and Minutomon shanweiense Huang, Mao & Huang, 2014 are LC176064 and LC176065, respectively. The phylogenetic tree of the 16S was reconstructed using BI analysis, with support values from ML analysis (Fig. 5). The tree strongly indicates that Yuebeipotamon does not belong to any one of the genera included in this study, giving support to the current taxonomic treatment, i.e. it is a new genus. From its basal position to most known genera from East Asia and Southeast Asia, it suggests that this genus might be from an ancient lineage. However, Yuebeipotamon is distributed in Guangdong Province, part of the Pearl River Basin, which is thought to have younger lineages due to its distance from the proposed center of origin for the Potamidae, Yunnan Province (Shih and Ng 2011). This indicates that the ancestor for the genus may have dispersed to the eastern regions of China earlier than previously thought. More genetic markers are necessary to reveal the exact relationship of this genus. In addition, the recently established Minutomon (see Huang et al. 2014) is also supported genetically and belongs to the "China-East Asia Islands" clade which is closely related to genera from continental China (Shih et al. 2009).



Figure 5. A Bayesian inference (BI) tree of 16S rDNA for the subfamily Potamiscinae, with the sequences in Shih and Ng (2009), as well as *Yuebeipotamon* gen. n. and *Minutomon* Huang, Mao & Huang, 2014 (gray highlighted). Probability values at the nodes represent support values for BI and maximum likelihood (ML). Only values > 50% are shown.

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RESEARCH ARTICLE



Revision of the Palaearctic Gasteruption assectator aggregate, with special reference to Sweden (Hymenoptera, Gasteruptiidae)

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Abstract

The Palaearctic species of the *Gasteruption assectator* aggregate (Hymenoptera, Gasteruptiidae) are revised and three species are recognised. Two species are re-instated: *Gasteruption boreale* (Thomson, 1883), **stat. n.** and *G. nigritarse* (Thomson, 1883), **stat. n.**, and both are excluded from the synonymy with *G. assectator* (Linnaeus, 1758). The general distribution of both species is given for Europe and in detail for Sweden. A key to the valid Palaearctic species of the *Gasteruption assectator* aggregate is given; key characters and primary types are illustrated. Four new synonyms are listed: *Foenus fumipennis* Thomson, 1883, *Trichofoenus breviterebrae* Watanabe, 1934, and *Gasteruption margotae* Madl, 1987, are synonymized with *Gasteruption boreale* (Thomson, 1883) and *Gasteruption brevicauda* Kieffer, 1904, with *G. undulatum* (Abeille de Perrin, 1879).

Keywords

Europe, Gasteruption boreale, Gasteruption nigritarse, key, new records, re-instated species, Sweden, synonyms

Introduction

The predator inquiline wasp Gasteruption assectator (Linnaeus, 1758) (Hymenoptera, Gasteruptiidae) has been considered a very common species with wide intraspecific variation concerning both morphology and colouration (e.g. van Achterberg and Talebi 2014). When working on an updated revision of the Nordic Gasteruption species the first author noticed that the specimens from a restricted geographical range could be clustered into three separate morphospecies. The discovery of a couple of hitherto unknown features of the females made it fairly easy to separate the morphospecies involved. Further studies of a larger number of specimens and conclusions drawn from practical field work showed that the three morphospecies have a significant difference in geographical distribution and habitat preference in Sweden. Studies of the type material of the supposed synonyms of G. assectator showed that Foenus borealis Thomson, 1883, and F. nigritarsis Thomson, 1883, are the oldest available names for these valid species. Nearctic synonyms listed by Smith (1996) are not accounted for here because their types need to be studied first. The synonymisation of Gasteruption brevicauda Kieffer, 1904, with G. assectator made by Madl (1987a) is here rejected and it is considered to be conspecific with G. undulatum (Abeille de Perrin, 1879), syn. n.

Material

The first author studied specimens of the *Gasteruption assectator* aggregate deposited in the Evolutionsmuseet, Uppsala; The Swedish Malaise Trap Project (SMTP); the Biologiska Museet (MZLU), Lund and the Naturhistoriska Riksmuseet (NHRS), Stockholm. In addition the private collections of Anders Nilsson, Johan Abenius, Sven Hellqvist, Bo G. Svensson and Niklas Johansson. The results were checked by the second author with specimens deposited in the Naturalis Biodiversity Center (RMNH), Leiden and the Oberösterreichisches Landesmuseum, Biologiezentrum (BZL), Linz.

Systematics

Gasteruption assectator (Linnaeus, 1758) sensu stricto

Figs 1-3, 25-26, 28

- *Ichneumon assectator* Linnaeus, 1758: 566, 1761: 407, 1767: 937; Scopoli 1763: 287; Fabricius 1775: 340, 1781: 435, 1787: 268; Gmelin 1790: 2696; Villers 1789: 174; Rossi 1790: 90; Christ 1791: 375; Petagna 1792: 365; Cederhjelm 1798: 163; Schrank 1802: 263; Hentschius 1804: 112; Illiger 1807: 74; Roman 1932: 2; Hedqvist 1973: 182; Fitton 1978: 376.
- *Foenus assectator*; Fabricius 1798: 240; Walckenaer 1802: 75; Latreille 1805: 195; Dahlbom 1831: 77; Curtis 1832: 423; Nees 1834: 308; Stephens 1835: 121;

Labram and Imhoff 1836: 24; Zetterstedt 1840: 408; Westwood 1843: 255; Taschenberg 1866: 93; Tournier 1877: ix (as *affectator*); Thomson 1883: 849. *Foenus affectator*, Abeille de Perrin 1879: 265, 266, 277.

Gasteruption assectator; Schletterer 1885: 276, 316, 1889: 384, 393, 395, 397; Dalla Torre 1902: 1063; Szépligeti 1903: 370 (as *affectator*); Kieffer 1912: 256 (id.); Lindemans 1921: 298 (id.); Roman 1932: 2; Schmiedeknecht 1930: 380, 383 (as *affectator*); Hedicke 1939: 5 (id.); Ferrière 1946: 235, 238, 240 (id.); Leclercq 1948: 75; Hellén 1950: 4; Townes 1950: 123–128; Šedivý 1958: 36, 37; Györfi and Bajári 1962: 48, 51; Schmidt 1969: 293; Hedqvist 1973: 181; Fitton 1978: 376; Dolfuss 1982: 22; Oehlke 1984: 169, 171, 175; Ortega and Baez 1985: 509, 515; Madl 1987a: 401, 1987b: 21, 1988: 37, 1989a: 159, 1989b: 41, 1990a: 127, 1990b: 480; Kozlov 1988: 245, 247; Kofler and Madl 1990: 320; Narolsky and Shcherbal 1991: 23, 24; Wall 1994: 150; Scaramozzino 1995: 3; Smith 1996: 492; Peeters 1996: 134; Neumayer et al. 1999: 220; Pagliano and Scaramozzino 2000: 11, 19; Saure 2001: 29; Yildirim et al. 2004: 1350; Turrisi 2004: 84; Westrich 2008: 7–8; van der Smissen 2010: 372; Zhao et al. 2012: 23–27; van Achterberg 2013: 82; van Achterberg and Talebi 2014: 57–61.

Gasteruption affectator; Semenov 1892: 200.

Ichneumon annularis Geoffroy in Fourcroy 1785: 398; Hedicke 1939: 7; Wall 1994: 148 (type lost). Synonymized by with *G. assectator* (Linnaeus) by Olivier (1792).

Type material. High resolution photos of the lectotype female of *G. assectator* in the Linnaean collection coll. no 2652- "49 *assectator*" (Figs 1–3) designated by van Achterberg and Talebi (2014) was studied. The specimen has an unusually short ovipositor and the pilosity of the sheath is longer than average, but within the variation of the species. The holotype female of *G. brevicauda* (Figs 4–7) from Algeria (Orléansville) was examined and the specimen, with its strongly sculptured mesoscutum, the strong antero-lateral teeth of the pronotum aswell as the orange hind tarsus clearly belongs to *Gasteruption undulatum* (Abeille de Perrin, 1879). The synonymisation with *G. assectator* made by Madl (1987a) is here rejected and *G. brevicauda* Kieffer, 1904, is a new synonym of *G. undulatum* (Abeille de Perrin, 1879) syn. n.

Additional material. Sweden (*Skåne*: Åhus, *Blekinge*; *Halland*: Breared; *Små-land*: Repperda, Bäckebo, Hälleskog, Tvärskog, Robacken, Igersdela, Skillingaryd, Södra Vi, Korsberga; *Gotland*: Ardre, Stora Karlsö, Fårö, Mullvalds; *Öland*: Halltorp, Ekerum, Glömminge; *Östergötland*: Simonstorp, Borensberg)

Diagnosis. Temples in dorsal view less parallel-sided and usually shorter than of *G. boreale*, head in dorsal view transverse, mostly distinctly wider than long. Occipital carina indistinct and not reflexed. Face mostly slightly narrower than that of *G. boreale*. Hypostomal bridge narrow, at most 0.5 times mandibular base (Fig. 25). Mesoscutum in most cases distinctly reticulate-coriaceous and without satin sheen (Fig. 26), medioposteriorly in front of scutellum distinctly rugose. Mesosoma and head silvery pilose. Mesosomal surface with a fatty gloss, quite distinct from the more opaque satin sheen in *G. boreale*. Antenna slightly longer than in *G. boreale*, with sixth segment about



Figures 1–3. Lectotype of *Gasteruption assectator* (Linnaeus). I habitus dorsal 2 habitus lateral 3 labels.

1.8 times longer than wide and subapical segment about 1.5 times longer than wide. Hind coxa dorsally striate-rugose. Hind tibia and basitarsus with white ring which might be interrupted ventrally. Metasoma mainly black with lateral orange patches on tergites 2–4 often merged. Fore and middle tibiae with small, but quite distinct white or yellow patch basally. Ovipositor sheath black or brown, 1.0–1.3 times as long as hind tibia and without prominent bristles but with thinner adpressed pubescence, appearing nearly naked (Fig. 28). The pilosity of equal intensity all over the surface not becoming scarcer towards the tip. In some specimens, especially when the sheath parts



Figures 4–7. Lectotype of *Gasteruption brevicauda* (Kieffer). **4** habitus lateral **5** metasoma lateral **6** labels **7** mesoscutum and head dorsal.

are twisted as in the lectotype female, the pilosity might be slightly raised. The species is closely related to *G. boreale* (Thomson, 1883) and *G. nigritarse* (Thomson, 1883), but the female can be distinguished by the slightly longer ovipositor without conspicuous bristles. The male is distinguishable by its slightly shorter head in dorsal view and the often more distinctly reticulate-rugose mesoscutum without satin sheen.

Distribution. *G. assectator* is the most widespread and common species of the *assectator* aggregate in Europe. Towards its northern distribution limits in northern Scandinavia it seems to be confined to coastal areas with more favorable climate than inland areas.

Biology. *Gasteruption assectator* occurs in a wide variety of habitats, varying from agricultural landscapes to deciduous forests and gardens. Most probably *Hylaeus* spp. are used as hosts.

Gasteruption boreale (Thomson, 1883), stat. rev.

Figs 8–18, 27, 29

Foenus borealis Thomson, 1883: 849; Hedicke 1939: 7; Hedqvist 1973: 181, 182 (invalid lectotype designation); Wall 1994: 148. Synonymized with *G. assectator* (Linnaeus) by Schletterer (1889) and with *G. minutum* (Tournier) by van Achterberg and Talebi (2014).

Gasteruption boreale; Schletterer 1885: 303.

- *Foenus fumipennis* Thomson, 1883: 848; Hedicke 1939: 7; Hedqvist 1973: 181, 182 (lectotype designation); Wall 1994: 148. Synonymized with *G. assectator* (Linnaeus) by Schletterer 1885. **Syn. n.**
- *Trichofoenus breviterebrae* Watanabe, 1934: 285; Hedicke 1939: 45. Synonymized with *G. assectator* (Linnaeus) by Pagliano and Scaramozzino (2000). **Syn. n.**
- *Gasteruption margotae* Madl, 1987c: 225–227, 1990b: 480; Wall 1994: 149. Synonymized with *G. assectator* (Linnaeus) by Madl (1990b). **Syn. n.**

Type material. In Thomsons collection in MZLU four males and one female are placed at the label *Foenus borealis*. Hedqvist (1973) states that the type series by Thomson only consisted of three males and one female but this is probably a simple miscalculation. The female (from Norway) was originally selected as lectotype by Hedqvist (1973) but the selection was declared as invalid (van Achterberg and Talebi 2014) because the listed original locality of the type series (= Lappland) excludes the selection of a lectotype from Norway. One male (Figs 8–11) was designated lectotype by van Achterberg and Talebi (2014) and the species was synonymized with *Gasteruption minutum* (Tournier, 1877). All males and the female in the type series belong to one distinct species (see key below). The wider malar space exhibited by the lectotype is clearly shorter than the mandibular base and fits within the range of this species and is not as long as in *G. minutum*.

The type series of *Gasteruption fumipenne* consists of the lectotype from Gotland. The size, habitus, antennae and smooth sculpture on the mesoscutum of the lectotype



Figures 8–11. Lectotype of *Gasteruption boreale* (Thomson). 8 habitus lateral 9 head dorsal 10 labels 11 head anterior.

(Figs 12–13) indicates, despite the lacking metasoma, that it concerns a female of *G. boreale*. This is the only specimen known from the Baltic island Gotland, but the type locality ("Olle hau"= Ulla hau, Fårö, Gotland, Sweden) was at the time of the collection an active sand dune field with old pines. It was quite a different ecosystem than at the mainland of Gotland where *G. assectator* is the most common (and now only?) of the three species. The other specimen under this label is a male of *G. assectator* from Scania (Skåne). The synonymisation with *G. assectator* made by Schletterer (1889) is rejected and *F. fumipennis* is to be regarded as a new synonym of *G. boreale* (Thomson).

The type series of *G. margotae* consists of the male holotype (Figs 14–15) from Finland (Madl 1987c); the holotype is a typical male of *G. boreale* (Thomson). The study of the holotype shows that the synonymisation with *G. assectator* made by Madl



Figures 12–13. Lectotype Foenus fumipennis Thomson. 12 habitus lateral 13 labels.

(1990) is unjustified after resolving the *G. assectator* aggregate and that *G. margotae* is clearly a new synonym of *G. boreale* (Thomson).

The examined type series of *G. breviterebrae* (Fig. 16) consists of the holotype female and a paratype male from Sakhalin (Far East Russia). The holotype shows the typical features of *G. boreale* (Thomson), viz., the less sculptured mesoscutum and the bristly ovipositor sheath. The slightly aberrant red marks on the metasoma fall within the geographical variation of *G. boreale*. The synonymisation with *G. assectator* made



Figures 14–15. Holotype of Gasteruption margotae Madl. 14 habitus lateral 15 labels.

by Pagliano and Scaramozzino (2000) is here rejected and *T. breviterebrae* is a new synonym of *G. boreale* (Thomson).

Additional material. Sweden (*Småland*: Bäckebo, Skillingaryd, Ränneslätt, Jönköping; *Västergötland*: Baskarp; *Södermanland*: Huddinge; *Uppland*: Rossholm; *Dalarna*: Leksand, Ludvika; Hälsingland: Hornslandet; *Västerbotten*: Vindeln, Hällnäs; *Lycksele lappmark*: Gällivare); Finland (*Åland*: Hammarland; *Åbo*: Harvaluoto).

Diagnosis. Head in dorsal view almost parallel-sided behind eyes, elongate, about as wide as long (Figs 9, 18). Occipital carina indistinct and not reflexed. Frons with satin sheen. Mesoscutum smooth, weakly rugose/shagreened with satin sheen, medioposteriorly in front of scutellum rugose-reticulate (Figs 17-18, 27). Mesosoma and head silvery pilose. Mesosoma with a satin sheen, quite distinct from the rather matt gloss occurring in G. assectator. Whitish pubescence of eye of female mostly distinctly longer and denser than of G. assectator. Antenna slightly shorter than in G. assectator with sixth segment about 1.5 times longer than wide and subapical segment about 1.2 times longer than wide. Only apical half of hind coxa weakly striate dorsally. Hind tibia and basitarsus with white ring which might be interrupted ventrally. Metasoma mainly black with orange lateral patches on tergites 2-4 which might be partially reduced, especially in northern specimens. Inner sides of tibiae often red brown to orange with white or yellow basal patch indistinct on fore and middle tibiae. Ovipositor sheath black or brown, 0.7-1.0 times as long as hind tibia, its apical half entirely with stout, rather scarce black bristles angled backwards at about 45° (Fig. 29). The species is closely related to G. assectator (Linnaeus) but the female can be distinguished by the shorter ovipositor sheath, the less sculptured mesoscutum and the more scarce promi-



Figure 16. Lectotype of Trichofoenus breviterebrae Watanabe, habitus lateral.

nent bristles on the apical half of the ovipositor sheath. The male is hard to distinguish from *G. assectator* and identification is not always possible with certainty. In most cases the male of *G. boreale* can however be separated from *G. assectator* by its slightly more elongated, parallel-sided head in dorsal view, the more or less enlarged malar space and its less sculptured mesoscutum.

Description. Female. Length of body 6–11 mm (fore wing 3.5–5.5 mm)

Head. Temples parallel-sided behind eyes in dorsal view. Occipital carina not raised. Frons and vertex with satin sheen. Malar space short, at most about 0.5 times mandibular base. Hypostomal bridge narrow, at most 0.5 times mandibular base. Eyes with dense white pubescence. Antenna short; sixth segment about 1.5 times longer than wide and subapical segment about 1.2 times longer than wide.

Mesosoma. Surface largely smooth with satin sheen, mesoscutal sculpture of almost equal intensity as on vertex. Antesternal carina narrow, non-lamelliform. Pronotal sides with very small pointed teeth antero-ventrally, but these are sometimes entirely absent. Upper half of mesopleuron mostly considerably weaker sculptured than its more rugose lower part.

Legs. Hind tibia stout as in *G. assectator*. Hind coxa often with weaker rugae apically than on basal half, dissolving amidst rugose background. Hind tibial spurs often brighter than hind tibia.



Figures 17–18. *Gasteruption boreale* (Thomson), Q. 17 habitus lateral 18 habitus dorsal.

Metasoma. Ovipositor sheath entirely black or brown, 0.7–1.0 times as long as hind tibia, its apical half entirely with stout, black bristles angled backwards at about 45° (Fig. 29).

Colour. Black. Mandible apically, hind tibial spurs and patches laterally on tergites 2–4 reddish brown. Patches rarely intercepted. Fore and middle tibiae mostly with indistinct yellow or ivory basal patch. Inner side of tibiae often orange. Basal ivory ring of hind tibia usually indistinct.

Distribution. As Thomson's name implies this species is most common in boreoalpine areas. In northern Europe it is quite widespread and common at higher latitudes and high altitude sites in the southern part, but becoming scarcer towards the southern lowlands in Sweden. Specimens are examined from Austria, Bulgaria, Finland, Germany, Netherlands (den Dolder, de Bilt, Tilburg, Wageningen, Groesbeek, Rhenen, Velp, Maastricht, Drimmelen, Rotterdam, Voerendaal), Norway, Russia, Serbia and Sweden.

Biology. The species occur in (boreal) landscapes dominated by coniferous forests where it can be locally common. Many of the sites in Scandinavia are at high altitude. *G. boreale* is lacking from more open localities as well as in regions dominated by deciduous forests. It has been observed searching high stumps of *Pinus* trees and it also attends old wooden walls and artificial bee nests in gardens. Probably it is a kleptoparasitoid of *Hylaeus* spp.

Gasteruption nigritarse (Thomson, 1883), stat. rev.

Figs 19-24, 30

Foenus nigritarsis Thomson, 1883: 849; Schletterer 1889: 398; Hedicke 1939: 7; Hedqvist 1973: 181, 182 (lectotype designation); Wall 1994: 149. Synonymized with *G. assectator* (Linnaeus) by Schletterer (1889). *Gasteruption nigritarse*; Schletterer 1885: 310.

Type material. Lectotype female (Figs 19–22) from Lund (Scania) selected by Hedqvist (1973). All 12 specimens under *Foenus nigritarsis*, both males and females (including the designated lectotype by Hedqvist) belong to the same distinct species and are well separable from *Gasteruption assectator sensu stricto* (see key below).

Additional material. Sweden (*Småland*: Bäckebo; *Skåne*; *Halland*; *Östergötland*: Svensksund; *Uppland*: Grisslehamn, Svartsjö, Roslagsbro, Skansen; *Öland*: Borgehage, Himmelsberga).

Diagnosis. Head dorsally more parallel-sided than in G. assectator, elongate and about as wide as long. Occipital carina indistinct and not reflexed. Mesoscutum superficially reticulate and (especially laterally) rugose, medio-posteriorly in front of scutellum more rugose-reticulate. Mesosoma laterally and face with long, thick golden pubescence. Hind tibia and basitarsus darker, often with the basal ring lacking or interrupted. Fore and middle tibiae often with large distinct ivory patch covering about one third of tibia. Metasoma mainly black with well-defined orange lateral patches on tergites 2-5 which might be partially reduced, especially in northern specimens. Ovipositor sheath entirely black or brown, 0.7–1.0 times as long as hind tibia and its apical half dorsally with stout, black bristles angled backwards at about 45°. The bristles are all conspicuously widened and bent apically, reminiscent of "velcro" (Fig. 30). The species is closely related to G. assectator (Linnaeus) and G. boreale (Thomson), but the female can be distinguished by the stout velcro-like bristles dorsally on the apical half of the ovipositor sheath, its denser pubescence of head and mesosoma and its broader hypostomal bridge. The male is distinguishable by its broader hypostomal bridge as well as the thick golden facial pubescence.

Description. Female. Length of body 8–11 mm (fore wing 4.0–5.5 mm)

Head. Temples parallel-sided behind eyes in dorsal view. Occipital carina not raised. Frons and vertex with satin sheen. Malar space short, at most about 0.5 times mandibular



Figures 19–22. Lectotype of *Gasteruption nigritarse* (Thomson). 19 habitus lateral 20 labels 21 head anterior 22 head ventral.

base. Hypostomal bridge at least 0.7 times width of mandibular base, medio-laterally often with distinct transverse striae. Face covered with dense golden pubescence.

Mesosoma. Surface vaguely reticulate and strongly shagreened. Antesternal carina narrow and non-lamelliform. Pronotal sides with very small pointed teeth antero-ventrally, but these are sometimes entirely absent.



Figure 23. *Gasteruption nigritarse* (Thomson), ♀, habitus dorso-lateral.

Legs. Hind tibia rather stout as in *G. assectator.* Hind tibial spurs and hind tibia mostly similarly coloured.

Metasoma. Ovipositor sheath entirely black or brown, 0.7–1.0 times as long as hind tibia, its apical half dorsally with stout, black bristles angled backwards at about 45° and conspicuously widened and bent backwards apically.

Colour. Black. Mandible apically orange. Sometimes small patch on hind tibia baso-ventrally white or ivory. Northern specimens of both sexes often with entirely black hind tibia. Fore and middle tibiae often with large distinct ivory patch covering about one third of tibia. Fore, middle and hind tarsus black. Patches laterally on tergites 2–5 reddish brown. Last sternite apically often extensively orange. Colour of hind tibial spurs variable, mostly black or dark brown.

Distribution. *Gasteruption nigritarse* is a rather rare locally but widespread species in Europe. Specimens examined from Austria, Germany, Czech Republic, Netherlands (Breda, Waalwijk, Putten (GE), Maastricht, Wageningen, Rhenen, Arnhem), Serbia, Sweden and Turkey.

Biology. The species primarily occur in small-scale agricultural landscapes where it is to be found especially on walls of log barns (Fig. 23). An association with the bees *Hylaeus difformis* and/or *Hylaeus pictipes* is highly probable, at least in Scandinavia and is based on observed behaviour of the wasps. *Gasteruption nigritarse* seems to have diminished dramatically in Scandinavia during the last century, probably due to the loss of habitat and is only known from a couple of localities.

Key to Palaearctic species of the Gasteruption assectator aggregate

1 _ 2 Hypostomal bridge 0.7-0.8 times as wide as mandibular base and weakly striate medio-laterally (Figs 22, 24). Occipital carina conspicuously bent inwards medio-ventrally, resulting in parallel running lower parts (Figs 22, 24). Hind tibia in northern populations often with reduced white markings. In lateral view apical half of ovipositor sheath with hooked bristles ("velcrotype"; Fig. 30). Facial pubescence thick and golden G. nigritarse (Thomson, 1883), stat. rev. Hypostomal bridge narrow, at most 0.5 times as wide as mandibular base and without striation medio-laterally (Fig. 25). Occipital carina evenly diverging medio-ventrally (Fig. 25). Hind tibia mostly with distinct white ring basally. Ovipositor sheath without "velcro"-type of bristles (Figs 28-29). Facial pu-Mesoscutum and coxae more opaque, less rugose, smoother and with satin 3 sheen (Figs 17-18, 27). Sculpture of mesoscutum and head similar, shagreened (Fig. 27). Ovipositor sheath 0.7-1.0 times as long as hind tibia, in dorsal view its apical half with more scarse distinct straight bristles, angled backwards at about 45° (Fig. 29) G. boreale (Thomson, 1883), stat. rev. Mesoscutum and coxae more shiny and rugose, with a "fatty" gloss (Fig. 26). Sculpture of mesoscutum distinctly rougher than that of head, reticulate-coriaceous (Fig. 26). Ovipositor sheath 1.0–1.3 times as long as hind tibia, in dorsal view normally only with dense bright adpressed pubescence, thus appearing naked in lower magnifications (Fig. 28)...... G. assectator (Linnaeus, 1758)



Figures 24–25. Hypostomal bridge of *Gasteruption nigritarse* (Thomson) (**24**) and *G. assectator* (Linnaeus) (**25**).



Figures 26–27. Mesoscutum of *Gasteruption assectator* (Linnaeus) (26) and *G. boreale* (Thomson) (27).

4	Hypostomal bridge 0.7-0.8 times as wide as width of mandibular base and
	weakly striate medio-laterally (Figs 22, 24). Occipital carina conspicuously
	bent inwards medio-ventrally, resulting in parallel running lower parts (Figs
	22, 24). Hind tibia in northern populations often with reduced white mark-
	ings. Facial pubescence thick and golden
	G. nigritarse (Thomson, 1883), stat. rev.
_	Hypostomal bridge narrow, at most 0.5 times width of mandibular base and
	without striation medio-laterally (Fig. 25). Occipital carina medio-ventrally
	evenly diverging. Hind tibia mostly with distinct white ring basally. Facial
	pubescence thin and silvery
5	Mesoscutum and upper half of mesopleuron rather smooth with small punc-
	ture-like grooves and with satin sheen almost of the same intensity as vertex



Figures 28–30. Ovipositor sheath of *Gasteruption assectator* (Linnaeus (**28**), *G. boreale* (**29**) and *G. nigritarse* (Thomson) (**30**). Scale bar 1mm.

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RESEARCH ARTICLE



Definition of the *jianfengling* species group of the ground beetle genus Orthogonius MacLeay (Coleoptera, Carabidae, Orthogoniini)

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Abstract

The *jianfengling* species group of the termitophilous carabid genus *Orthogonius* MacLeay, 1825 is defined and reviewed. This species group ranges from southern China, crossing Indochina and Myanmar to eastern India. To date, the *jianfengling* species group is composed of ten species, including six new species which are hereinafter described and illustrated: *O. wrasei* **sp. n.** (Myanmar), *O. bellus* **sp. n.** and *O. limbourgi* **sp. n.** (Vietnam), *O. politior* **sp. n.**, *O. aberlenci* **sp. n.** (Laos) and *O. meghalayaensis* **sp. n.** (India). Habitus, elytral apices and male genitalia of all species are illustrated. A key to species and a distribution map of *jianfengling* species group are provided.

Keywords

Carabids, Indo-Burma, new species, taxonomy, termitophilous

Introduction

Indo-Burma is one of the 25 biodiversity hotspots in the world for conservation priorities (Myers et al. 2000). It covers a large tropical area of more than two million square kilometers, from eastern India, through Myanmar to southern China to the east, and to northern of Malaya Peninsula to the south. The termitophilous ground beetles of the tribe Orthogoniini are very rich in species diversity in this area, represented by approximately 100 species (Tian and Deuve 2000, 2006, 2010, Tian et al. 2012).

The majority of orthogoniine species recorded in tropical Asia belongs to the prominent genus *Orthogonius* MacLeay, 1825. As the series works to understand the faunal composition and to set up a rational classification system of *Orthogonius*, the *lancangjiang* and *baconii* species groups have already been dealt with (Tian and Deuve 2013, 2016). In the present paper we define the *jianfengling* species group.

The *jianfengling* species group is comprised of members of large, polish, depressed and glabrous orthogoniine beetles. The first species of this group, *O. jianfengling* Tian & Deuve, was described in 2000 from Hainan Island, China. Then, *O. himalayicus* Tian & Deuve, 2005 (from Bhutan and Sikkim, India), *O. freyi* Tian & Deuve, 2006 (from Myanmar) and *O. duboisi* Tian & Deuve, 2006 (from Yunnan, China) were recorded respectively (Tian and Deuve 2005, 2006). To date, ten *Orthogonius* species are designed as members of the *jianfengling* species group, including six new species described below.

Material and methods

All specimens for this study are dry and mounted materials. Dissection and observation of the specimens were made using a WILD M32 binocular microscope. Detailed descriptions are provided for the new species, while only diagnostic character states are given for the known species. Digital photographs were taken and processed in the same as in Tian and Deuve (2013).

Body length was measured from apex of right mandibles to apex of elytra; body width = width of elytra.

Abbreviations of measurements used in the text are as followings:

- HL head length (from apex of right mandible to base of vertex);
- HW head width (maximum distance across head, including eyes);
- PL length of pronotum (measured from front to basal margins, through midline);
- **PW** width of pronotum (greatest width of pronotum);
- EL length of elytra (measured from base to apex of elytra, through suture);
- EW width of elytra (greatest transverse distance across both elytra).

Abbreviations of collections mentioned in the text are as following

CDW	Collection of David Wrase, Berlin
CIB	Collection of Ingo Brunk, Dresden
CIRAD	Agricultural Research for Development, Montpellier
HNML	Natural History Museum, London
IRSNB	Institut royal des Sciences naturelle de Belgique, Brussels
MNHN	Muséum national d'Histoire naturelle. Paris

NHMB	Naturalhistorisches Museum, Basal
NHMV	Natural History Museum of Vienna, Vienna
SCAU	South China Agricultural University, Guangzhou

Taxonomic treatment

Genus Orthogonius MacLeay, 1825

jianfengling species group

Definition. The *jianfengling* species group shares the following combination of morphological characteristics: (1) Medium to large sized; body elongate and more or less flat; polish and very smooth, strongly shiny, impunctate; (2) Labrum more or less concave or emarginate at frontal margin; ligula bisetose at apex; each of mentum and submentum bisetose; mentum edentate in most species (but toothed medially in O. meghalayaensis sp. n.); (3) Hind angle of pronotum rounded off; (4) Elytra well-bordered at base, nearly parallel-sided medially; striae shallow or deep, intervals convex, the 3rd interval with three setiferous pores (but median one wanted in *O. freyi*), the 7th interval not carinated; apex of elytra obliquely truncate, bisinuate or sinuate, outer apical angle rounded off, the inner apical angle various shaped, obtuse or rectangular, denticulate at tip or not; (5) Middle coxae setose medially in most species (but asetose in O. bellus sp. n., and O. limbourgi sp. n.); middle and hind tibiae slender, not dilated in male; femora moderately long; hind tibial spurs moderate in length, very sharp; the 4th hind tarsomere bilobed; hind femora bisetose to quadrisetose posteriorly; tarsal claws especially the hind ones weakly pectinate; (6) Abdominal ventrite VII in male not emarginate at apical margin; and (7) Male genitalia not exposed; aedeagus short and stout, widely opened dorsally, apical lamella short; parameres normal for Orthogonius.

Sexual dimorphism. In male, 1st (apical half) to 3rd protarsomeres bearing two rows of spongy setae which are flat and located along median portion (Figs 1–2); ventrite VII not emarginated in both sexes.

Distribution of the *jianfengling* **species group.** The *jianfengling* species group are distributed in the following countries: China (O. *jianfengling* and O. *duboisi*), Vietnam (O. *bellus* sp. n. and *limbourgi* sp. n.), Laos (O. *politior* sp. n. and O. *aberlenci* sp. n.), Myanmar (O. *freyi* and O. *wrasei* sp. n., India (O. *meghalayaensis* sp. n., and O. *himalayicus*) and Bhutan (O. *himalayicus*) (Fig. 3).

Key to species of *jianfengling* species group



Figures 1–2. Protarsi of *O. limbourgi* sp. n. 1 male 2 female.

_	Combination of other differences other species groups of Orthogonius
2	Prosternal process unbordered at apex
_	Prosternal process bordered at apex
3	Mentum with a median tooth, abdominal ventrite VII bearing three pairs of
	setae O. meghalayaensis sp. n.
_	Mentum edentate, abdominal ventrite VII bearing two pairs of setae4
4	Head slender, labrum deeply emarginate at front, eyes less convex
	O. himalayicus
_	Head stouter, labrum shallowly emarginate at front, eyes more convex
5	Pronotum and elytra entirely reddish brown; scutellum widely obtuse; lateral
	expanded margins of pronotum distinctly reflexed O. jianfengling



Figure 3. Distribution of the *jianfengling* species group **a** *O. jianfengling* **b** *O. limbourgi* sp. n. **c** *O. bellus* sp. n. **d** *O. politior* sp. n. **e** *O. aberlenci* sp. n. **f** *O. duboisi* **g** *O. wrasei* sp. n. **h** *O. freyi* **i** *O. himalayicus* **j** *O. meghalayaensis* sp. n.

-	At least disc of pronotum light dark brown; scutellum narrowly obtuse; lat-
	eral expanded margins of pronotum either flat or slightly reflexed
6	Elytral interval 3 with only two setiferous pores, middle one wanted O. freyi
_	Elytral interval 3 with three setiferous pores7
7	Body brown or yellowish brown8
_	Body black or dark brown10
8	Middle coxae setose medially, apical inner angle of elytra obtuse, not denticu-
	late at tip
_	Middle coxae asetose medially, apical inner angle of elytra nearly rectangular,
	distinctly denticulate at tip9

<i>i</i> sp. n.
s sp. n.
luboisi
<i>i</i> sp. n.
 i :

Orthogonius jianfengling Tian & Deuve, 2000

Figs 3a, 4, 14

Orthogonius jianfengling Tian & Deuve, 2000: 298.

Length. 15.0–17.0 mm; width: 5.5–6.0 mm. Habitus as in Fig. 4.

Description. Body elongate, strongly shiny, smooth and glabrous; head dark brown, pronotum, elytra and legs red brown, impunctate, microsculptural engraved meshes isodiametric. Head slightly longer than wide, HL/HW = 1.11; clypeus bisetose, labrum sex-setose, shallowly emarginate at apical margin; mentum edentate; each of mentum and submentum bisetose. Pronotum strongly transverse, PW/PL = 1.55, disc moderately convex, apical and basal margins well beaded, sides evenly expanded, widest at middle; base slightly wider than apex, lateral expanded margin well-defined, evenly and distinctly reflexed. Elytra elongate, and rather flat, EL/EW = 1.84, widest at about middle, parallel-sided at middle, basal border complete, apex bisinuate, inner angle blunt, not denticulate at tip (Fig. 14), the 3rd interval with three setiferous pores. Legs slender, fore tibiae expanded at apex, with outer angle strongly protrude, outer margin smooth, sub-serrate; middle coxae setose medially, hind coxae smooth and glabrous, the 3rd and 4th hind tarsomeres subequal in length, the 4th bilobed at apex; all tarsal claws weakly pectinate. Prosternal process bordered at apex.

Male: Unknown.

Remarks. It is probably close to *O. limbourgi* sp. n., distinguished from the latter by its distinctly reflexed lateral expanded margins of pronotum (slightly reflexed in *O. limbourgi*), and setose middle coxae (asetose in *O. limbourgi*).

Material examined. 1 female, the holotype, "Hainan: Ledong Xian: Jianfengling, 15. VI. 1983, Gu Maobin leg.", in SCAU; 1 female, a paratype, ibid. in MNHN.

Distribution. China (Hainan Island) (Fig. 3a).

Orthogonius himalayicus Tian & Deuve, 2005

Figs 3i, 5, 15, 24-25

Orthogonius himalayicus Tian & Deuve, 2005: 61.

Length. 19.0–20.0 mm; width: 6.5–7.0 mm (measurements in the original description were not correct). Habitus as in Fig. 5.



Figures 4–7. Habitus 4 *O. jianfengling* 5 *O. himalayicus* 6 *O. limbourgi* sp. n. 7 *O. politior* sp. n. Scale bar: 5.0 mm.

Description. Head and pronotum black, antennae and elytra dark brown, slender and very shinny; body smooth and glabrous, head and pronotum impunctate, elytral odd intervals with a few, sparse punctures which are irregularly arranged; head intricately wrinkled; microsculptural engraved meshes densely isodiametric, faint on head and pronotum, clear on elytra. Head slightly longer than wide, HL/HW = 1.15-1.18; labrum deeply emarginate at front; eyes comparatively small, but prominent; mentum edentate and bisetose, submentum bisetose. Pronotum transverse, PW/PL = 1.91–1.94, rather flat, lateral expanded margin wide and slightly reflexed, widest at middle. Elytra elongate and rather slender, EL/EW = 1.74-1.77, parallel-sided at middle, moderately convex, widest slightly behind middle, apex obliquely truncate, inner angles roundly obtuse (Fig. 15); striae very deep, intervals distinctly convex, the 3rd intervals generally with three setiferous pores. Legs moderately long, middle coxae setose, the 3rd and 4th hind tarsomeres subequal in length, all tarsal claws weakly pectinate. Prosternal process unbordered at apex. Male genitalia (Figs 24–25) with the median lobe of aedeagus comparatively small and short, middle portion convex, apex pointed in lateral view, turning to right, obtuse dorsally.

Remarks. Differs from other congeners by its deeply concaved labrum and broadly obtuse inner apex of elytron.

Material examined. 1 male, the holotype: "British Bootang, L. Durel, 1898", in MNHN; 2 males, paratypes, "Pedong, A. Desgodins"; 1 male and 2 females, paratypes, "British Bootang, Maria Basti. L. Durel"; 1 male, a paratype: "Sikkim" only; 1 female, a paratype: "Museum Paris, Bhutan. R. Oberthür, 1900", all in MNHN, except one male in SCAU.

Distribution. Bhutan and Sikkim (Fig. 3i).

Orthogonius limbourgi sp. n.

http://zoobank.org/107BEE2C-7B57-4A69-9CF4-E7B8FC708376 Figs 1–3b, 6, 16, 26–27

Holotype. Male, label "Coll. I.R.Sc.N.B./ Vietnam: Tam Dao NP, 25-28. VII, 2010, light trap, Leg. J. Constant & P. Limbourg; I.G. 31.668", in IRSNB.

Paratypes. 1 female, "Coll. I.R.Sc.N.B./ Vietnam: Cuc Phuong, Day coll. NG/ IG, 31.668, 11-18. VIII, 2010, Leg J. Constant & P. Limbourg", in IRSNB; 25 males & 14 females, "Coll. I.R.Sc.N.B./ Vietnam: Cuc Phuong N. P., 20.19'00" N, 105.36'30" E, 19-23. VII, 2011, night collecting, Leg J. Constant & P. Bresseel; I.G. 31.933", in IRSNB, MNHN and SCAU, respectively; 1 male, "N. Vietnam, Tamdao NP env. 1000 m, VI. 2012, ex coll. A. Popadic", in CIB; 2 females, in CIB and MNHN respectively.

Diagnosis. A medium-sized species, light dark brown in general, with rather flat and slender body which is smooth and glabrous, and asetose midcoxae and well bordered prosternal process.

Description. Length: 15.0–16.0 mm; width: 6.0–6.5 mm. Habitus as in Fig. 6.



Figures 8-11. Habitus 8 O. aberlenci sp. n. 9 O. bellus sp. n. 10 O. freyi 11 O. duboisi. Scale bar: 5.0 mm.

Body elongate, strongly shiny, smooth and glabrous. Head and disc of pronotum dark brown; elytra, lateral expanded margins of pronotum, mouthparts, antennae, legs and ventral surface reddish brown; whole body dark brown in several specimens. Surface impunctate, head moderately wrinkled on frons, smooth on vertex and pronotum. Microsculptural engraved meshes isodiametric on elytra, indistinct on head and pronotum.

Head longer than wide, HL/HW = 1.12-1.14, eyes small but prominent, frons and vertex moderately convex, frontal impressions large and deep; clypeus bisetose, basal portion even; labrum sex-setose, shallowly emarginate at front margin; palps slender, subcylindrical, the 3^{rd} maxillary palpomere as long as the 4^{th} which is glabrous, the 3^{rd} palpomere with two short setae at apex; the 2^{nd} labial palpomere slightly longer than the 3^{rd} , bisetose in inner margin, with several additional setae at subapex and apex, the 3^{rd} labial palpomere with a few setae at basal part; ligula small, bisetose at apex; mentum edentate; each of mentum and submentum bisetose (but an additional seta on the left of mentum in male), palpiger asetose. Antennae moderately long, extending over base of elytra; pubescent from basal 1/3 of the 4^{th} antennomere, slightly expanded at pubescent portion of the 4^{th} ; the 3^{rd} antennomere almost as long as the 4^{th} .

Pronotum strongly transverse, PW/PL = 1.59–1.62, disc moderately convex, apical and basal margins well beaded, sides evenly expanded, widest at middle; base slightly wider than apex, bisinuate on base, slightly concave on apical margin; hind angle rounded off; lateral expanded margin well defined, evenly and indistinctly reflexed; both transversal impressions distinct, basal foveae small and deep.

Elytra elongate, and rather flat; EL/EW = 1.64-1.65; widest at about middle, sides parallel at middle, basal border complete, apex bisinuate, inner angle broad, with a small denticle (Fig. 16); striae moderately deep, intervals convex, odd and even intervals subequal in width in middle, the 3^{rd} interval with three setiferous pores; the 7^{th} interval simple.

Legs slender, fore tibiae expanded at apex, with outer angle strongly protrude, outer margin serrate; hind femora rather slender, with three posterior setae; middle coxae setose, hind coxae smooth and glabrous; middle tibiae quite straight, slightly expanded at apex; hind tibiae slightly expanded at apex, apical spurs long and sharp, the 3rd hind tarsomere slightly longer than the 4th which bilobed at apex; all tarsal claws weakly pectinate.

Prosternal process well bordered at apex. Abdominal ventrite VII of male complete; ventrite VII with two pairs of setae on either side of apical margin in both male and female.

Male genitalia: Stout, dorsal opening wide and long, base dilated, gently bent ventrally towards apex, which almost pointed in lateral view; in dorsal view, not contracted before apical lamella, which short and broad, as long as wide, surface granulated.

Remarks. It is allied to *O. politior* sp. n., but its clypeus bisetose (quadrisetose in *O. politior*), prosternal process well-bordered at apex (unbordered in *O. politior*), and the apical lamella of aedeagus shorter and broad at apex (longer and narrow in *O. politior*).

Etymology. In honor of P. Limbourg (Brussels), one of the holotype collectors. **Distribution.** Northern Vietnam (Fig. 3b).



Figures 12–13. Habitus 12 *O. meghalayaensis* sp. n., holotype, male 13 *O. wrasei* sp. n., paratype, female. Scale bar: 5.0 mm.

Orthogonius politior sp. n.

http://zoobank.org/32834F91-D7F4-4801-B957-936966D44D1F Figs 3d, 7, 17, 28–30

Holotype. Male, "Laos-NE: Houa Phan Province, 20.13N/104.00E, Phou Pane Mt., 1350-1500 m, 1-16. VI. 2009, M. Brancucci leg.", "NHMB Basel, NMP Prague, Laos 2009 expedition: M. Brancucci, M. Geiser, Z. Kraus, D. Hauck, V. Kuban", in NHMB.

Diagnosis. A species similar to but larger than the above one, with also a rather flat and slender body, but having an unbordered prosternal process, a quadrisetose clypeus and setose midcoxae.

Description. Length: 19.0 mm; width: 6.8 mm. Habitus as in Fig. 7.

Body elongate, strongly shiny, smooth and glabrous. Head and disc of pronotum dark brown; elytra, lateral expanded margins of pronotum, mouthparts, antennae, legs and ventral surface reddish brown. Head and pronotum impunctate, elytral odd intervals with a few punctures; head moderately wrinkled; pronotum and elytra largely smooth. Microsculptural engraved meshes isodiametric on elytra, rather transverse on pronotum, indistinct on head.

Head longer than wide, HL/HW = 1.09, eyes small but prominent, frons and vertex convex, frontal impressions large and deep; clypeus quadrisetose in male, basal portion even; labrum sex-setose, shallowly emarginate at apical margin; palps slender, subcylindrical, the 3^{rd} maxillary palpomere as long as the 4^{th} which glabrous, the 3^{rd} maxillary palpomere with two short setae at apex; the 2^{nd} labial palpomere slightly longer than the 3^{rd} , bisetose on inner margin, with several additional setae at subapex and apex, the 3^{rd} labial palpomere with a few setae at basal part; ligula small, bisetose at apex; mentum edentate; each of mentum and submentum bisetose (but a third seta on the left of mentum in the holotype), palpiger asetose. Antennae moderately long, extending over base of elytra; pubescent from basal 1/3 of the 4^{th} antennomere, slightly expanded at pubescent portion of the 4^{th} ; the 3^{rd} as long as the 4^{th} .

Pronotum strongly transverse, PW/PL = 1.60, disc quite flat, apical and basal margins well beaded, sides evenly expanded, widest at middle; base wider than apex, bisinuate on base, slightly concave on apical margin; lateral expanded margin well defined, evenly and distinctly reflexed; both transversal impressions distinct, basal foveae small and deep.

Elytra elongate-ovate, EL/EW = 1.75; widest at about middle, sides parallel at middle, basal border complete, apex bisinuate, inner angle broad, but shortly denticulate (Fig. 17); striae moderately deep, intervals convex, subequal in width of odd and even intervals in middle, the 3^{rd} interval with three setiferous pores; the 7^{th} interval normal.

Legs rather slender, hind femora rather slender, with three posterior setae; fore tibiae expanded at apex, with outer angle strongly protrude, outer margin serrate; middle coxae setose, hind coxae smooth and glabrous; middle tibiae quite straight, slightly expanded at apex; hind tibiae slightly expanded at apex, apical spurs long and sharp, the 3rd hind tarsomere slightly longer than the 4th which bilobed at apex; all tarsal claws weakly pectinate.

Prosternal process unbordered at apex.

Male genitalia (Figs 28–30): Median lobe of aedeagus stout, dorsal opening wide and long, base dilated, gently bent ventrally towards apex, which almost pointed in profile; in dorsal view, median lobe distinctly contracted before apical lamella which short and thin, longer than wide.

Female: Unknown.

Remarks. It is very similar to *O. limbourgi*, with the differences mentioned above. **Etymology.** Referred to its polished body.

Distribution. Laos (Fig. 3d).

Orthogonius aberlenci sp. n.

http://zoobank.org/15B791FA-FBB3-4DD9-8349-3BE664F7228D Figs 3e, 8, 18, 31–32

Holotype. Male, label: "Laos: Province de Khammouane, rivière Hin Boun, Ban Nathan, campe de l'Agame, 17°59.773'N / 104°49.395'E, V. 2012, Piège Lumineux, IBCFL, Opération Canopée, H.-P. Aberlenc leg.", in MNHN.



Figures 14–23. Apex of right elytron 14 *O. jianfengling* 15 *O. himalayicus* 16 *O. limbourgi* sp. n. 17 *O. politior* sp. n. 18 *O. aberlenci* sp. n. 19 *O. bellus* sp. n. 20 *O. freyi* 21 *O. duboisi* 22 *O. meghalayaensis* sp. n. 23 *O. wrasei* sp. n.

Paratypes. 1 male & 3 females, in MNHN, SCAU and CIRAD (Montpellier).

Diagnosis. A medium-sized and stout species, with a shiny, smooth, and glabrous body which is brownish in general, and having an edentate mentum, a well bordered prosternal process and setose midcoxae.

Description. Length: 15.0–16.5 mm; width: 6.0–6.5 mm. Habitus as in Fig. 8.

Middle sized, stout, strongly shiny, smooth and glabrous. Head black; palps, pronotum, elytra, the 1st-3rd antennomeres, legs and ventral surface brownish; labrum, clypeus, the 4th to 11th antennomeres dark brown. Head and pronotum impunctate, elytral odd intervals with a few fine and feeble punctures. Microsculptural engraved meshes isodiametric on elytra, indistinct on head and pronotum.

Head as long as wide, eyes moderate and prominent, frons and vertex convex, frontal impressions large and deep, extending posteriorly to the level of anterior supraorbital pores; clypeus bisetose, basal portion decorated by three short, longitudinal furrows which crossing base of clypeus and joining frontal impressions on either sides, and against frons medially; labrum sex-setose, moderately emarginate at apical margin; palps stout, subcylindrical, the 3rd maxillary palpomere slightly shorter than the 4th, glabrous on the 4th, the 3rd with three short setae at apex; the 2nd labial palpomere slightly longer than the 3rd, bisetose on inner margin, with several additional setae at subapex and apex, the 3rd labial palpomere with a few setae at basal part; ligula small, bisetose at apex; mentum edentate; each of mentum and submentum bisetose, setae subequal in length, palpiger asetose. Antennae moderately long, extending over base of elytra; pubescent from basal 1/3 of the 4th, the 1st with a long seta at subapex.

Pronotum strongly transverse, PW/PL = 1.65, disc quite flat, apical and basal margins well beaded, sides evenly expanded, widest at middle; base wider than apex, bisinuate on base, slightly and widely concave on apical margin; lateral expanded margins well defined, narrow, evenly and distinctly reflexed; fore transversal impression faint, hind one distinct, basal foveae small but deep.

Elytra elongate, EL/EW = 1.70-1.75; widest at about middle, almost parallelsided at middle, basal border complete, apex widely and nearly obliquely truncated on outer margin, then reversely truncated nearly the tip, inner angle nearly rectangular, not denticulate (Fig. 18); striae moderately deep, intervals convex, intervals subequal in width in middle, the 3^{rd} interval with three setiferous pores; the 7^{th} interval normal.

Legs moderately long, hind femora moderately expanded, with three posterior setae in male and four or five in female; fore tibiae expanded at apex, with outer angle shortly protruded, outer margin sub-serrate; middle coxae glabrous in holo-type, but with one to three setae in median portion in other specimens, hind coxae smooth and glabrous; middle tibiae not modified in male, quite straight, serrate, gradually expanded towards apex; hind tibiae slightly expanded at apex, apical spurs very long and sharp, the 1st hind tarsomere distinctly longer than the 2nd, the 3rd as long as the 4th which deeply and symmetrically bilobed at apex; all tarsal claws weakly pectinate.

Prosternal process bordered at apex. Abdominal ventrite VII of male complete, with two pairs of setae on either side of apical margin in both male and female.

Male genitalia (Fig. 31–32): Short and stout, dorsal opening wide and long, somewhat expanded medially, gently bisinuate ventrally towards apex, which short and blunt in lateral view; apical lamella short and small, not parallel-sided, wider than long, apex broad.


Figures 24–30. Male genitalia (right lateral view and dorsal view for apical lamella) 24–25 *O. himalayicus* 26–27 *O. limbourgi* sp. n. 28–30 *O. politior* sp. n.

Remarks. It is probably allied to *O. limbourgi* sp. n. as they have similar structure of aedeagus. But it is easily separated from *O. limbourgi* and *O. politior* sp. n. by its short and more convex body, and short apical lamella of the aedeagus.

Etymology. The name of this new species is in honor of the collector, Henri-Pierre Aberlenc (CIRAD, Montpellier), an excellent entomologist and a friend of the second author.

Distribution. Laos (Fig. 3e).

Orthogonius bellus sp. n.

http://zoobank.org/AD1E656E-12F7-4F88-AD5C-ACF59BA56445 Figs 3c 9, 19, 36–37

Holotype. Male, "S. Vietnam, 40 km NW An Khe, Buon Luoi, 620-750 m", "14.10 N / 108.30 E, 28. III-12.IV. 1995, Pacholatko & Dembicky", "Mus. Wien."

Paratype. 1 male, ibid. in NHMV.

Diagnosis. A fairly large species, with a yellowish and slender body which is less depressed, but strongly shiny, smooth and glabrous, having a bordered prosternal process, an edentate mentum and asetose midcoxae.

Description. Length: 18.5 mm; width: 7.0 mm. Habitus as in Fig. 9.

Body elongate, strongly shiny, smooth and glabrous. Yellowish to brown, except head which dark brown. Head and pronotum impunctate, elytral odd intervals with a few punctures. Microsculptural engraved meshes isodiametric on head and elytra, transverse on pronotum.

Head longer than wide, HL/HW = 1.12, eyes small but prominent, frons and vertex convex, frontal impressions deep and long, extending beyond the level of fore supraorbital setae; clypeus bisetose, basal portion of surface uneven; labrum sex-setose, faintly emarginate at apical margin; palps slender, subcylindrical, the 3^{rd} maxillary palpomere as long as the 4^{th} which glabrous; the 3^{rd} maxillary palpomere with two short setae at apex; the 2^{nd} labial palpomere slightly longer than the 3^{rd} , bisetose on inner margin, with several additional setae at subapex and apex; the 3^{rd} labial palpomere with a few setae at basal part; ligula small, bisetose at apex; mentum edentate; each of mentum and submentum bisetose, palpiger asetose. Antennae rather long, extending basal one third of elytra; pubescent from basal one third of the 4^{th} antennomere, slightly expanded at pubescent portions; the 3^{rd} antennomere almost as long as the 4^{th} .

Pronotum strongly transverse, PW/PL = 1.64, disc moderately convex, apical and basal margins well beaded, sides evenly expanded, widest at middle; lateral expanded margin well defined and reflexed, surface uneven; both transversal impressions well marked, basal foveae small and deep.

Elytra elongate-ovate, EL/EW = 1.69; widest at about apical one third of elytra, sides parallel at middle, basal border complete, apex broadly sinuate, inner angle nearly rectangular (Fig. 19); striae deep, intervals distinctly convex, subequal in width with each other at middle, the 3^{rd} interval with three setiferous pores.



Figures 31–37. Male genitalia (right lateral view and dorsal view for apical lamella) 31–32 *O. aberlenci* sp. n. 33–35 *O. bellus* sp. n. 36–37 *O. freyi*.

Legs rather slender, fore tibiae slightly expanded at apex, outer angle distinctly protruding and sharp, outer margin somewhat sub-serrate; middle and hind coxae smooth and glabrous; middle tibiae quite straight, slightly expanded at apex; hind tibiae hardly expanded at apex, apical spurs long and sharp; the 3rd hind tarsomere as long as the 4th which deeply bilobed at apex; claws weakly pectinate.

Prosternal process faintly bordered at apex.

Male genitalia (Figs 36–37): Stout, slightly sinuate at ventral side, dorsal opening wide, apex densely spinulate, apical lamella very short, broad, much wider than long.

Female: Unknown.

Remarks. Probably close to *O. limbourgi* sp. n., but its body less depressed, and hind 3rd tarsomere as long as 4th (versus in *O. limbourgi*).

Etymology. "*Bellus*", a Latin word meaning beautiful, refering to the charmof this species.

Distribution. Southern Vietnam (Fig. 3c).

Orthogonius freyi Tian & Deuve, 2006

Figs 3h, 10, 20, 36-37

Orthogonius freyi Tian & Deuve, 2006: 126.

Length. 17.0 mm; width: 6.5 mm. Habitus as in Fig. 10.

Description. Dark brown to black, body rather slender and depressed, smooth and glabrous, microsculptural engraved meshes isodiametric on head, pronotum and elytra. Head longer than wide, HL/HW = 1.16, eyes rather small; labrum straight at frontal margin, sex-setose, clypeus bisetose; each of mentum and submentum each with a pair of setae. Pronotum strongly transverse, PW/PL = 1.60, widest a little before middle, lateral expanded margins well-marked, almost flat; elytra elongate-ovate, EL/ EW = 1.73, convex, base well-bordered, parallel-sided, striae deep, intervals strongly convex, apex faintly sinuate, inner angle nearly rectangular, distinctly denticulate at tip (Fig. 20); the 3rd interval with two well-marked setiferous pores, median one wanted. Legs moderately long, the 4th hind tarsomere shorter than the 3rd, lobes of 4th tarsal emarginations half long as the tarsomere; tarsal claws weakly pectinate. Prosternal process well-bordered at apex; abdominal ventrite VII not emarginate at apical margin in male. Male genitalia (Figs 36–37) stout and short, apical lamella short, as wide as long at apex, tip almost truncated.

Remarks. Differs from other members of this species group by its labrum straight at front, and 3rd elytral interval with only two setiferous pores.

Material examined. The type series only: 1 male, holotype, "Birmah Momeit", "Doherty" "Frey Coll. 1905.100", "60827" and "*Orthogonius* sp." (By Andrewes); 1 female, a paratype, ibid, in NHML.

Distribution. Myanmar (Fig. 3h).



Figures 38–43. Male genitalia (right lateral view and dorsal view for apical lamella) **38–39** *O. duboisi* **40–41** *O. meghalayaensis* sp. n. **42–43** *O. wrasei* sp. n., right lateral view and ventral view.

Orthogonius duboisi Tian & Deuve, 2006

Figs 3f, 11, 21, 38–39

Orthogonius duboisi Tian & Deuve, 2006: 124

Length. 19.0 mm; width: 7.0 mm. Habitus as in Fig. 11.

Description. Dark brown, shiny, glabrous and smooth, but faint striate on head and pronotum; microsculptural engraved meshes isodiametric. Head slightly longer than wide, HL/HW = 1.10, eyes moderately sized, prominent, labrum slightly emarginate at frontal margin, clypeus bisetose; antennae backwardly exceeding elytral humeri. Pronotum transverse, PW/PL = 1.77, widest a little before middle, lateral expanded margin quite wide, slightly reflexed. Elytra elongate-ovate, rather flat, base well-bordered, parallel-sided medially, apex moderately sinuate, outer angle indistinct, inner angle large and obtuse, not denticulate (Fig. 21); striae deep, intervals slightly convex, subequal in width at middle; the 3^{rd} interval with three setiferous pores. Hind tibial spurs quite short, but sharp; the 4^{th} tarsomere shorter than the 3^{rd} , deeply emarginate at apex, with lobes nearly half as long as joint. The prosternal process bordered at apex. Male genitalia (Figs 38–39): Short and stout, apex broad, the apical lamella very short and broad.

Female: Unknown.

Remarks. It is similar to *O. freyi*, but its inner apical angle of elytra is obtuse and not denticulate at tip (versus in *O. freyi*), and the male aedeagus is stouter.

Material examined. 1 male, the holotype, "CHINE-Yunnan, Tongbiguan, 24°36'N, 97°35'E, 1180 m", "14. VI. 2001, Deuve, Mantilleri, Rougerie & Tian leg.", in SCAU.

Distribution. China (Yunnan).

Orthogonius meghalayaensis sp. n.

http://zoobank.org/7151FBFF-2EDC-44EA-AF90-C54217557D6A Figs 3j, 12, 22, 40–41

Holotype. Male, "NE India: Meghalaya, W. Garo Hills, Nokrek National Park, ca 1000 m", "25.29.06 N / 90.19.05 E, 9-17. V. 1996, leg. Jendek & Sausa", "Mus. Wien."

Paratype. 1 female, ibid, in NHMV.

Diagnosis. A dark brown or black species, having a elongate body, a deeply emarginate labrum, an edentate mentum, a well bordered prosternal process, setose midcoxae and a 6-setose ventrite VII.

Description. Length: 18.5–19.0 mm; width: 6.5 mm. Habitus as in Fig. 12.

Body elongate, shiny. Head and disc of pronotum black to dark brown; elytra, lateral expanded margins of pronotum, mouthparts, antennae, legs and ventral surface reddish dark brown. Female darker than male. Head and pronotum impunctate, elytral odd intervals with a few punctures; head densely and moderately wrinkled and intricate; pronotum transversally striate, elytra smooth. Microsculptural engraved meshes isodiametric on elytra, rather transverse on head and pronotum. Head longer than wide, HL/HW = 1.04, eyes small but prominent, frons and vertex convex, frontal impressions deep; clypeus bisetose, basal portion of surface uneven; labrum sex-setose, deeply emarginate at apical margin; palps slender, subcylindrical, the 3^{rd} maxillary palpomere as long as the 4^{th} which glabrous, the 3^{rd} maxillary palpomere with two short setae at apex; the 2^{nd} labial palpomere slightly longer than the 3^{rd} , bisetose on inner margin, with several additional setae at subapex and apex, the 3^{rd} labial palpomere with a few setae at basal part; ligula small, bisetose at apex; mentum with a median tooth, but not well-defined; each of mentum and submentum bisetose, palpiger asetose. Antennae moderately long, extending over base of elytra; pubescent from basal one third of the 4^{th} antennomere, slightly expanded at pubescent portion; the 3^{rd} antennomere almost as long as the 4^{th} .

Pronotum strongly transverse, PW/PL = 1.60, disc moderately convex, apical and basal margins well beaded, sides evenly expanded, widest at middle; lateral expanded margins well defined, flat and not reflexed, surface uneven; both transversal impressions distinct, basal foveae small and deep.

Elytra elongate ovate, EL/EW = 1.67, widest at about middle, sides parallel at middle, basal border complete, apex broadly sinuate, inner angle broadly acute (Fig. 22); striae shallow, intervals rather flat or slightly convex, subequal in width with each other in middle, the 3^{rd} interval with three setiferous pores; the 7th interval normal.

Legs rather slender, fore tibiae slightly expanded at apex, outer angle distinctly protrude, outer margin somewhat sub-serrate; middle and hind coxae smooth and glabrous; middle tibiae quite straight, slightly expanded at apex; hind tibiae hardly expanded at apex, apical spurs long and sharp, the 3rd hind tarsomere much longer than the 4th which deeply emarginate at apex (lobes half as long as the joint); claws weakly pectinate.

Prosternal process unbordered at apex. Ventrite VII with three pairs of setae on either side of apical margin in both sexes.

Male genitalia (Figs 40–41): Short and stout, nearly straight ventrally, dorsal opening very large, almost extending to base; apical lamella short, contracted towards apex, slightly wider than long.

Remarks. Separable from other congeners by sex-setose on ventrite VII, deeply emarginate labrum at apical margin and dentate mentum.

Etymology. Referred to the type locality.

Distribution. India.

Orthogonius wrasei sp. n.

http://zoobank.org/5A827310-DE68-48D9-B997-B7161BCBA7DC Figs 3g, 13, 23, 42–43

Holotype. Male, "Myanmar (Magway State), nr. Kyeto Pass between Padaukaing-Ngabyin, 850 m, 21.88911 N / 094.41589 E, (secondary forest, lux), 30. VI. 2005, leg. M. Hoffmann & T. Ihle", "Coll. Wrase BERLIN", in CDW.

Paratypes. 5 females, ibid. in CDW, MNHN and SCAU.

Diagnosis. A black (including lateral expanded pronotal margins) and stout species, having nearly rectangular inner apical angles of elytra, an edentate mentum, a well bordered prosernal process and setose midcoxae.

Description. Length: 18.5–19.0 mm; width: 6.5 mm. Habitus as in Fig. 13.

Body elongate, smooth and shiny. Body black, except palps dark brown. Head and pronotum impunctate, elytral odd intervals with small and sparse punctures; head and pronotum obscurely striate. Microsculptural engraved meshes isodiametric on head and elytra, transverse on pronotum.

Head longer than wide, HL/HW = 1.11-1.13, eyes rather small but prominent, frons and vertex convex, frontal impressions deep, extending beyond level of the fore supraorbital setae; clypeus bisetose, surface almost even; labrum sex-setose, slightly emarginate at apical margin; palps slender, subcylindrical, the 3^{rd} maxillary palpomere as long as the 4^{th} which glabrous, the 3^{rd} with two short setae at apex; the 2^{nd} labial palpomere slightly longer than the 3^{rd} , bisetose on inner margin, with several additional setae, the 3^{rd} labial palpomere with a few setae at basal part; ligula small, bisetose at apex; mentum without distinct tooth; each of mentum and submentum bisetose, palpiger asetose. Antennae moderately long, extending over base of elytra; pubescent from basal two fifth of the 4^{th} antennomere, slightly expanded at pubescent portion; the 3^{rd} almost as long as the 4^{th} .

Pronotum strongly transverse, PW/PL = 1.76–1.77, disc moderately convex, apical and basal margins well beaded, sides strongly expanded, widest at middle; lateral expanded margins well defined, flat and not reflexed, surface somewhat striate; fore and hind angles broadly rounded; both transversal impressions well marked, basal foveae small and deep.

Elytra elongate ovate, EL/EW = 1.66–1.68, widest at about middle, parallel-sided at middle, basal border complete, apex broadly and obliquely truncated, inner angle nearly rectangular (Fig. 23); striae deep, intervals strongly convex, subequal in width at middle, the 3rd interval with three setiferous pores.

Legs rather slender, fore tibiae slightly expanded at apex, apical margin sinuate, outer angle distinctly protrude, outer margin sub-serrate; middle coxae setose, hind ones smooth and glabrous; middle tibiae quite straight, slightly expanded at apex; hind tibiae slightly expanded at apex, apical spurs long and sharp, the 3rd hind tarsomere longer than the 4th which deeply emarginate at apex (lobes one third as long as the segment); claws strongly pectinate.

Prosternal process well bordered at apex. Ventrite VII quadrisetose in both male and female;

Male genitalia (Figs 42–43): Stout and robust, shallowly sinuate ventrally, dorsal opening very wide, gently and gradually narrowed towards apex, which blunt in lateral view; apical lamella very short, and very sharp in ventral view.

Remarks. Recognized by its black body and nearly rectangular inner apical angles of elytra.

Etymology. This new species is named in honor of David W. Wrase (Berlin). **Distribution.** Myanmar.

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RESEARCH ARTICLE



Two new species of the genus *Timia* and a redescription of *Timia mongolica* (Diptera, Ulidiidae)

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Abstract

Two new species of the genus *Timia* Wiedemann, 1824 are described and illustrated. *Timia lazebnayae* sp. n. from Uzbekistan has yellow body and is similar to *Timia gobica* Zaitzev, 1982, differing from it only by the entirely yellow flagellomere 1. *Timia shatalkini* sp. n. from Mongolia has dark body and differs from all other dark-colored representatives of the genus by the cell r_{4+5} being completely closed, forming petiole at the wing apex. *Timia mongolica* Zaitsev, 1982 is redescribed and an updated key for yellow-coloured *Timia* is provided.

Keywords

Diptera, new species, redescription, Timia, Ulidiidae

Introduction

Timia Wiedemann, 1824 is a Palaearctic genus which includes 60 described species, commonly found in semi-arid and arid regions (Becker 1906; Hendel 1908; Hennig 1940; Zaitzev 1982; Zaitzev 1984a, b; Kameneva 1996, 2000, 2010; Galinskaya

2011, 2014; Morgulis and Freidberg 2014). While studying the Ulidiidae material from the collection of the Zoological Institute of the Russian Academy of Sciences, Saint-Petersburg (ZISP), the author came across several specimens of flies, belonging to the two new species described below. Examination of the type material of *Timia mongolica* Zaitsev, 1982 revealed several disparities between the specimens and the description of the species (Zaitsev 1982); for example, the body appeared to be yellow and not dark brown. The previously unknown female of *T. mongolica* was found in the collection of ZISP. Therefore, this species is redescribed here and an updated key for the yellow-coloured *Timia* is provided.

Material and methods

Specimens examined were obtained from or deposited in the collections of the following institutions:

ZISP Zoological Institute of the Russian Academy of Sciences, Saint-Petersburg, Russia
ZMUM Zoological Museum of the Lomonosov State University, Moscow, Russia
MNKB Museum für Naturkunde, Leibniz-Institut für Evolutions und Biodiversitätsforschung, Berlin, Germany

Morphological terminology and abbreviations generally follow White et al. (1999). Series of photos were taken directly by the Canon EOS 40D camera with Canon MP-E 65 mm lens and then composed with CombineZM software (Hadley 2007). Measurements are provided in millimetres.

Taxonomy

Timia Wiedemann 1824

Type species. Timia erythrocephala Pallas in Wiedemann, 1824 (by monotypy).

Diagnosis. Yellow or black flies. Frons usually with dents and bumps, shiny or almost shiny, sometimes with white microtrichose areas. Antennal grooves deep, oval, well-separated by wide facial carina. Thorax and abdomen shining or shagreened, sometimes almost matt, sometimes with green metallic shine, often with microtrichose areas. Wing hyaline, in some species with dark cells bc, c, sc and apical spot. Male genitalia: epandrium subovoid; surstylus hook-like, sometimes with marked postero-dorsal lobe; cerci clearly bilobed; phallus long, coiled and partially flattened divided into two parts, with a pair of sclerotized taeniae ending approximately at its mid-length and another pair of taeniae beginning at phallus middle almost reaching phallus apex;

phallus apical half bearing long membranous appendix ("caecum"); distiphallus apex bowed and bearing numerous spurs, and "glans" formed by hooks or lobes surrounding gonopore. Surstylus hook-like, sometimes with marked postero-dorsal lobe. Cerci clearly bilobed. Female terminalia: aculeus moderately long and wide, with short oval cercal unit; three spherical spermathecae with smooth or micropapillose surface. Separation of *Timia* and *Ulidia* Meigen, 1826 is difficult. The characters used so far are mainly as follow: frons smooth (in *Timia*) or dimpled (in *Ulidia*) (with some exceptions), head and thorax microtrichose (in *Timia*) or bare (in *Ulidia*, but some species assigned to *Timia* have shiny head and thorax, and *Ulidia metope* Kameneva, 2010 has frons widely microtrichose) (Chen and Kameneva 2009; Kameneva 2010). In addition, species of the genus *Ulidia* always have entirely black bodies without yellow parts of thorax and abdomen.

Remarks. Adult *Timia*, as well as many other ulidiids, are attracted to various organic residues (decaying plant tissue, animal corpses, excrements). In arid habitats, the surface of organic residues is quickly covered with a dried crust, under which semiliquid substrate is preserved for a relatively long time. The proboscis of *Timia* has longitudinal rows of pointed outgrowths located on the labellum; flies make a hole in a crust with these appendages and then penetrate with long tubular appendage of the proboscis into it, absorbing semi-liquid substrate (Zaitzev 1982).

Key to the yellow-bodied species of the genus *Timia* and *Timia shatalkini* sp. n.

1	Body yellow (Figure 1)2
_	Body black (Figure 2)11
2	Parafacialium wider than flagellomere 1 and twice as wide as antennal groove
	(Figure 10) 3
_	Parafacialium as wide as or narrower than flagellomere 1, and narrower than
	antennal groove (Figure 1)
3	Posteroapical extension of cell bcu more than 1.5 times as long as maxi-
	mum width of cell. Mesonotum shining, without microtrichose areas (Fig-
	ure 10). Female: cercal unit with 1 pair of long basal setae. Male: cerci
	rounded apically
_	Posteroapical extension of cell bcu at most as long as cell width at its middle.
	Mesonotum silvery white microtrichose, with rows of merging shining spots
	around setae (Figure 11). Female: cercal unit with 2 pairs of long setae. Male:
	cerci with obtuse angulate apex
4	Cell r_{4+5} open. Parafacialium almost as wide as flagellomere 1 (Figure 6) 6
_	Cell r _{4.5} completely closed, forming petiole at wing tip. Parafacialium less
	than half as wide as flagellomere 1 (Figure 8)5
5	Flagellomere 1 entirely black (Figure 8)T. gobica Zaitzev, 1982
_	Flagellomere 1 entirely orange (Figure 1) Timia lazebnayae sp. n.

Distance between apices of veins R₄₊₅ and M longer than cross-vein R-M. 6 Scape and pedicel entirely black, contrasting with yellow flagellomere 1 Distance between apices of veins $R_{4,5}$ and M shorter than cross-vein R-M. Scape and pedicel entirely yellow (or brown-yellow, in this case flagellomere 1 black)7 7 Flagellomere 1 and apex of palpus yellow. Distance between apices of veins $R_{4,5}$ and M more than half as long as cross-vein R-M. Male fore femur with moderately long and thin setae (Figures 4, 7, 9).....9 Flagellomere 1 and palpus apex black or dark brown. Distance between apices of veins R_{4.5} and M 0.1 as long as cross-vein R-M. Male fore femur either 8 Frons yellow, with black triangle medially, two times wider and six times longer than ocellar triangle. Thorax with a black pattern. Occiput black medially, narrow yellow laterally (Figure 3). Male fore femur with posteroventral Frons yellow, with black ocellar triangle. Thorax with a light brown pattern. Occiput yellow. Male fore femur with thin setulae, without posteroventral series of short black spines (Figure 5) T. canaliculata Becker, 1906 9 Microtrichose areas on mesonotum forming a medial vitta and spots of various shapes at its sides (Figures 4, 9).....10 Mesonotum entirely silvery white microtrichose, without rows of merging shining spots around setae (Figure 7)T. flaveola Galinskaya, 2011 10Mesonotum with lateral vittae of silver microtrichia extending posterior to wing level. Parafacialium >0.25 times as wide as eye. Posteroapical extension of cell bcu shorter than (0.75 times as long as) width of the cell at its middle. Face >1.4 times wider than high (1.85 times as wide as its height) (Figure 4) Lateral vittae of silver microtrichia not extending posterior to wing level. Parafacialium <0.25 times as wide as eye. Posteroapical extension of cell bcu longer than (1.7 times as long as) width of the cell at its middle. Face <1.4 times wider than high (as wide as high) (Figure 9) Cell r_{445} apically completely closed, forming petiole. Parafacialium about half 11 as wide as flagellomere 1. Thorax with microtrichose area on mesonotum forming a medial vitta; thorax light brown, with dark brown lateral spots (mainly on presutural area of scutum) and medial stripes (Figure 2) Cell $r_{4.5}$ open. Parafacialium width variable. Thorax with or without pattern of microtrichia, but without a medial vitta; thorax coloration variable, predominantly uniformly black or black with yellow scutellum other *Timia* species

Timia lazebnayae sp. n.

http://zoobank.org/498E6DD0-14BF-4AD3-B0FD-17A2C59B0E4B Figure 1

Type material. *Holotype male:* **Uzbekistan:** "70 км СЗ Газли, пески Кызыл Кум, Зайцев, 27.V.1965" [70 km NW Gazli, sands of Kyzyl Kum, Zaitzev leg. 27.V.1965] (ZISP).

Paratype: Uzbekistan: 1 male, same data as holotype (ZISP).

Diagnosis. *Timia lazebnayae* sp. n., *Timia shatalkini* sp. n. and *Timia gobica* differ from all other *Timia* species by cell r_{4+5} completely closed, forming petiole at wing tip and by parafacialium narrow, almost 0.3 times as narrow as first flagellomere. *Timia lazebnayae* sp. n. differing from *Timia shatalkini* sp. n. and *Timia gobica* by first flagellomere entirely yellow. *Timia lazebnayae* sp. n. is characterised by body yellow; cell r_{4+5} apically completely closed, forming petiole at wing tip; parafacialium less than half as wide as flagellomere 1, and narrower than antennal groove; flagellomere 1 entirely orange.

Description.

Male: *Head.* Frons pale yellow, evenly covered with short and thin brown setulae. Frons with band of weak microtrichosity along eye margin. Ocellar triangle shining black. Lateral part of vertex entirely yellow without dark markings. Occiput pale yellow, covered with short and thin black setulae. Medial occipital sclerite yellow; lateral occipital sclerite yellow, with silver-white microtrichosity. Gena yellow, 0.25 times as low as eye. Postgena yellow. Parafacialium yellow, 0.3 times as narrow as antennal grooves, 0.5 times as narrow as flagellomere 1. Eye as high as wide. Lunula pale yellow. Face yellow, without darkened band along the ventral edge. Clypeus yellow. Scape and pedicel yellow. Flagellomere 1 completely yellow, matted, roundish. Arista yellow. Antennal groove yellow, with thin silver-white microtrichosity. Proboscis yellow, slightly darkened distally. Palpus yellow.

Thorax. Yellow, with pale brown lateral spots (mainly on presutural area of scutum) and with medial stripe. Mesonotum with silver-white microtrichosity, lateral spots and medial stripe. Postpronotum slightly microtrichose. Scutellum nearly shiny, yellow, with short black setulae ventrally. Mediotergite dark brown. Pleura yellow, slightly microtrichose on dorsal edge of anepisternum. Pleura covered with sparse short black setulae. Katepisternum with pale brown spot in ventral portion.

Setae. Two small orbital setae (dorsal seta little longer than ventral seta), one divergent ocellar seta, one divergent postocellar seta, one long divergent lateral vertical seta, five divergent postocular setae, one short paravertical seta, and one long convergent medial vertical seta. Three postgenal setulae, one long genal seta. Two groups of supracervical setae over occipital foramen. One postpronotal seta, two notopleural setae, one supra-alar seta, one prescutellar dorsocentral seta, one postalar seta, one intra-alar seta; two scutellar; anepimeral setae absent; one anepisternal seta, one katepisternal seta, anepisternum and katepisternum covered with short black setulae.

Legs. Entirely yellow.



Figure I. *Timia lazebnayae* Galinskaya sp. n., paratype male; **A** habitus, lateral view **B** head and thorax, dorsal view **C** head, lateral view **D** head, anterior view **E** epandrium, dorsal view **F** wing.

Wings. Hyaline. Cell r_{4+5} completely closed, forming petiolate at wing apex. Posteroapical extension of cell bcu 0.2 times as short as width of the cell at its middle. Halter yellow.

Abdomen. Yellow, non-microtrichose.

Male genitalia. Surstylus with anteroventral lobe, curved proximally at right angle, bearing two spines at its medial margin; and with short posteromedial lobe covered

with setulae ventrally. Cerci tapered at apex, separated by narrow slit, covered with long setulae.

Body length, 3.5–3.6 mm. Wing length, 3.1–3.2 mm. Female: unknown. Etymology. The name is in dedication to a good friend V.S. Lazebnaya. Distribution. Uzbekistan.

Timia shatalkini sp. n.

http://zoobank.org/FC9FEE83-97B7-4CBC-8130-F7DF09D5F2E0 Figure 2

Type material. *Holotype male:* Mongolia: "Монголия, Кобдоский аймак, Ур. Елхон, 20 км ЮВ Алтая на Бодончи, Кержнер, 27.VII.1970" [Khovd Province, Elhon, 20 km SE Altai to Bodonchi, 27.VII.1970, I.M. Kerzhner leg.] (ZISP).

Paratype: Mongolia: 1 female, label as in the holotype .

Diagnosis. *Timia shatalkini* sp. n., *Timia lazebnayae* sp. n. and *Timia gobica* differ from all other *Timia* species by cell r_{4+5} completely closed, forming petiole at wing tip and by parafacialium narrow, almost 0.3 times as narrow as flagellomere. *Timia shatalkini* sp. n. differing from *Timia lazebnayae* sp. n. and *Timia gobica* by dark brown body. *Timia shatalkini* sp. n. is characterised by body black; cell r_{4+5} apically completely closed, forming petiole at wing tip; parafacialium about half as narrow as flagellomere 1. Thorax with microtrichose area on mesonotum forming a medial vitta; thorax light brown, with dark brown lateral spots (mainly on presutural area of scutum) and medial stripes.

Description.

Male: *Head.* Frons pale brown dorsally, dark brown ventrally, evenly covered with short and thin brown setulae. Frons with band of weak microtrichosity along eye margin. Ocellar triangle shining black. Lateral part of vertex brown. Occiput brown, covered with short and thin black setulae. Medial occipital sclerite brown; lateral occipital sclerite brown, with band of weak microtrichosity along eye margin. Gena 0.25 times as low as eye, brown, with yellow band along eye margin. Postgena brown. Parafacialium yellow, 0.3 times as narrow as antennal groove, 0.5 times as narrow as flagellomere 1. Eye as high as wide. Lunula pale yellow. Face yellow, with darkened band along the ventral edge. Clypeus yellow. Scape and pedicel light brown. Flagellomere 1 light brown proximally, dark brown distally, matt, roundish. Arista light brown. Antennal groove yellow, slightly darkened. Proboscis dark brown. Palpus dark brown.

Thorax. Pale brown, with dark brown lateral spots (mainly on presutural area of scutum) and medial stripe. Mesonotum with silver-white microtrichose medial stripe. Postpronotum with silver-white microtrichosity. Scutellum subshining light brown, with short black setulae over dorsal surface. Mediotergite dark brown. Pleura dark brown, slightly microtrichose on dorsal edge of anepisternum. Pleura covered with sparse short black setulae.



Figure 2. *Timia shatalkini* Galinskaya, sp. n., paratype female; **A** habitus, lateral view **B** head and thorax, dorsal view **C** head, anterior view **D** head, lateral view **E** epandrium of male, dorsal view **F** wing.

Setae. Four small orbital setae (dorsal orbital seta little longer than other ones), one divergent ocellar seta, one divergent postocellar seta, one long convergent medial vertical seta, five divergent postocular setae, one short paravertical seta, and one long

divergent lateral vertical seta. Three genal setulae, five postgenal setulae, one long genal seta. Two groups of supracervical setae over occipital foramen.

One postpronotal seta, two notopleural setae, one supra-alar seta, one prescutellar dorsocentral seta, one postalar seta, one intra-alar seta; two scutellar; anepimeral setae absent; one anepisternal, one katepisternal seta, also anepisternum and katepisternum covered with short black setulae.

Legs. Coxa, trochanter and femur dark brown, tibia and tarsus yellow.

Wings. Hyaline. Cell r₄₊₅ completely closed, forming petiolate at wing apex. Posteroapical extension of cell bcu 0.4 times as long as the width of cell at its middle. Halter yellow. *Abdomen*. Dark brown, non-microtrichose.

Male genitalia. Surstylus with anteroventral lobe curved proximally at right angle, bears two spines at its medial margin; and with short posteromedial lobe covered with setulae on its ventral side. Cerci tapered apically, separated by narrow slit, covered with long setulae.

Body length 3.1 mm. Wing length 2.3 mm.

Female: similar to male, except genital structures.

Body length 5.1 mm. Wing length 3.2 mm.

Etymology. Named in honor of Dr. A.I. Shatalkin, supervisor of my PhD thesis. **Distribution.** Mongolia.

Timia mongolica Zaitzev, 1982 Figure 3

Type material. *Holotype male:* **Mongolia:** "Монголия, Баян-Хонгорский аймак, 30 км ССВ родн. Шара-Хулсны-Булак, Зайцев, 4.IX.1970" Bayankhongor Province, 30 km NNE spring Shara Hulsny-Bulak, 4.IX.1970, Zaitzev leg. (ZISP).

Paratype: Mongolia: 1 male, label as in the holotype (ZISP).

Additional material: Mongolia: 3 males, 1 female, label as in the holotype (ZISP). Redescription.

Male: *Head.* Frons yellow, with large black triangle medially, without microtrichosity, evenly covered with short and thin black setulae. Ocellar triangle shining black. Lateral part of vertex entirely yellow without dark marks. Occiput yellow, medially black, slightly microtrichose close to eye margin, covered with short and thin black setulae. Gena yellow, 0.4 times as low as eye. Postgena yellow. Parafacialium yellow, 0.5 times as narrow as antennal grooves and as wide as flagellomere 1. Eye 1.3 times as high as wide. Lunula yellow. Face yellow, without narrow darkened band along the ventral edge. Clypeus pale yellow, darkened along ventral edge. Scape and pedicel light brown. Flagellomere 1 brown at base, almost black at apex, matted. Arista light brown. Antennal groove yellow, with thin silver-white microtrichosity. Proboscis brown. Palpus brown at base, black at apex.

Thorax. Yellow, with black pattern. Mesonotum with silver-white microtrichose medial stripe. Postpronotum with silver-white microtrichosity. Scutellum subshining



Figure 3. *Timia mongolica* Zaitzev, 1982, paratype male **A** habitus, lateral view **B** head and thorax, dorsal view **C** head, anterior view **D** epandrium, dorsal view.

yellow, faintly sculptured, with short black setulae over its ventral surface. Mediotergite yellow, with brown spot at the middle. Pleura yellow.

Setae. Four small orbital setae (dorsal orbital seta little longer than other ones), one divergent ocellar seta, one divergent postocellar seta, one long divergent lateral vertical seta, four divergent postocular setae, one short paravertical seta and one long convergent lateral vertical seta. Four genal setulae, eight postgenal setulae, one long genal seta. Two groups of supracervical setae over occipital foramen.

One postpronotal seta, two notopleural setae, one supra-alar seta, one prescutellar dorsocentral seta, one postalar seta, one intra-alar setae, two scutellar; anepimeral setae absent; one anepisternal, one katepisternal setae, also anepisternum and katepisternum covered with short black setulae.

Legs. Yellow. Tarsus, mid- and hindcoxae darkened to pale brown.

Wings. Hyaline, with yellowish apex. Cell r_{4+5} narrowly opened. Apices of veins R_{4+5} and M almost in contact. Posteroapical extension of cell bcu 0.3 times as short as the width of cell at its middle. Halter yellow.

Abdomen evenly short and thin, black, setulose. All tergites laterally yellow, brown at middle, syntergite 1+2 and tergite 3 completely brown. Sternites yellow.

Male genitalia. Surstylus with anteroventral lobe curved proximally at right angle, bears two spines at its medial margin; and with short posteromedial lobe covered with setulae on its ventral side. Cerci tapered at top, separated by narrow slit, covered with long setulae.

Body length, 4.7–5.5 mm. Wing length, 3.5–4.5 mm. **Female:** similar to male, except for genital structures. Body length, 5.0–7.0 mm. Wing length, 3.5–4.8 mm. **Distribution.** Mongolia.

Additional material examined

Timia canaliculata Becker, 1906 Figure 5

Type material. *Lectotype male:* China: "syntypus, Бугась у Хами, на ЮВ от Тянь-Шаня, РобКозлов, 25.VIII.1895" [Bugaz near Hami, to the SE from Tien Shan, 25.VIII.1895, Roborowsky and Kozlov leg.] (ZISP).

Paralectotypes: China: 3 males, 1 female, 25.VIII.1895, label as in lectotype; 1 male 28.VIII.1895, 1 female 20.VIII.1895, labels as in lectotype (ZISP); 4 males, 1 female, label as in lectotype (MNKB).

Additional material: Mongolia: 1 female, Mongolia, Ömnögovi Province, south coast of Buir Lake, 17.VII.970, I.M. Kerzhner leg.; 3 males, 1 female, Mongolia, Khovd Province, Elhon, 20 km SE Altai to Bodonchi, July 27, 1970, I.M. Kerzhner leg. (ZISP).



Figure 4. *Timia berlandi* (Seguy, 1953), holotype male **A** habitus, lateral view **B** head, lateral view **C** thorax, lateral view **D** label **E** thorax, dorsal view **F** wing (after Kameneva 2010, with permission).



Figure 5. *Timia canaliculata* Becker, 1906, lectotype male **A** habitus, lateral view **B** head and thorax, dorsal view **C** head, lateral view **D** head, anterior view **E** wing.

Timia dimidiata Becker, 1906

Figure 6

Type material. *Holotype male:* China: "50812." "Kaschgar, V.1903", "dimidiata Beck." [original Becker's handwritten labels], "Typus" [red printed label] (MNKB).

Additional material: Iran:1 female, from Gurmuk to W and NW Sistan (Expedition to Persia, 1898), 5–20.IX.1898, Zarudny leg.; 2 males, 1 female, Senetang (Expedition to Persia, 1898), 13–17.V.1898, Zarudny leg. (ZISP); Kazakhstan: 1 male, Karakul-Sarah Tugay, Syr Darya, South Kazakhstan Province, 18.V.1898, Heyer leg.; 9 females, 3 males, SE Muyunkum, 110 km NW Jambyl, Zone in front of sands, on the *Alhagi* (camel thorn), 2.VII.1963, Sugonyaev leg. (ZISP); **Tajikistan:** 2 females, neighborhood of Gandzhin, NW Qurghonteppa, 12.V.1961, I.M. Kerzhner leg. (ZISP); **Turkmenistan:** 1 male, Karakul, 65 km. N of Ashgabat, 19.IV.1963, Ponomareva leg.; 3 males, lake Topyatan 15 km NNE Yashhan, Uzboy, 18–19.V.1987, Vereshchagina leg.; 1 male, 12 km SE Tejen, 24.V.1964, Ponomareva leg.; 2 females, 20 km N Kizil Arvat, 3.VI.1952, Shteinberg leg.; 1 male, Merv, 15.VI.1930, V. Popov



Figure 6. *Timia dimidiata* Becker, 1906, male **A** habitus, lateral view **B** head and thorax, dorsal view **C** wing **D** head, anterior view.

leg.; 1 male, 1 female, station Akhcha-Kuyma, 5.VII.1934, V. Popov leg.; 2 females, Karadegish, valley of. the Atrek river, 21.VIII.1932, Ushinsky leg.; 1 specimen without abdomen, Arman Saad-Kizil Arvat, Transcaspian region, 1896, Anger leg.; 1 female, neighborhood of Bugdali, SW Turkmenistan, 6.VII.973, Nartshuk leg.;1 male, 28 km SW Kumdag, Turkmenistan, saline, 5.VI.1973, Nartshuk leg. (ZISP); **Uzbekistan:**1 female, Buchara, Mer. occ., Yargak, pr. Chatyrtshy, 20.VI.1928, L. Zimin leg.; 11 males, 8 females, Kamak, Kattakurgan, near Samarkand, 29.VI.1929, L. Zimin leg.; 1 male, the same place, 10.VII.1929, L. Zimin leg.; 5 males, 4 females, the same place, 1.VII.1932, L. Zimin leg.; 1 male, 2 females, 100 km ENE Taxtakoʻpir, Uzbekistan, O.G. Ovtshinnikova leg., 14.VI.1987; 5 males, 2 females, 32 km NNE Tashkömür, Uzbekistan, O.G. Ovtshinnikova leg., 12.VI.1987 (ZISP); **Armenia:**1 female, Arazdayan, 8.VI.1956, Zimina leg. (ZMUM).

Timia flaveola Galinskaya, 2011

Figure 7

Туре material. *Holotype female:* Turkmenistan: "Репетек, личинки в корнях Convolvulus. Лет – конец июня, собр. Каплин, 27.IV.1980" [Repetek, larvae on the roots of *Convolvulus* (Convolvulaceae), hatching in the late June, 27.IV.1980 Kaplin leg.] (ZISP). Paratypes: 5 females, label as in holotype; 1 female, Turkmenistan: "Репетек, Туркм. саксаульник близ усадьбы, Стальмакова, 21.VI.1947" [Repetek (*Haloxylon* near the estate), 21.VI.1947 Stal'makova leg.] (ZISP).

Timia gobica Zaitzev, 1982

Figure 8

Type material. *Holotype male:* Mongolia: "Монголия, Южно-Гобийский аймак, 20 км CB Булгана, песчаная пустыня с саксаулом, Кандыбина, 10.VII. 1971" [Ömnögovi Province, 20 km NE Bulgan, sand desert, on *Alhagi*, 10.VII.1971, Kandybina leg.] (ZISP).

Additional material: Kazakhstan: 4 males, 60 km. NW Dzhingilda, Kyzyl Kum, 24.V.1965, Zaitzev leg. (ZISP); Mongolia: 4 males, 8 females, Ömnögovi Province, station Bulgan, on *Alhagi*, 20 km NE Bulgan, 22.VI.1971, Kandybina leg.; 1 female, Südgobi Province, 100 km W v. Grenzposten Ovot Chuural, 1250 m; Nr. 835, 23.VI.1967, exp. Dr. Z. Kaszab; 2 males, 1 female, Ömnögovi Province, 20 km NE Bulgan (*Haloxylon ammodendron*), 2.VII.1971; 4 females, Ömnögovi Province, 20 km NE Bulgan (sandy desert with the *Alhagi*), 19.VII.1971, Kandybina leg.; 1 female, Govi-Altai Province, spring Haichi-Bulak, 60 km. SE Bugat, 19.VII.1970, Yemelyanov leg.; 2 males, Khovd Province, Elhon, 20 km SE Altai to Bodonchi, 27.VII.1970, Nartshuk leg.; 2 females, Ömnögovi Province, Dzemgin Gobi, 25 km. SSW Haylastyn-Khuduk, I.M. Kerzhner leg.(ZISP); **Uzbekistan:** 1 male, Khiva, Ravat, 29.VII.1927 L.Zimin leg. (ZISP).



Figure 7. *Timia flaveola* Galinskaya, 2011, paratype male **A** habitus, lateral view **B** head, anterior view **C** head and thorax, dorsal view **D** wing.



Figure 8. *Timia gobica* Zaitzev, 1982, male **A** habitus, lateral view **B** head and thorax, dorsal view **C** head, anterior view **D** wing.

Timia orientalis Zaitzev, 1982

Figure 9

Type material. *Holotype male:* **Mongolia:** "Монголия, Южно-Гобийский аймак, Бордзон-Гоби, 80 км. ЮЮВ Номгона, Зайцев, 5–8.VIII.1967" [Ömnögovi Province, Bordzon Gobi, 80 km. SSE Nomgon, 5–8.VIII.1967, V.Zaitzev leg.] (ZISP).

Paratypes: 8 females, 3 males: **Mongolia:** "Монголия, Южно-Гобийский аймак, Бордзон-Гоби, 80 км. ЮЮВ Номгона, Зайцев, 5–8.VIII.1967" [Ömnögovi Province, Bordzon Gobi, 80 km. SSE Nomgon, 5–8.VIII.1967, Emelianov leg.] (ZISP). **China:** 3 females, "Баинхудук; с. Алашань, Козлов, 20.V.1909" [Bainhuduk, north Alashan Plateau, Gobi Desert, 20.V.1909, Kozlov leg.] (ZISP).

Additional material: China: 1 male, Etszin-gol, north Alashan Plateau, Gobi Desert, 18.VI.1909, Kozlov leg. (ZISP); Mongolia: 1 female, Ömnögovi Province, Bordzon Gobi, 80 km. SSE Nomgon (sands), 5–8.VIII.1967, Kerzhner leg.; 8 females, 3 males, Ömnögovi Province, Bordzon Gobi, 80 km. SSE Nomgon (sands), 5–8.VIII.1967, Yemelyanov leg. (ZISP).

Timia testacea Portschinsky, 1891

Figure 10

Туре material. Lectotype female: "syntypus, Опр. Порчинский, 1892" [syntypus, 1892, Portshinsky leg.] (ZISP). Paralectotype: 1 female, label as in lectotype (ZISP). **China:** 1 male, "Kaschgar, V.1903, 50811, Typus, Typus von *T. mellina* Becker" [Kashgar, V.1903, 50811, Type, Type of *T. mellina* Becker] (MNKB).

Additional material: Kazakhstan: 1 male, Karauzyak, Kyzylorda Province, 15.VI.1916, N. Pulikovskaya leg.; 6 males, 1 female, Tash-Suat, boundary between Kyzylorda and Shymkent Province, 24.V.1896, Heyer leg.; 1 male, Kyzylorda, 17.VI.1926, Ruzaev leg.; 1 male, Kyzylorda, 27.VII.1926, Ruzaev leg.; 1 female, Almaty Province, Sharyn River, Sortogoi, Lehr leg.(ZISP); 1 male, Karachingil, estuary of Syr Darya, 29.VI.1900, L.S. Berg leg.; 2 males, Kazaly, on Tamarix, 4.VII.1900, L.S. Berg leg. (ZMUM); Tajikistan: 2 males, 1 female, Isfara, IV–VI.1927 (ZISP); Uzbekistan: 2 males, station Fedchenko, 1926, L. Zimin leg.; 1 male, Khiva, VII.1929, Gerasimov leg.; 1 male, Pahtalyk-Kul, 15.VI.1925; 1 male, NO Yazyavan, Sary su River, Andijan Province, 22.V.1961, Sugonyaev leg.; 2 females, 1 male, near Tashkent (on *Glycyrrhiza* glabra), 1.VII.1963, Valieva leg. (ZISP); Kyrgyzstan: 1 female, Gulcha, N.N. Filippov leg. (ZMUM); Turkmenistan:1 female, Charshangu, 30 km from Kelif, Amu Darya, Petrischeva leg., 7.VI.934; 1 female, Ispas, 70 km NW Türkmenabat, Nartshuk leg. (ZISP); China: 1 female, Gobi desert, Taklamakan, Hauser leg., 1900, 54535, Timia testacea Portsch. V.Korneyev det. 1999; 1 female, 1 male, Gobi desert, Taklamakan, Hauser leg., 1900, 54535; 1 female, Gobi desert, Taklamakan, Hauser leg., 1900, 54535 Timia testacea Portsch. (MNKB).



Figure 9. *Timia orientalis* Zaitzev, 1982, male **A** habitus, lateral view **B** head and thorax, dorsal view **C** head, anterior view **D** wing.

Timia zaitzevi Galinskaya, 2011

Figure 11

Туре material. Holotype male: **Kazakhstan:** "Ю.-З. Кызыл-Кумы, Ю. Казахст. обл., Лер, 20.V.1960" [SW Kyzyl Kum, 20.V.1960, Lehr leg.] (ZISP). Paratypes: 12 males, 3 females, label as in holotype (ZISP).



Figure 10. *Timia testacea Portschinsky*, 1891, male **A** habitus, lateral view **B** head, lateral view **C** head and thorax, dorsal view **D** head, anterior view **E** wing.

Additional material: Kazakhstan: 1 female, Turkestan station, Kazakhstan, Lukyanovich leg., 30.V.936; 3 females, SW. Kyzyl Kum, S. Kazakhstan Region, Lehr



Figure 11. *Timia zaitzevi* Galinskaya 2011, paratype male **A** habitus, lateral view **B** head and thorax, dorsal view **C** head, anterior view **D** wing.

leg., 20.V.960; **China:** 1 female, sand desert near River Ili, 40 km from Lake Chianka, Poyarkov leg.; **Tajikistan:** 1 female, right bank of Amu Darya, near Aivadj, Shaartuz Region, Tajik SSR, 4.VI.1975, Volkovich leg. (ZISP).

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RESEARCH ARTICLE



Review of Pseudacrobasis Roesler, 1975 from China (Lepidoptera, Pyralidae, Phycitinae)

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Abstract

The genus *Pseudacrobasis* is reviewed for China. *Pseudacrobasis dilatata* **sp. n.** is described as new and compared with *P. tergestella* (Ragonot, 1901). Images of adults and illustrations of genital structures are provided.

Keywords

China, Lepidoptera, new species, Pyralidae, Phycitinae, Pseudacrobasis

Introduction

Pseudacrobasis is a monotypic genus established by Roesler in 1975 for the type species *Pseudacrobasis nankingella* Roesler, 1975 from Nanjing, China. It is widely distributed in China, Korea, Japan, and south of Russian Far East (Roesler 1975; Inoue 1982; Song and He 1997; Bae 2004; Bae, Byun and Paek 2008; Kirpichnikova 1999; Ren and Li 2009, 2012; Yamanaka 2013; Ren 2014), and its discovery in Europe was once

thought to be of an alien species (Asselbergs 1998, 2002; Billi 2010; Asselbergs 2002). In 2014, Vives unveiled *Pseudacrobasis nankingella* was a junior synonym of *Psorosa tergestella* (Ragonot, 1901), and transferred *tergestella* (Ragonot, 1901) from *Psorosa* Zeller, 1848 to *Pseudacrobasis* Roesler, 1975, which eliminated "the wrong identification leading to the wrong conclusion on a putatively 'invasive' species". Scalercio (2015) elaborated its biology, ecology, and distribution for the first time.

It has been more than 40 years since the genus establishment, and since then the type species *Pseudacrobasis tergestella* (Ragonot, 1901) is known only. Here, a second species *P. dilatata* sp. n. is described, based on specimens collected from different localities in China, and it is compared with *P. tergestella* (Ragonot, 1901).

Material and methods

Genitalia dissections were carried out following the methods introduced by Li (2002). Photographs of adults and venation were taken with a Leica M205A, and photographs of genitalia and details of the head were taken with a Leica DM750, using Leica Application Suite 4.6 software to capture images. The type specimens are deposited in the Insect Collection of Nankai University, Tianjin, China.

Taxonomy

Pseudacrobasis Roesler, 1975

Pseudacrobasis Roesler, 1975: 100.

Type species. Pseudacrobasis tergestella (Ragonot, 1901).

Diagnostic characters. *Pseudacrobasis* is characterized by the combination of the following characters: the male antennal scape with a distal scale projection on the inner side, the several basal flagellomeres slightly incurved, forming a shallow sinus containing a smaller scale tuft, the first and several other flagellomeres beyond the sinus bearing a small spine dorsally (Figs 1c, 2c), the ventral surface of the flagellum bearing dense, elongate cilia approximately as long as width of the flagellum, the female antenna simple and weakly pubescent; the labial palpus upturned just beyond the vertex (Figs 1a, 1b, 2a, 2b); the forewing having a fuscous patch on the inner side of the antemedial line and a tuft of scales near the fuscous spot, R_{3+4} and R_5 stalked in basal 2/3, M_2 and M_3 long-stalked (Fig. 1d) or fused (Fig. 2d), M_{2+3} shortly stalked with CuA₁; in the male genitalia, the apical process of the gnathos tapered, the separated transtilla strongly sclerotized, the valva with a small fingerlike clasper at base, the U-shaped juxta with a pair of finger-like lateral lobes, and the phallus with many slightly sclerotized crimples and microtrichia (Figs 3, 4); in the female genitalia, both
apophyses anteriores and posteriores of medium length, the former slightly shorter than the latter, the antrum weak-sclerotized or not sclerotized, the membranous ductus bursae with many spinules near the junction with the corpus bursae, the ovate membranous corpus bursae shorter than the ductus bursae, the signum developed as a small, rounded, granulate plate, and the ductus seminalis arising from the corpus bursae posteriorly (Figs 5, 6).

This genus is similar to *Caradjaria* Roesler, 1975 in appearance, but can be distinguished from the latter by the following characters: the male antennal scape with an angular scale process at its inner terminal, which disappears once the scales are removed; the forewing with M_2 and M_3 very short-stalked; the apical process of the gnathos tapered, the transtilla separated, and the phallus with sclerotized wrinking and a few minute and weak scobinations in the male genitalia; and the membranous corpus bursae in the female genitalia. In the genus *Caradjaria*, the male antennal scape is enlarged at its inner terminal to form a thorn-like process; M_2 and M_3 of the forewing are stalked approximately half of their length; the apical process of the gnathos is enlarged in distal part, the transtilla is connected, and the phallus has small dense spines in the male genitalia; the corpus bursae of the female genitalia is weakly sclerotized in its posterior half.

Pseudacrobasis tergestella (Ragonot, 1901)

Figs 2, 4, 6

Psorosa tergestella Ragonot, 1901: 107–108. TL: Italy (Trieste). *Pseudacrobasis nankingella* Roesler, 1975: 100. TL: China (Jiangsu). *Pseudacrobasis tergestella* (Ragonot, 1901): Vives, 2014: 401.

Diagnosis. Adults (Fig. 2) with wingspan 13.5–18.0 mm. *Pseudacrobasis tergestella* is characterized by the large uncus narrowed from broad base to 3/5, mushroom-like in the distal 2/5; the transtilla distally produced to a stout digitate dorsal process and a curved slender ventral process in the male genitalia (Fig. 4); and by the posterior margin of the eighth tergite deeply concave, U-shaped and with a sclerotized semicircular decoration at middle of anterior 1/3 in the female genitalia (Fig. 6).

Distribution. China (Fujian, Gansu, Guangdong, Guangxi, Guizhou, Hainan, Henan, Hubei, Hunan, Liaoning, Jiangsu, Jiangxi, Jilin, Shaanxi, Shandong, Shanghai, Sichuan, Yunnan, Zhejiang, Taiwan), Korea, Japan, south of Russian Far East, France, Portugal, Italy.

Notes. *Pseudacrobasis tergestella* (Ragonot, 1901) is widely distributed in China. Its identification in this study is based on the examination of 88 male and 67 female specimens. Scalercio (2015) pointed out "the currently known [...], the distribution of *P. tergestella* is quite unique with occurrences in the far east and in the far west of the Palearctic region, no records are currently available for Central Asia and East Europe, where suitable habitats are present". We believe *P. tergestella* is likely to distribute the Central Asia and East Europe region with the depth of the investigation.

Pseudacrobasis dilatata sp. n.

http://zoobank.org/AD43828F-F38B-4D24-929D-4F9EC463A172 Figs 1, 3, 5

Diagnosis. This new species can be distinguished from its allied species *P. tergestella* by the following characters: M_2 and M_3 of the hindwing stalked for approximately 3/5 of their lengths; in the male genitalia by the subtriangular uncus gradually narrowed to truncate apex, the transtilla with its distal part developed into two small horns of nearly equal sizes; in the female genitalia by the posterior margin of the eighth tergite only slightly concave and lacking decoration. In *P. tergestella* (Figs 2, 4, 6), M_2 and M_3 of the hindwing are completely fused (Fig. 2d); in the male genitalia, the uncus is mushroom-like in distal 2/5, the distal part of the transtilla is developed as one large and one small horns (Fig. 4); in the female genitalia, the posterior margin of the eighth tergite is deeply concave and U-shaped, and has a sclerotized semicircular mark in the middle of the anterior 1/3 (Fig. 6).

Description. Adult (Fig. 1): Wingspan 14.0–19.0 mm. Head (Fig. 1a) greyish brown. Antenna (Fig. 1c) with scape greyish brown dorsally, greyish white ventrally, flagellum greyish brown. Labial palpus (Fig. 1b) with first segment greyish white, second and third segments about equal length, greyish brown. Patagium, tegula and thorax greyish brown. Forewing greyish brown, densely dusted with white; basal 1/4 greyish white; antemedial line brown, curving obliquely from basal 1/4 of costa to 2/5 of termen, bordered inwardly by a rounded-triangular fuscous patch, edged with a black ridge of raised scales on inner side, edged inwardly with white; large, triangular, greyish white blotch between antemedial and postmedial lines, reaching near dorsum; discal spots brownish fuscous, rounded, distinct and separated; postmedial line grayish white, sinuate, concave at M_1 and CuA_2 , edged with brown; terminal line black; cilia greyish white basally, light brown distally. Hindwing greyish white, with ten veins, M_2 and M_3 stalked for about 3/5 of their length; termen and cilia light gray. Legs brownish fuscous dusted with greyish white, tarsomeres white at end. Abdomen greyish brown, each segment yellowish brown at end.

Male genitalia (Fig. 3). Uncus subtriangular, broad in basal half, distinctly narrowed at middle, then gradually narrowed to truncate apex. Gnathos with apical process triangular, about 2/5 length of uncus, shorter than lateral arm; lateral arm widely banded, slightly narrowed distally. Transtilla with basal 2/3 narrowed, clubbed, distal 1/3 dilated, stout; apex concave medially, forming a small horn-shaped dorso- and ventro-apical process of about equal size. Valva three times as long as wide, distal 3/5 densely setose, broader than basal 2/5; clasper placed near costa at basal 1/5 of valva, small, fingerlike; costa a narrowed club, extending to end of valva; sacculus fusiform, about 1/3 length of valva, bearing long stouter setae. Juxta nearly quadrate, slightly concave at middle of posterior margin; lateral lobe finger-like, as long as apical process of gnathos, with sparse setae distally. Vinculum U-shaped, moderately long with transverse posterior margin. Phallus cylindrical, about same length as valva, with sclerotized crimples, granulate distally.



Figures 1–2. Adult of *Pseudacrobasis* species: **I** *P. dilatata* sp. n. (**Ia** head **Ib** labial palpus, slide No. YLL15173 **Ic** Antenna, slide No. YLL15173 **Id** wing, slide No.YLL15171w) **2** *P. tergestella* (**2a** head **2b** labial palpus, slide No. YLL15175 **2c** Antenna, slide No. YLL15175 **2d** wing, slide No. LJY10581w). Scale bars: 2.0 mm.



Figures 3-4. Male genitalia structures of *Pseudacrobasis* species: 3 *P. dilatata* sp. n. (3a culcita), slide No. YLL15176 4 *P. tergestella* (4a culcita), slide No. RYD04497. Scale bars: 0.5 mm.

Female genitalia (Fig. 5). Anal papillae subtriangular, narrowed in distal 1/4, rounded at apex. Eighth abdominal segment collar-shaped, approximately 2.5 times wider than long, with tergite arched at middle of anterior margin, slightly concave



Figures 5-6. Female genitalia structures of *Pseudacrobasis* species: 5 *P. dilatata* sp. n., slide No. LHX14109 6 *P. tergestella*, slide No. YLL15177. Scale bars: 0.5 mm.

on posterior margin. Apophyses posteriores slightly longer than apophyses anteriores. Antrum oval, weak-sclerotized. Ductus bursae with posterior 3/5 smooth, moderate in width, anterior 2/5 gradually broadened, with dense spinules from anterior 1/5 to

2/5. Corpus bursae ovate, about 3/4 length of ductus bursae, punctate near signum; signum at posterior 1/5, consisting of concentrically arranged minute scobination, shallowly concaved in middle. Ductus seminalis attached to posterior margin of corpus bursae.

Material examined. Holotype δ , China: Shaanxi, Danfeng, Tieyupu, (33.63°N, 110.53°E; elevation 680 m), 28 May 1994, leg. Jin Zhou. Paratypes: 1 \mathcal{Q} , Gansu, Wenxian, Bifenggou, (32.95°N 104.67°E; elevation 860 m), 10 July 2005, leg. Hai-Li Yu; 1 Å, Guizhou, Chishui, Suoluo, (28.44°N, 106.03°E; elevation 390 m), 27 May 2000, leg. Yan-Li Du; $4 \bigcirc \bigcirc$, Guizhou, Xishui, Linjiang, (28.21°N, 106.18°E; elevation 500 m), 3 June 2000, leg. Yan-Li Du; 1 3, Guizhou, Fanjingshan, Heiwan, (27.94°N, 108.61°E; elevation 530 m), 2 June 2002, leg. Xin-Pu Wang; 3 33, Guizhou, Daozhen, Dashahe, (28.87°N, 107.61°E; elevation 600 m), Xiannvdong, 28 May 2004, leg. Shu-Lian Hao; 1 3, Guizhou, Daozhen, Dashahe, (28.87°N, 107.61°E; elevation 600 m), Xiannvdong, 17 August 2004, leg. Yun-Li Xiao; 2 33, Guizhou, Daozhen, (28.87°N, 107.61°E; elevation 1300 m), Chengjiashan, 19 August 2004, leg. Yun-Li Xiao; 1 👌, Hebei, Jingxing, Mt. Xiantai, (38.12°N, 113.84°E; elevation 100 m), 23 July 2000, leg. Hai-Li Yu; 4 33, Henan, Huixian, Baligou, (35.59°N, 114.00°E; elevation 780 m), 12 July 2002, leg. Xin-Pu Wang; 3 33, Henan, Huixian, Guanshan, (35.50°N, 113.59°E; elevation 550 m), 25-26 July 2006, leg. Deng-Hui Kuang, Hui Zhen; 2 33, Henan, Jiyuan, Wangwushan, (35.15°N, 112.28°E; elevation 1100 m), 30 July 2006, leg. Deng-Hui Kuang, Hui Zhen; 2 ්ථ, Henan, Yiyang, Huaguoshan, (34.34°N, 111.89°E; elevation 1000 m), 1 August 2006, leg. Deng-Hui Kuang, Hui Zhen; $4 \sqrt[3]{3}$, $1 \sqrt{2}$, Hubei, Shennongjia, Bajiaomiao, $(31.76^{\circ}N, 110.57^{\circ}E;$ elevation 1100 m), 19 July 2003, leg. Shu-Lian Hao; 1 $^{\circ}$, Hubei, Shennongjia, (31.34°N, 110.57°E; elevation 1700 m), Wenquan, 21 July 2003, leg. Shu-Lian Hao; 1 👌, Hubei, Shennongjia, Songbaizhen, (31.75°N, 110.66°E; elevation 1200-1400 m), 17 July 2003, leg. Shu-Lian Hao; 1 3, Qinghai, Xunhua, Mengda, (35.83°N, 102.69°E; elevation 2240 m), 15 July 1995, leg. Hou-Hun Li, Shu-Xia Wang. 2 32, 3 99, Shaanxi, Yangling, (34.27°N, 108.08°E; elevation 450 m), 3–11 June 1985, leg. Hou-Hun Li; $4 \stackrel{\circ}{\rightarrow} \stackrel{\circ}{\rightarrow}, 6 \stackrel{\circ}{\rightarrow} \stackrel{\circ}{\rightarrow}$, same data as holotype; $1 \stackrel{\circ}{\rightarrow}$, Shaanxi, Baihe, Qianpo, (32.81°N, 110.11°E; elevation 200 m), 16 May 1994, leg. Jin Zhou; 51 ♂♂, 47 ♀♀, Shanxi, Jincheng, Lingchuan, Xizhashuicun, (35.78°N, 113.28°E; elevation 900 m), 12–18 July 2010, leg. Hai-Yan Bai, Lin-Lin Yang; 1 Å, Sichuan, Jianyang, Pingquan, (30.34°N, 104.64°E; elevation 350 m), 4 May 1994, leg. Jin Zhou; 1 ♀, Sichuan, Mabian, Yonghong, (28.55°N, 103.42°E; elevation 1200 m), 22 July 2004, leg. Ying-Dang Ren; 2 🖧, Sichuan, Tianquan, Lamahe, (30.35°N, 102.42°E; elevation 1300 m), 29 July 2004, leg. Ying-Dang Ren; 2 ථሪ, Zhejiang, Mt. Jiulong, (28.21°N, 118.68°E; elevation 400 m), 4-5 August 2011, leg. Lin-Lin Yang, Na Chen.

Distribution. China (Gansu, Guizhou, Hebei, Henan, Hubei, Qinghai, Shaanxi, Sichuan and Zhejiang).

Etymology. The specific name is derived from the Latin *dilatatus* (dilate), referring to the dilated distal part of the transtilla.

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