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***Quisarctus yasumurai* gen. et sp. nov. (Arthrotardigrada: Halechiniscidae) from a submarine cave, off Iejima, Ryukyu Islands, Japan**

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Abstract

Quisarctus yasumurai gen. et sp. nov. (Arthrotardigrada: Halechiniscidae) is described from the submarine cave ‘Daidokutsu’, off Iejima, Okinawa Islands, Ryukyu Islands, Japan. It is characterised by a cylindrical body, long primary clava and shorter lateral cirrus inserted on a common cirrophore, and simple digits of unequal lengths (without folds, peduncles, proximal pads, pretarsi, or wrinkles) that terminate in a sheathed, small, crescent-shaped claw with a minute calcar. Quisarctinae subfam. nov. is erected for this unique new genus.

Key words: Heterotardigrada, marine, meiobenthos, phylogeny, Quisarctinae subfam. nov., Tardigrada, taxonomy

Introduction

Halechiniscidae (Heterotardigrada: Arthrotardigrada) is a marine tardigrade family, which is probably polyphyletic (Jørgensen *et al.* 2010), with 29 genera in seven subfamilies. It is characterised by the presence of a complete set of cephalic cirri, primary clavae, four digits with claws on each leg, and the absence of sclerotized plates.

In this paper, I describe a new genus and subfamily of this probable ‘rag bag family’, which is erected based on a unique new species, with a cylindrical body, long primary clavae and simple digits, that was collected from a submarine cave in Japan.

Material and methods

Tardigrades were obtained from three litres of grey calcareous mud collected from the depth of 29 m in the submarine cave ‘Daidokutsu’, off the eastern coast of Iejima, Okinawa Islands, Ryukyu Islands, Japan (26°43'18"N, 127°50'00"E) on 6th November 2013 by Koshin Yasumura. The mud sample was fixed in 3% buffered formaldehyde. To concentrate the sample, it was rinsed with distilled water to remove formaldehyde, stirred with water-diluted Ludox® HS-40 colloidal silica (density ca. 1.20 g cm⁻³), kept still for at least 15 min. to allow the mud particles to settle. Any animals trapped in the supernatant were retained on a 32 µm mesh net (modified from Burgess (2001) density separation method). This procedure was repeated four times to increase the recovery. The specimens were sorted under a stereomicroscope and mounted in glycerol for phase-contrast microscopy (Olympus BX53). All micrographs were taken at 1000× magnification and multiple micrographs were composited with Adobe Photoshop CS6 (Adobe Systems Incorporated) for full view of objects larger than field of view.

Systematics

Order Arthrotardigrada Marcus, 1927

Family Halechiniscidae Thulin, 1928

Quisarctinae subfam. nov.

Diagnosis. Halechiniscidae with cylindrical body; primary clava and lateral cirrus arise from common cirrophore; primary clava longer than lateral cirrus; secondary clava undeveloped; leg IV sense organ as papilla; legs terminate in digits without folds, peduncles, proximal pads, pretarsi or wrinkles; internal digits longer than external digits; each digit terminates in sheathed, small, crescent-shaped claw with minute calcar; pair of ventrally opening seminal receptacles each with slender, sinuous duct terminating in spherical vesicle.

Etymology. Formed from the type genus.

Type genus. *Quisarctus* gen. nov.

Quisarctus gen. nov.

Diagnosis. Halechiniscidae with cylindrical body; primary clava and lateral cirrus arise from common cirrophore; primary clava longer than lateral cirrus; secondary clava undeveloped; stylet supports present; leg IV sense organ as elongate papilla with distal spine; legs terminate in digits without folds, peduncles, proximal pads, pretarsi or wrinkles; internal digits longer than external digits; each digit terminates in sheathed, small, crescent-shaped claw with minute calcar; pair of ventrally opening seminal receptacles each with slender, sinuous duct terminating in spherical vesicle.

Etymology. The generic epithet is a combination of *Quis*, a latin word for 'Who?' or 'What?' referring to the author's first impression of this animal, and *arctus*, a Latinised Greek word for 'bear' referring to the common name of tardigrades.

Type species. *Quisarctus yasumurai* sp. nov.

Quisarctus yasumurai sp. nov.

(Figs. 1, 2; Table 1)

Diagnosis. Halechiniscidae with cylindrical body; primary clava and lateral cirrus arise from common cirrophore; primary clava longer than lateral cirrus; secondary clava undeveloped; stylet supports present; leg I sense organ consists of three parts; leg II sense organ as unarticulated spine; leg III sense organ as unarticulated spine; leg IV sense organ as elongate papilla with distal spine; all legs terminate in four digits without folds, peduncles, proximal pads, pretarsi or wrinkles; internal digits longer than external digits; each digit terminates in sheathed, small, crescent-shaped claw with minute calcar; pair of ventrally opening seminal receptacles each with slender, sinuous duct terminating in spherical vesicle.

Material examined. *Holotype*: KUZ Z1649, adult female. *Paratypes*: KUZ Z1650, adult female; KUZ Z1651, four-digit juvenile.

Type locality: Water depth of 29 m, 'Daidokutsu' cave, off Iejima, Okinawa Islands, Ryukyu Islands, Japan (26°43'18"N, 127°50'00"E). Coll. by Koshin Yasumura on 6th November 2013.

Type depository: The type series is deposited in the Zoological Collection of Kyoto University.

Description of holotype (Figs. 1, 2A, 2B). Adult female, body length 106 µm. Cylindrical body with smooth cuticle. Round cephalic region without lobes. Complete set of three parted cephalic cirri present. Dorsal median cirrus with scapus (2 µm), tubular portion (7 µm) and flagellum (3 µm) inserted 13 µm posterior to anterior margin of body. Dorsal internal cirrus with scapus (2 µm), tubular portion (5 µm) and flagellum (4 µm) inserted on short cirrophore. Ventral external cirrus with scapus (4 µm), tubular portion (4 µm) and flagellum (2 µm) slightly exterior to internal cirrus. Internal cirri and external cirri situated near anterior margin of body. Lateral cirrus with scapus (3 µm), tubular portion (5 µm) and flagellum (4 µm) inserted on short lateral cirrophorus with primary clava. Elongated, proximally-inflated primary clava (27 µm) with distal pore. Lateral cirrus situated antero-dorsal to primary clava. Secondary clava undeveloped. Mouth directed anteriorly. Observation of bucco-pharyngeal apparatus incomplete due to limited visibility in mounting media. Bucco-pharyngeal apparatus with buccal tube

(21 μm), three placoids, pharyngeal bulb (11 $\mu\text{m} \times 11 \mu\text{m}$) and two stylets (furca not observed). Stylet supports present but not observed in holotype. Cirrus E (20 μm) with proximal portion, distal portion and tapering spine inserted on short cirrophore. Leg I sense organ (5 μm) articulated into three parts. Leg II and III sense organs (8 μm , 7 μm) as unarticulated spines. Leg IV sense organ (5 μm) with proximal papilla and short, distal tapering. Each leg terminates in four digits without folds, peduncles, proximal pads, pretarsi or wrinkles. Internal digits longer than external digits. Digits on leg IV longer than that of legs I–III. Each digit terminates in sheathed, small, crescent-shaped claw without dorsal spur. Minute calcar recognised in well-positioned claws of internal and external digits. Rosette-like female gonopore opens 5 μm anterior to caudal anus. Pair of seminal receptacles present. Slender, sinuous seminal receptacle duct opens ventrally near lateral margin of body slightly posterior to level of gonopore and runs exteriorly and then dorsally terminating in spherical vesicle. In holotype, seminal vesicle directed dorsally gives spherical appearance.

Remarks on paratypes. In the paratypic female, KUZ Z1650, the digits were in better position for observation (Figs. 2C, D). In addition, the stylet supports were more obvious (Fig. 2E) and the seminal vesicles were observed from lateral view (Fig. 2F). The other paratype, KUZ Z1651, was identified as a four-digit juvenile based on difficulty in observing the gonopore region and absence of both seminal receptacles and spermatozoa.

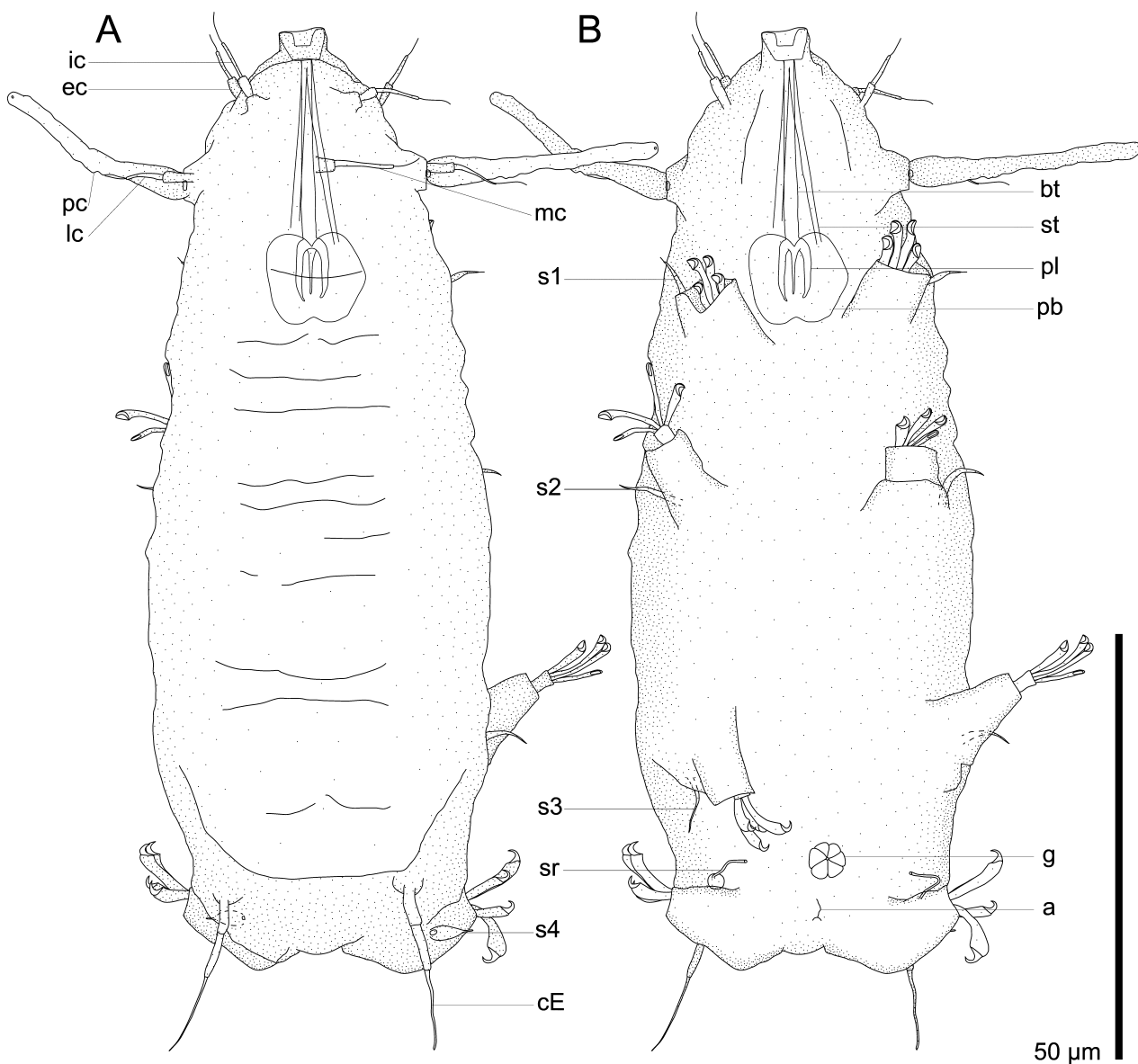


FIGURE 1. Drawing of *Quisarctus yasumurai* gen. et sp. nov., holotypic female [KUZ Z1649]. (A) Dorsal view; (B) ventral view. a, anus; bt, buccal tube; cE, cirrus E; ec, external cirrus; ic, internal cirrus; g, gonopore; lc, lateral cirrus; mc, median cirrus; pb, pharyngeal bulb; pc, primary clava; pl, placoid; sr, seminal receptacle; st, stylet; s1–4, sense organs of legs I–IV.

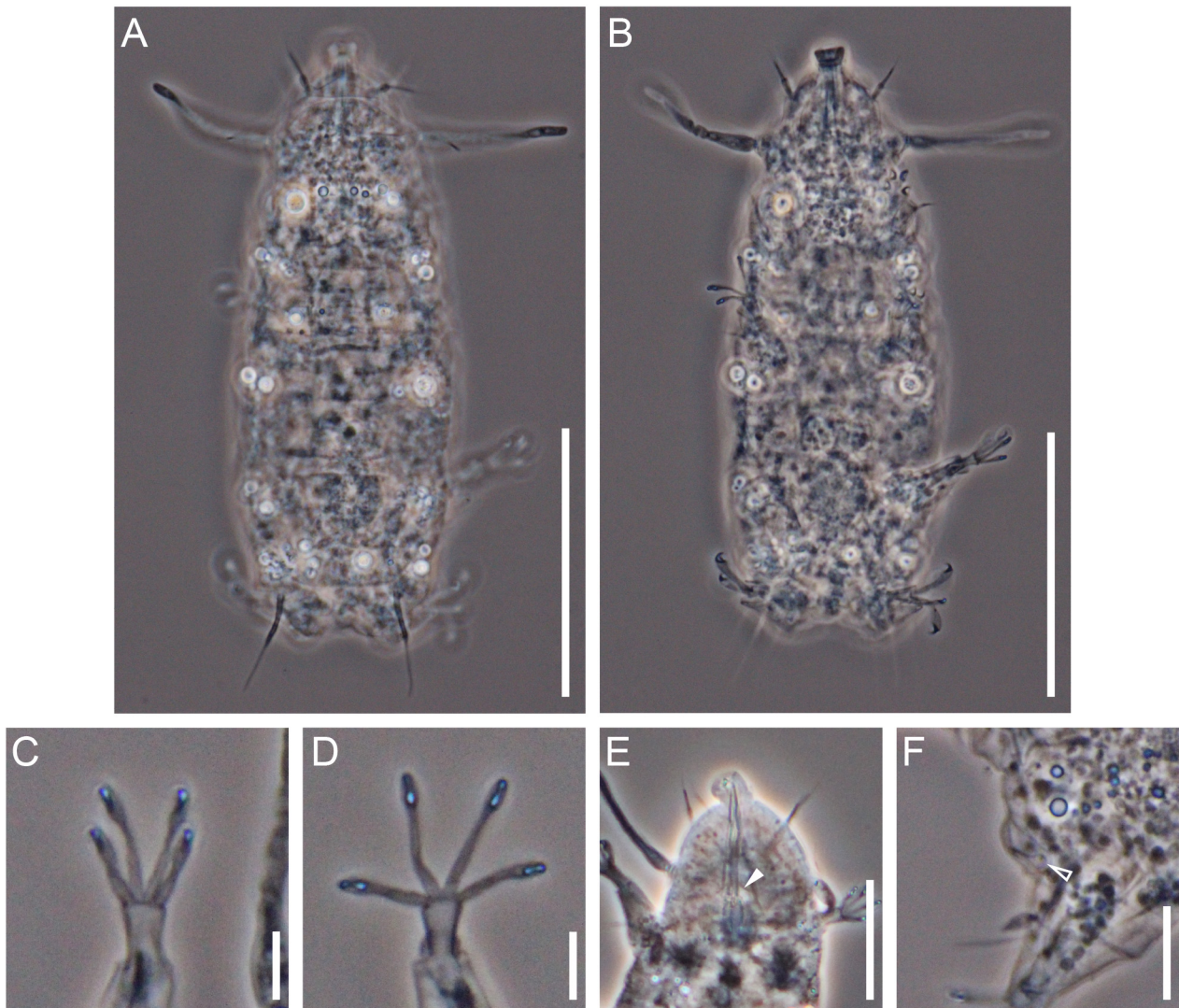


FIGURE 2. Phase contrast micrographs of *Quisarctus yasumurai* **gen. et sp. nov.** (A) Composite image: dorsal view of holotype [KUZ Z1649], scale bar= 50 μ m; (B) composite image: ventral view of holotype [KUZ Z1649], scale bar= 50 μ m; (C) leg I digits of paratype [KUZ Z1650], scale bar= 5 μ m; (D) leg IV digits of paratype [KUZ Z1650], scale bar= 5 μ m; (E) stylet support (white arrow head) of paratype [KUZ Z1650], scale bar= 10 μ m; (F) seminal vesicle (hollow arrow head) of paratype [KUZ Z1650], scale bar= 10 μ m.

Etymology. The specific epithet, *yasumurai*, is named in honor of Koshin Yasumura who collected the sediment samples from which the new species was obtained.

Discussion

The apparent cylindrical body of *Quisarctus yasumurai* **gen. et sp. nov.** resembles that of the four genera of Styraconyxinae Kristensen & Renaud-Mornant, 1983 (amended diagnosis, Kristensen & Higgins, 1984): *Angursa* Pollock, 1979 (amended diagnosis, Bussau, 1992); *Bathyechiniscus* Steiner, 1926; *Pleocola* Cantacuzène, 1951 (referring to Kristensen & Renaud-Mornant, 1983); and *Styraconyx* Thulin, 1942 (amended diagnosis, Kristensen & Higgins, 1984). However, the assignment of this species to any genera of this subfamily, excluding *Bathyechiniscus*, is easily ruled out by its digits without the characteristic peduncles and proximal pads of the subfamily. The original description of *Bathyechiniscus* did not describe the presence of these characters (Steiner, 1926), but it possesses another diagnostic character of the subfamily, i.e., ‘claws with two or more points’. The single-point claws of the new species do not accord with this trait. In addition, in contrast to Styraconyxinae

(except some *Angursa* species—See Table 1 in: Villora-Moreno, 1998), the primary clava of the new species is clearly longer than the lateral cirrus, as in Dipodarctinae Pollock, 1995, Florarctinae Renaud-Mornant, 1982, and Tanarctinae Renaud-Mornant, 1980, which Pollock (1995) suggested as a distinct character state at the subfamily level. These differences clearly distinguish the new species from Styraconyxinae.

TABLE 1. Morphometrics (μm) of *Quisarctus yasumurai* **gen. et sp. nov.** Dashes (-) represent unmeasured traits.

	Holotype	Paratype	Paratype
KUZ no.	Z1649	Z1650	Z1651
Status	Female	Female	Four-digit juvenile
Body length	106	111	97
Median cirrus	12	-	-
Internal cirrus	11	11	9
External cirrus	9	11	8
Lateral cirrus	12	13	8
Primary clava	27	27	20
Cirrus E	20	21	22
Leg I sense organ	5	7	6
Leg II sense organ	8	7	-
Leg III sense organ	7	7	6
Leg IV sense organ	5	4	4

The new species is easily distinguished from the other subfamilies as follows: by the body shape, the relative length of the primary clavae to the lateral cirrus and the digit patterns (referring to Pollock (1995) who considered these as distinct characters at subfamily level) and by the seminal receptacle morphology (referring to Noda (1986) who considered this a distinct character at subfamily level) and by the state of the secondary clavae (consistent in Dipodarctinae, Euclavarctinae, and Florarctinae). Specifically, *Quisarctus yasumurai* **gen. et sp. nov.** differs from:

- Dipodarctinae Pollock, 1995 (amended diagnosis, Jørgensen *et al.*, 2014) by the undeveloped secondary clavae, all legs with external digits shorter than internal digits, digits without peduncles, and ventrally opening seminal receptacles, which are considered to open laterally in species of *Dipodarctus* Pollock, 1995 (amended diagnosis, Jørgensen *et al.*, 2014).
- Euclavarctinae Renaud-Mornant, 1983 by the undeveloped secondary clavae, simple digits of unequal lengths without wrinkles, and morphology of the seminal receptacles.
- Florarctinae Renaud-Mornant, 1982 (amended diagnosis, Kristensen, 1984) by the cylindrical body without alae, undeveloped secondary clavae, digits without peduncles, and morphology of the seminal receptacles.
- Halechiniscinae Thulin, 1928 (amended diagnosis, Grimaldi de Zio *et al.*, 1990) by the cylindrical body, longer primary clavae, and simple digits of unequal lengths.
- Orzeliscinae Schulz, 1963 (amended diagnosis, Gross *et al.*, 2014) by the cylindrical body, longer primary clavae, and digits without adhesive organs.
- Tanarctinae Renaud-Mornant, 1980 by the cylindrical body, unmodified papillate leg IV sense organs, and simple digits without pretarsus.

The above discussion justifies the erection of a new genus and subfamily for the new species. However, to elucidate its phylogenetic relationships with the other subfamilies, future molecular phylogenetic studies of Halechiniscidae and related families are necessary.

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