

## Two new species of harpacticoid copepods from anchialine caves in karst area of North Vietnam

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Two new harpacticoid species belonging to the genera *Microarthridion* Lang, 1944 (Tachidiidae) and *Nitocra* Boeck, 1864 (Ameiridae) are recorded from underground caves in the karst area of Ninh Binh Province, North Vietnam. *Microarthridion thanhi* n. sp. is distinguished from congeneric species by the number of setae on the antennary exopod, the structure of leg 5 in both sexes, and the finger-like process modified from an outer distal pinnate seta of the third endopodal segment of leg 2 in the male. *Nitocra vietnamensis* n. sp. has the character combination of six setae on the ellipsoidal exopod of leg 5 in both sexes, the first endopodal segment of leg 1 shorter than the whole exopod, the seta/spine armature of an inner seta of P2-P4 enp-1 and four elements on P2 enp-3, and the reduction of the proximal endite to a seta on the maxillary syncoca.

**Keywords:** Anchialine; Harpacticoida; *Microarthridion*; new species; *Nitocra*; Vietnam

### Introduction

The family Tachidiidae is a rather small group which currently comprises 14 species of six genera (Huys et al. 2005). In the family, *Microarthridion* Lang, 1944 is the largest genus containing seven species (Boxshall and Halsey 2004). Recently, Kihara and Rocha (2007) described a new species, *M. corbisierae* Kihara and Rocha, 2007, from Brazil, and provided a short key to the eight species in the genus. Members of the genus *Microarthridion* are mainly marine and brackish, but some species are occasionally found in freshwaters.

The genus *Nitocra* Boeck, 1864, belonging to the family Ameiridae, currently contains 61 valid species and subspecies (Karanovic and Pesce 2002; Huys et al. 2005; Chang 2007). Most members of the genus are basically marine, but some species extend their habitat range to brackish waters or even to freshwaters such as karstic, anchialine caves (Karanovic and Pesce 2002; Boxshall and Halsey 2004).

In Vietnam, taxonomic studies on harpacticoid copepods have as of yet been poorly studied. Borutzky (1967) recorded seven species belonging to Canthocamptidae and Phyllognathopodidae, including two new species, *Elaphoidella vietnamica* and *Attheyella (Canthosella) vietnamica*, from caves in the karst area of Chi Ne, Lac Thuy District, Hoa Binh Province, North Vietnam. Ho and Tran (2007) have added five harpacticoid species to Vietnam fauna: *Neotachidius triangularis* (Shen and Tai, 1963), *Elaphoidella coronata* Sars, 1904,

*Onychocamptus mohammed* (Blanchard and Richard, 1891), *Enhydrosoma bifurcarostratum* Shen and Tai, 1965, and *Stenhelia (Delavalia) ornamentalia* Shen and Tai, 1965. Apostolov (2007) recorded two species from Lan Hoang cave in Huu Lung District, Lang Son Province, northern Vietnam: *E. bidens* (Schmeil, 1894) and *O. mohammed*. Recently, Chertoprud et al. (2009) reported two species of genus *Nitocra* and *Microarthridion* from Nha Trang Bay, South Vietnam: *N. cf. affinis* and *M. litospinatus* Shen and Tai, 1973.

The senior author participated in a field survey research on the underground caves in the karst area, which is located in the Trang An Tourism Area, Ninh Binh Province, North Vietnam, about 90 km south of Hanoi and about 40 km southwest of the coast. He collected several anchialine harpacticoid species, including two new species of the genera *Microarthridion* and *Nitocra*, which are described herein.

### Materials and methods

The harpacticoid copepods were collected from Son Duong cave (20°14'46.34''N, 105°55'39.94''E) and Ba Giot cave (20°14'55.31''N, 105°55'31.86''E) among five anchialine caves surveyed at the Trang An Tourism Area on 24 May 2010. Both are limestone caves studded with stalactites, where a subterranean river flows past slowly on a muddy bottom containing sands (about 1–2.5 m deep; distance from the exit 258 m and 272 m, respectively; 0.05‰ in salinity) (Table 1).

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Table 1. Information on the ground waters of five anchialine caves and their neighboring rivers in the Trang An Tourism Area, Ninh Binh Province, North Vietnam, surveyed on 24 May 2010.

	Son Duong cave*	Vung Tham cave	Sinh cave	Ba Giot cave*	Tran cave	River 1	River 2
Temp. (°C)	28.5	28.2	29.0	28.4	28.8	30.4	27.9
pH	6.8	7.3	6.6	7.2	7.4	7.5	7.4
DO (mg/l)	6.6	6.5	6.3	5.8	6.4	5.5	5.6
COD (mg/l)	7	9	10	11	8	15	12
NH <sub>4</sub> <sup>+</sup> (mg/l)	0.21	0.49	0.55	0.61	0.36	0.58	0.62
NO <sub>3</sub> <sup>-</sup> (mg/l)	0.44	0.39	0.32	0.37	0.36	0.67	0.55
PO <sub>4</sub> <sup>-</sup> (mg/l)	0.62	0.31	0.83	0.43	0.55	0.84	0.88
EC (µS/cm)	98	104	220	109	210	130	293
Distance from the exit (m)	258	310	160	272	125		
Water depth (m)	0.9–2.7	0.5–3.2	1.4–2.1	1.3–2.3	1.3–1.5	1.8	
Width (m)	2.2–4.5	2.5–10.8	2.5–4.7	2.0–4.5	1.8–3.2		
Salinity (‰)	0.05	0.05	0.11	0.05	0.10	0.06	0.14
Bottom type	mud and sand	mud and stones	mud, sand, and rock	mud, sand and rock	mud, sand and rock	thin mud	thin mud

\* type localities of two new harpacticoid species. DO, Dissolved oxygen; COD, Chemical oxygen demand; EC, Electrical conductivity.

Samples were taken with a conical plankton net of 80 µm mesh aperture by dragging it from the near-bottom to the surface on a boat. Copepods were fixed and stored in 4% buffered formalin.

Specimens were dissected and mounted in lacto-phenol. Specimens were observed under a differential interference contrast microscope (CH40, Olympus, Japan). All drawings were made with the aid of a camera lucida.

Type specimens are deposited in the Institute of Ecology and Biological Resources, Hanoi, Vietnam.

Abbreviations used in the text and figure legends follow the conventional ones frequently used for the taxonomy of copepods: enp-1 to enp-3 or exp-1 to exp-3, the first to third endopodal or exopodal segments of each leg; L/W, length to width ratio; P1-P6, first to sixth legs.

**Family Tachidiidae** Boeck, 1865

**Genus *Microarthridion*** Lang, 1944

***Microarthridion thanhi* n. sp.** (Figures 1–3)

**Description**

*Female.* Body (Figure 1A-B) µm (ranging 413–439 µm,  $m = 422$ ,  $N = 6$ ) in length; cycloform, with distinct separation between prosome and urosome, prosome ellipsoidal, urosome sub-cylindrical. Rostrum slightly protruding, discernible in dorsal view; triangular and its tip blunt with two pairs of sensilla.

Cephalosome shorter than next three prosomites combined, with spinules along posterior margin and setules around anteroventral margins; with median nuchal organ and paired lateral accessory nuchal organs.

Pedigers 2–4 each with paired accessory nuchal organs laterally; free margins of tergites with long

spinules along posterior and lateral margins; dorsal surfaces with three to four spinule rows. Pediger 5 trapezoidal with small spinules along posterior margin; dorsal and lateral surfaces with spinule rows; without accessory nuchal organ laterally.

Genital double-somite with large subcuticular chitinous suture marking line of fusion dorsally and laterally; about 0.84 times longer than wide in dorsal view; lateral surfaces of anterior half of double-somite with about five spinule rows; genital apertures fused medially forming a common genital slit; isolated copulatory pore round; pore canal slightly curved; raised ventral surface with one transverse spinule row posteriorly and two arched spinule rows laterally; long spinules arrayed all along posterior margin of genital double-somite.

Next urosomite with spinules around posterior margin; two to three spinule rows on ventral surface as in Figure 2A. Dorsal surface of penultimate urosomite smooth without spinule or setule ornamentation, lacking setule row along posterior margin; two spinule rows on ventral surface, setule row along posterior margin. Anal somite with one spinule row ventrally and row of six to nine minute spinules along insertion of each ramus ventrally. Anal operculum (Figure 1A, 2B) semicircular with about 20 or more fine setules along posterior edge.

Caudal rami (Figure 2B) columnar, about 1.17 times broader than long; armed with one oblique and inward-curving row of fine setules medially, and five to six spinules along posterior margin ventrally. Lateral caudal seta small, bare, locating near proximal end of lateral margin of caudal ramus. Outer caudal seta stout, spiniform, nearly as long as inner caudal seta. Terminal caudal setae well developed, pinnate, with

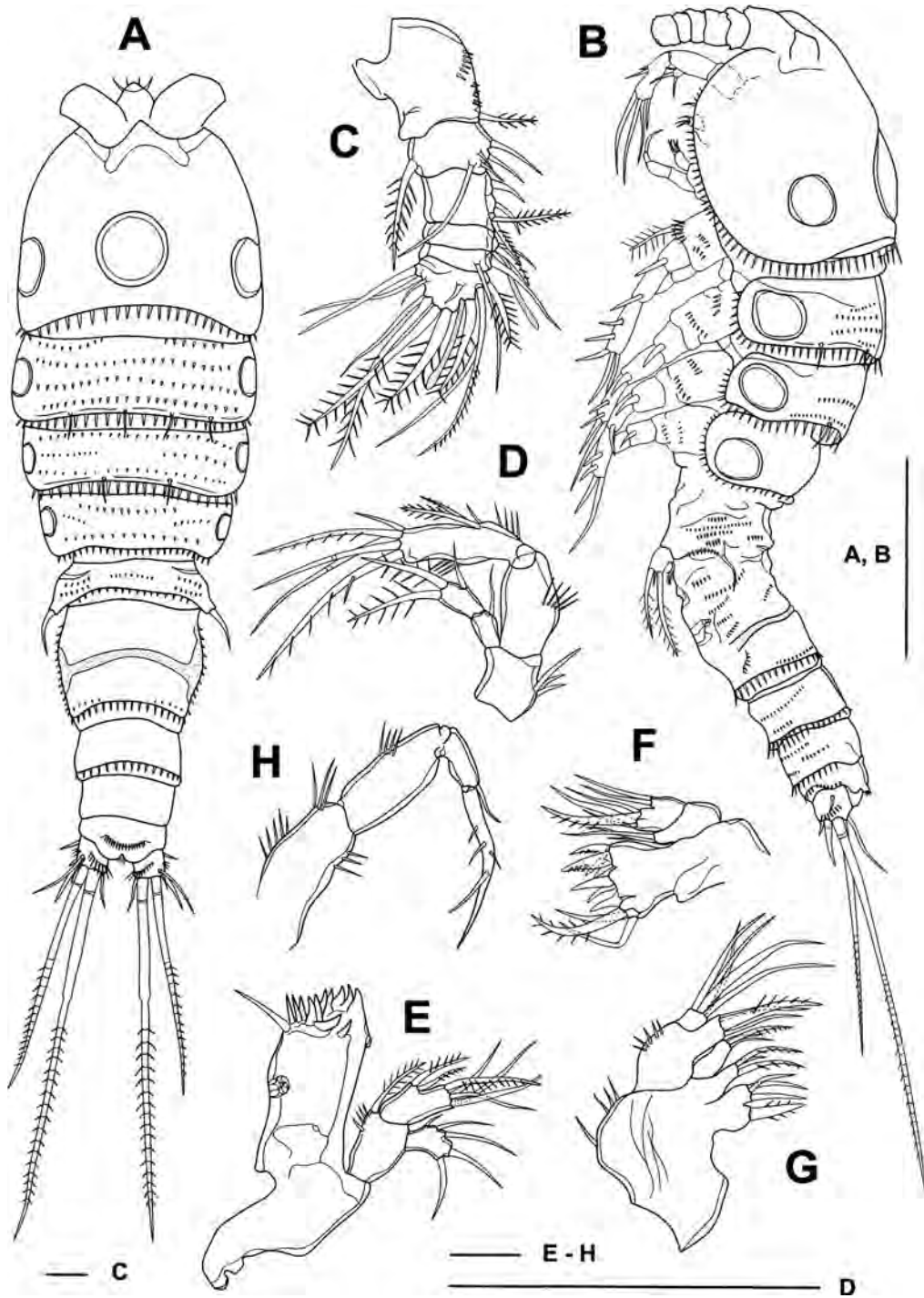


Figure 1. *Microarthridion thanhi* n. sp., female. A, habitus, dorsal; B, habitus, lateral; C, antennule; D, antenna; E, mandible; F, maxillule; G, maxilla; H, maxilliped. Scale bars = 100 μm (A, B, D) and 10 μm (C, E-H).

fracture plane. Dorsal caudal setae paired, locating a little laterally, both issuing from small projection; inner one very small, outer one naked, about 1.5 times as long as outer caudal seta.

Antennule (Figure 1C) short and blunt; 6-segmented; segment 1 with spinule row anteriorly; segment 2 with 1 pinnate seta on posterior margin; segments 4

and 5 small; segment 6 blunt. Setal formula: 1-[1], 2-[6], 3-[4], 4-[2 + aesthetasc], 5-[2], 6-[12 + aesthetasc].

Antenna (Figure 1D) with setule rows on medial margin of basis and proximal endopodal segment. Distal endopodal segment with two plumose setae and one naked seta in proximal third of lateral margin, and one naked seta, two plumose setae, and two

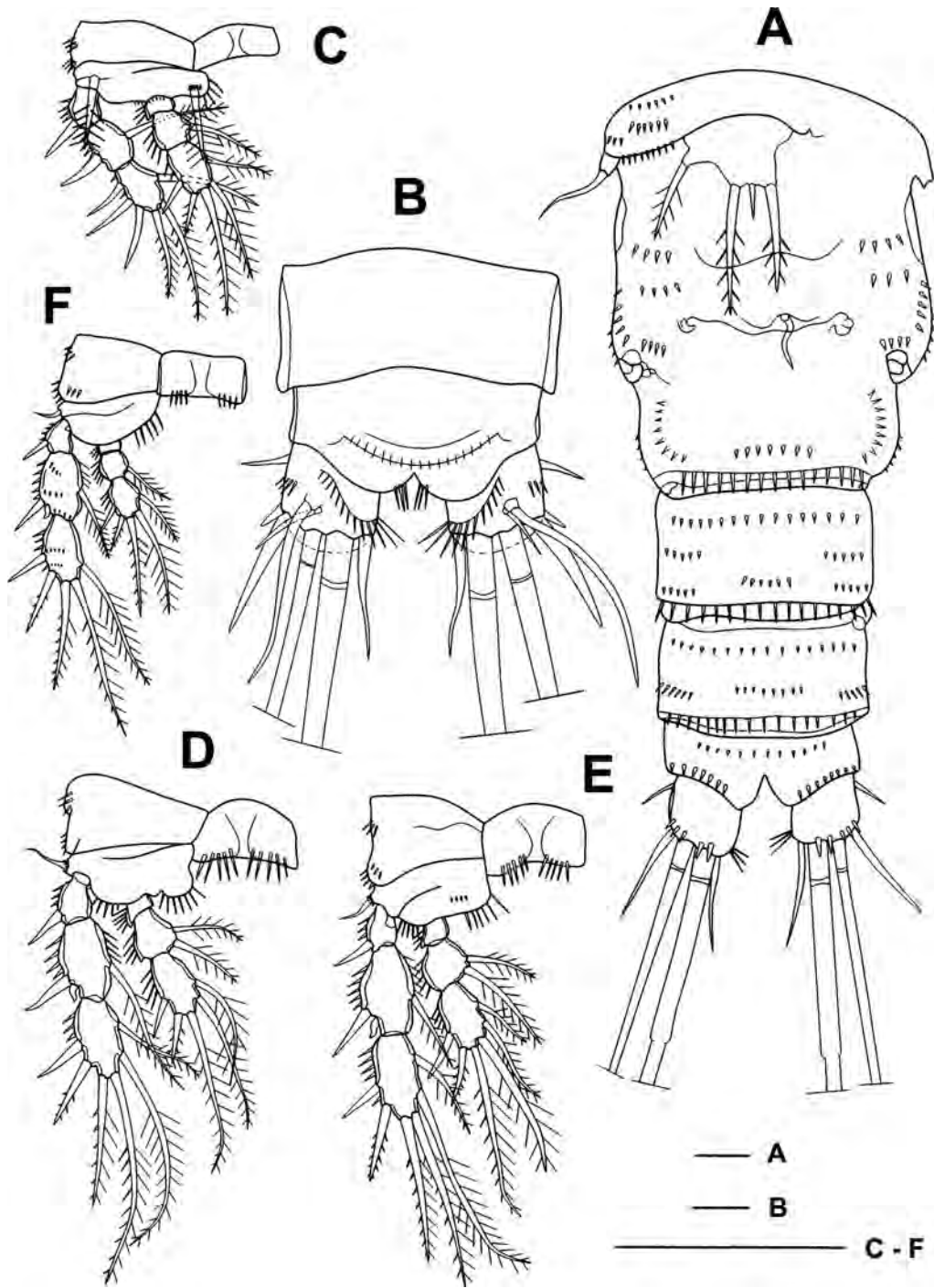


Figure 2. *Microarthridion thanhi* n. sp., female. A, urosome, ventral; B, anal somite and caudal rami, dorsal; C–F, P1– P4. Scale bars = 100  $\mu$ m (A, B) and 10  $\mu$ m (C–F).

geniculate setae apically. Exopod 2-segmented; proximal segment with one naked seta, distal segment with one naked lateral seta and two plumose apical setae.

Mandible (Figure 1E) with well-developed coxal gnathobase bearing about 12 teeth along distal margin, with one naked seta at dorsal corner; basis with one inner distal plumose seta flanking a setule row;

endopod with two pinnate inner setae and five apical setae (one pinnate and four naked setae); exopod 1-segmented, armed with three inner, one apical, and one outer distal setae.

Maxillule (Figure 1F) with well-developed praecoxal arthrite bearing seven spines, one pinnate seta and one geniculate seta, and one naked seta around distal

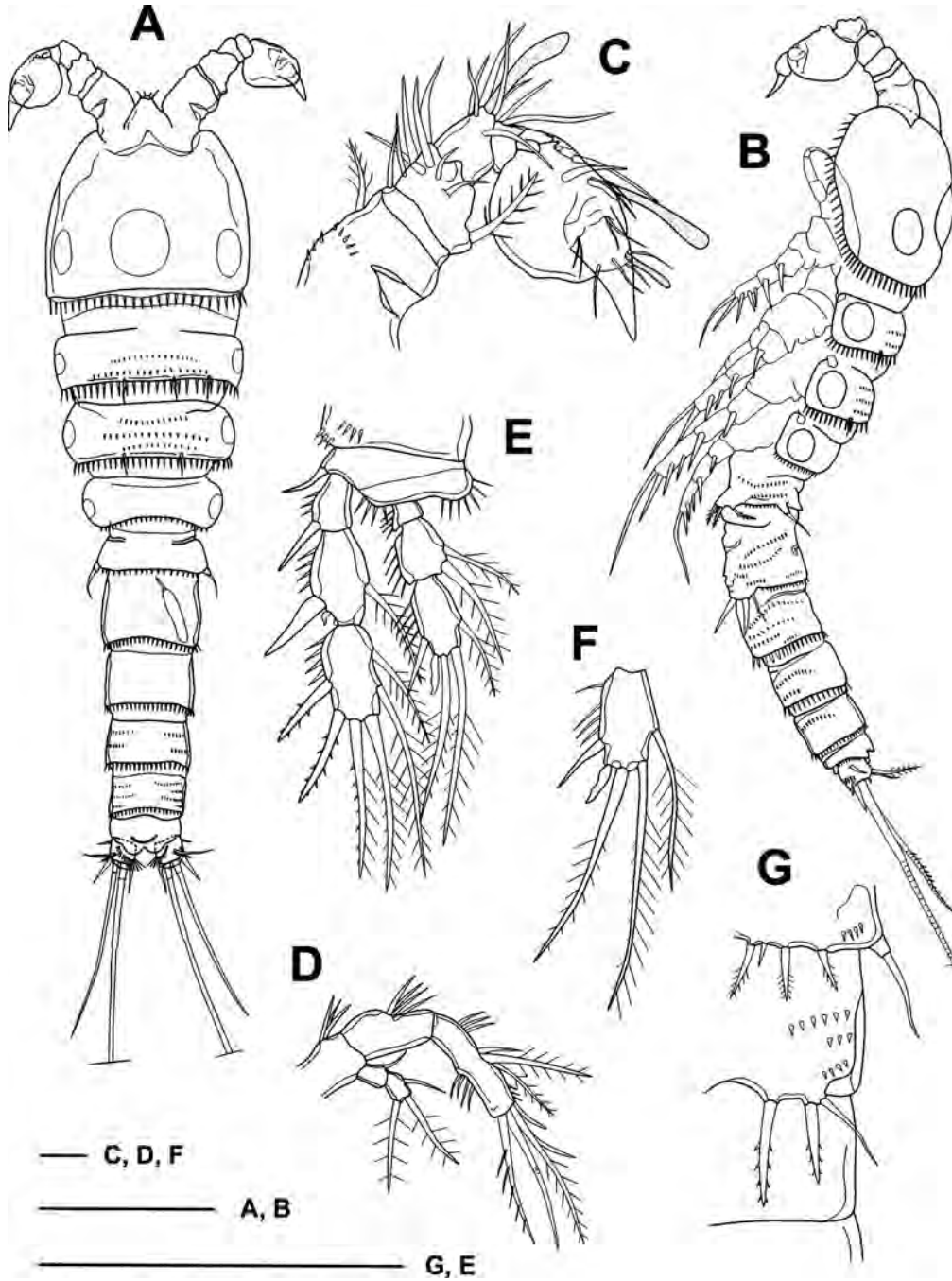


Figure 3. *Microarthridion thanhi* n. sp., male. A, habitus, dorsal; B, habitus, lateral; C, antennule; D, antenna; E, P2; F, P2 endopod 3; G, P5 and P6. Scale bars = 100 μm (A, B, E, G) and 10 μm (C, D, F).

margin; coxal endite with three setae; endopod and exopod fused to basis, lobe not obviously divided, bearing six setae in total.

Maxilla (Figure 1G), syncoxa with three endites and a setule row along outer margin; proximal endite expanded distally with one large pinnate seta and three naked setae; middle endite bearing one naked and two pinnate setae; distal endite with two naked and one pinnate setae; allobasis forming 1 strong

pectinate claw, flanked by two setae, with setule row along outer margin; endopod 1-segmented armed with five setae.

Maxilliped (Figure 1H) subchelate; syncoxa elongate, with setule rows near inner and outer distal corners and on outer margin; basis with setule row near middle of outer margin; endopod one-segmented, bearing one elongate claw armed with four spinules as accessory armature and one naked seta distally.

Table 2. Character comparison of *Microarthridion thanhi* n. sp. with congeneric species.

	Antennular segments, ♀	Setae on antennary exopod	Seta + spine on P1 enp-3	Inner seta on P2-P4 enp-2	Segments of P4 endopod	Seta/spine on endopodal lobe of P5, ♀
<i>berberum</i>	6	4	5	1:1:1	3	4/0
<i>corbisierae</i>	5	5	4	1:1:0	2	3/1
<i>fallax</i>	7	5	5	1:1:1	3	4/0
<i>laurenticum</i>	5	5	5	2:2:1	3	3/0
<i>litospinatus</i>	6	5	5	2:2:1	3	3/1
<i>littorale</i>	6	4	5	2:2:1	3	3/0
<i>perkinsi</i>	6	5	5	2:2:1	3	4/0
<i>reductum</i>	6	5	5	2:2:1	3	4/0
<i>thanh</i> n. sp.	6	4	5	2:2:1	3	3/1

P1 (Figure 2C). Intercoxal sclerite bilobate, without spinules on anterior surface. Coxa with two spinule rows on outer margin. Basis with inner distal corner produced, bearing setule row along inner margin and one pinnate seta, its tip nearly reaching to middle of enp-3. Both endopod and exopod 3-segmented. Endopod slightly shorter than exopod, with enp-1 small, lacking inner seta; enp-3 with one spine, two pinnate, and two plumose setae; exp-1 without inner seta; exp-3 with three outer spines, two pinnate apical, and one plumose inner setae. P2–P4 (Figure 2D–F). Intercoxal sclerites with spinules anteriorly along posterior margin. Coxa typically with spinule rows on outer margin and outer distal corner. Inner margin of basis forming lobate setulose expansion, lacking inner seta. Exopod longer than endopod; both rami 3-segmented; distal outer margin of exp-2 of P2–P4 with arched apophysis; exp-3 with two outer spines, two pinnate apical, two plumose inner setae (on P2–P3) or one plumose inner seta (on P4); enp-1 very small, lacking inner seta, bearing outer margin with spinule row; enp-2 with two plumose inner setae on P2–P3 and one plumose inner seta on P4; enp-3 of P2 and P4 with one outer spine, two pinnate apical, and two plumose inner setae; enp-3 of P3 with one outer spine, two pinnate apical, and three plumose inner setae. Seta/spine formula of P1–P4:

P1	basis 1-1	exp I-0; I-1; III,2,1	enp 0-0; 0-1; I,2,2
P2	basis 1-0	exp I-0; I-1; II,2,2	enp 0-0; 0-2; I,2,2
P3	basis 1-0	exp I-0; I-1; II,2,2	enp 0-0; 0-2; I,2,3
P4	basis 1-0	exp I-0; I-1; II,2,1	enp 0-0; 0-1; I,2,2

P5 (Figure 2A), fused to fifth pediger basally, with spinule row along posterior margin of fifth pediger; forming a plate-like inner lobe, bearing three stout pinnate setae and one minute spine between innermost and median setae, innermost seta shorter median seta; outer angle produced dorsolaterally to a knob-like projection with a basal seta.

*Male.* Sexually dimorphic in size, urosome ornamentation and segmentation, antennule, P2 endopod, P5 and P6. Body (Figure 3A–B) 438 µm in length (ranging 423–449 µm,  $m = 434$ ,  $N = 5$ ), relatively narrower than females; prosome ellipsoidal, urosome cylindrical. All somites armed with spinule row along posterior margin. Spermatophore narrow, elongated.

Antennule (Figure 3C) 7-segmented, geniculate between segments 5 and 6; segment 1 with spinule rows anteroproximally, one plumose seta distally; segment 2 short with one seta; segment 3 with one plumose seta posteroproximally and seven naked setae dorsally and anteriorly; segment 4 small, with six setae and one aesthetasc; segment 6 very large and swollen, anterior margin forming multicuspidate process proximally and one long aesthetasc extending to middle of segment 7; last segment hook-shaped with six naked setae and one small aesthetasc. Armature formula: 1-[1], 2-[1], 3-[8], 4-[6 + aesthetasc], 5-[2], 6-[4 + aesthetasc], 7-[6 + aesthetasc].

P2 endopod, outer distal pinnate seta modified as finger-like process with repand outer edge, as in Figure 3E–F.

P5 (Figure 3G) vestigial, represented by a small plate incorporated into pediger 5, armed with three stout pinnate setae and one minute spine between innermost and median setae; outer angle produced to a knob-like projection with basal seta, as in female.

P6 (Figure 3G) represented by a small plate incorporated into outer distal corner of genital somite, bearing two inner stout spines and one outer naked seta.

#### *Specimens examined*

*Holotype:* female (IEBR-COP2313), *allotype:* male (IEBR-COP2314), dissected and mounted on slide, Son Duong cave (20°14'46.34''N, 105°55'39.94''E), Ninh Binh Province, North Vietnam, 24 May 2010,

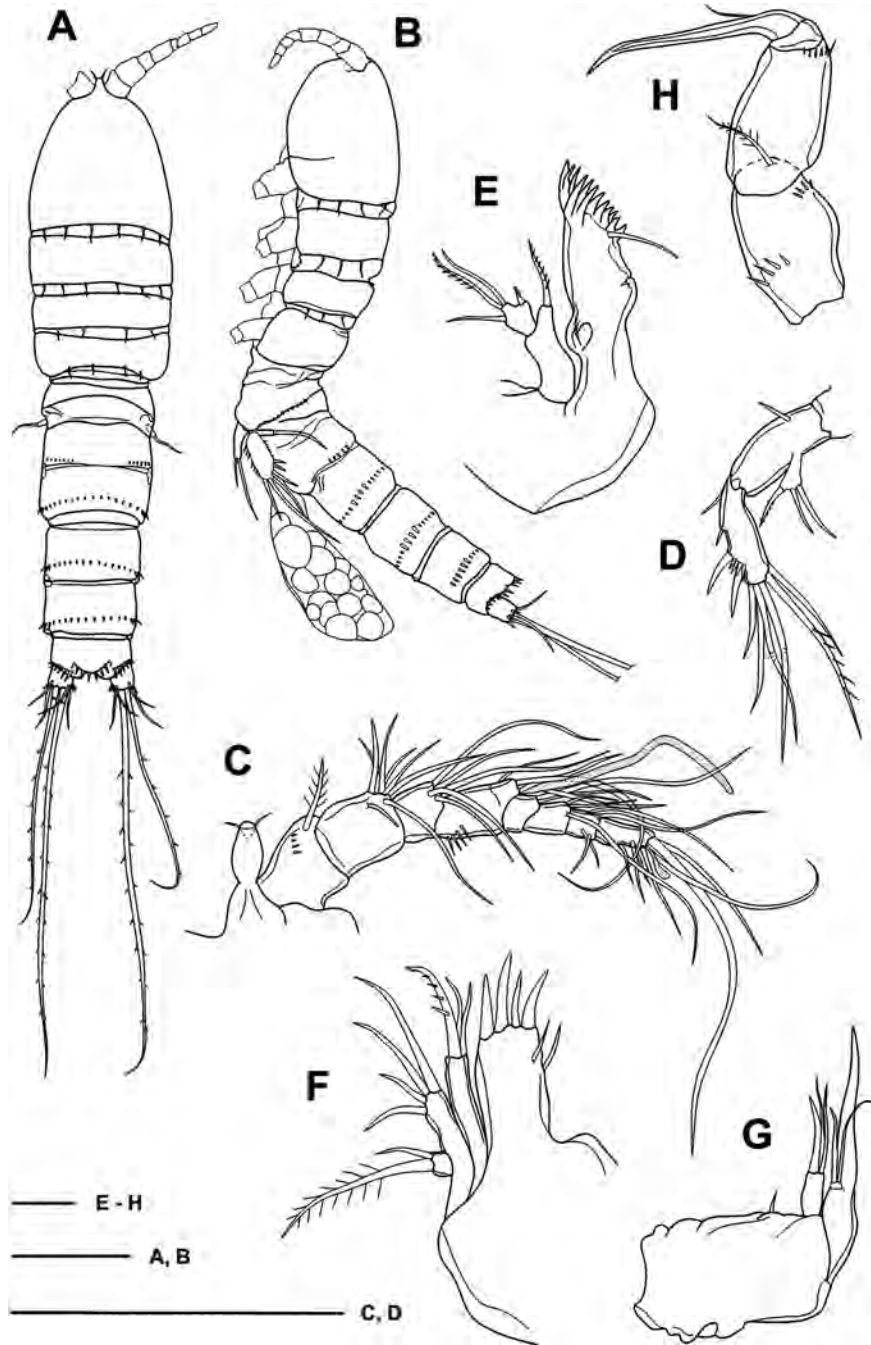


Figure 4. *Nitokra vietnamensis* n. sp., female. A, habitus, dorsal; B, habitus, lateral; C, antennule; D, antenna; E, mandible; F, maxillule; G, maxilla; H, maxilliped. Scale bars = 100 μm (A–D) and 10 μm (E, F).

leg. DL Tran. *Paratypes*: 5f (IEBR-COP2315-2319) and 3m (IEBR-COP2320-2322), dissected in lactophenol, collection details same as in holotype; 1f (IEBR-COP2323), 1m (IEBR-COP2324), dissected, Ba Giot cave (20°14'55.31"N, 105°55'31.86"E), near the locality of holotype, 24 May 2010, leg. DL Tran. *Other materials*: 9f, 4m, 3 copepodites, undissected and preserved in 70% alcohol (IEBR-COP-AED0182003), collection details same as in holotype.

**Etymology**

The species is dedicated to Prof. Dr. Dang Ngoc Thanh, acknowledging his contribution to the systematics of Vietnamese copepods.

**Ecology**

The species was found near the bottom of the cavern river. This species co-occurred with *Pseudodiaptomus*

*bulbosus* (Shen and Tai, 1964) (Calanoida), *Mesochra suiifunensis* Borutzky, 1952 (Harpacticoida), *Halicyclops sinensis* Kiefer, 1928 (Cyclopoida), *Moina dubia* Guerne and Richard, 1892 and *Diaphanosoma sarsi* Richard, 1894 (Cladocera). The cavern water is nearly freshwater (0.5‰ in salinity) but seems to be slightly affected by seawater, in light of the co-occurring brackish-water copepods. Furthermore, taking into consideration that the inside environment (cf. Table 1) is directly or indirectly connected with the outside by the cavern river, the present new species is not supposed to be a genuine troglobiont.

### Remarks

Among eight recognized species of *Microarthridion*, *M. corbisierae* is easily distinguished from the congeneric species by the 2-segmented endopod of P4 and reduction in the setal armature of P1–P4 (Table 2).

*Microarthridion thanhi* n. sp. differs clearly from other four congeneric species, *M. reductum* (Monard, 1935), *M. laurenticum* (Nicholls, 1940), *M. fallax* Perkins, 1956, and *M. perkinsi* Bodin, 1970, which have five setae in total on the antennary exopod and four (or three) unmodified setae on the endopodal lobe of P5 in both sexes.

The new species is allied to *M. littorale* (Poppe, 1881) and *M. berberum* (Monard, 1935) in sharing four setae on the antennary exopod. However, the new species is different from them in the armature of the endopodal lobe of P5 in both sexes, that is, three stout pinnate setae and a smooth slender spine between the innermost and median setae.

Considering the armature of the endopodal lobe of P5, *M. thanhi* n. sp. resembles *M. litospinatus* Shen and Tai, 1973. However, in addition to the number of setae on the antennary exopod (five in *M. litospinatus* versus four in *M. thanhi*), they differ from each other in the shape of the projection of the P2 enp-3 in males (modified as a crochet-form projection in *M. litospinatus*, while being nearly a finger-shape in *M. thanhi*).

**Family Ameiridae** Monard, 1927

**Genus Nitocra** Boeck, 1864

***Nitocra vietnamensis* n. sp. (Figure 4–6)**

### Description

**Female.** Body (Figure 4A–B) 550 µm in length (ranging 525–615 µm,  $m = 562$ ,  $N = 8$ ), elongated, cylindrical, without clear distinction between prosome and urosome. Rostrum narrow, protruding anteriorly, not defined at its base, with two delicate dorsal sensilla. Cephalothorax nearly as long as next three prosomites combined; minute hairs scattered on dorsal surface,

with six sensilla along posterior margin. Distolateral margin of each prosomite not protruded laterally, with several sensilla of posterior margin. Urosome comprising fifth pedigerous somite, genital double-somite and three free abdominal somites. Genital double-somite, first and second free abdominal somites with hyaline fringe posteriorly. Genital double-somite about 1.1 times broader than long, subdivided dorsally, bearing row of small cuticular spines marking line of fusion dorsally, armed with spinule row along its posterior margin on dorsal side, continued laterally and one spinule row ventrally. First free abdominal somite with spinule row along posterior margin, continued laterally and ventrally. Second free abdominal somite ornamented as in posterior part of genital double-somite (Figure 4A–B, 5A). Posterior margin of anal somite (Figure 5B) with about 9 to 12 large spinules dorsally and ventrally, with row of spinules around insertion of each ramus on ventral side. Anal operculum convex with five to six strong marginal spinules.

Caudal rami nearly rectangular, about 1.05 times longer than wide in dorsal view (ranging 1.04–1.09,  $m = 1.07$ ,  $N = 6$ ); with one transverse setule row along inner face, three spinules on distomedial edge of caudal ramus ventrally, and two spinules near outer distal caudal seta. Lateral caudal seta locating dorsolaterally; outer distal caudal seta about 1.3 times longer than inner distal caudal seta; dorsal caudal seta about 0.5 times shorter than inner distal caudal seta, inserted near distomedial corner with chitinous ridge produced; terminal caudal seta very strong, pinnate, about twice as long as outer terminal caudal seta (Figure 4A, 5A–B).

Antennule (Figure 4C) 8-segmented, with one very long aesthetasc on segment 4 and one short aesthetasc on last segment. Armature formula: 1-[1], 2-[8], 3-[5], 4-[4 + aesthetasc], 5-[2], 6-[3], 7-[4], 8-[7 + aesthetasc]. All setae smooth, except plumose seta on segment 1.

Antenna (Figure 4D) with allobasis incorporating first endopodal segment, with one spine on abexopodal margin; distal endopodal segment with some spinules and two curved spines on outer margin, with six setae apically; exopod 1-segmented, armed with three distal setae, outer seta longest.

Mandible (Figure 4E) with well-developed, elongate coxal gnathobase bearing about 12 bicuspidate teeth along distal margin and with one lateral seta; palp 2-segmented, basis elongated, with only one plumose seta; endopod fused to basis; exopod small with one inner lateral seta, one outer lateral seta, and two apical setae.

Maxillule (Figure 4F) with praecoxal arthrite bearing four elements on distal edge and two spinules on inner edge; coxal arthrite with cylindrical endite bearing one strong spiniform seta and two slender apical setae; basal endite bearing four naked setae apically; endopod fused to basis; exopod 1-segmented,



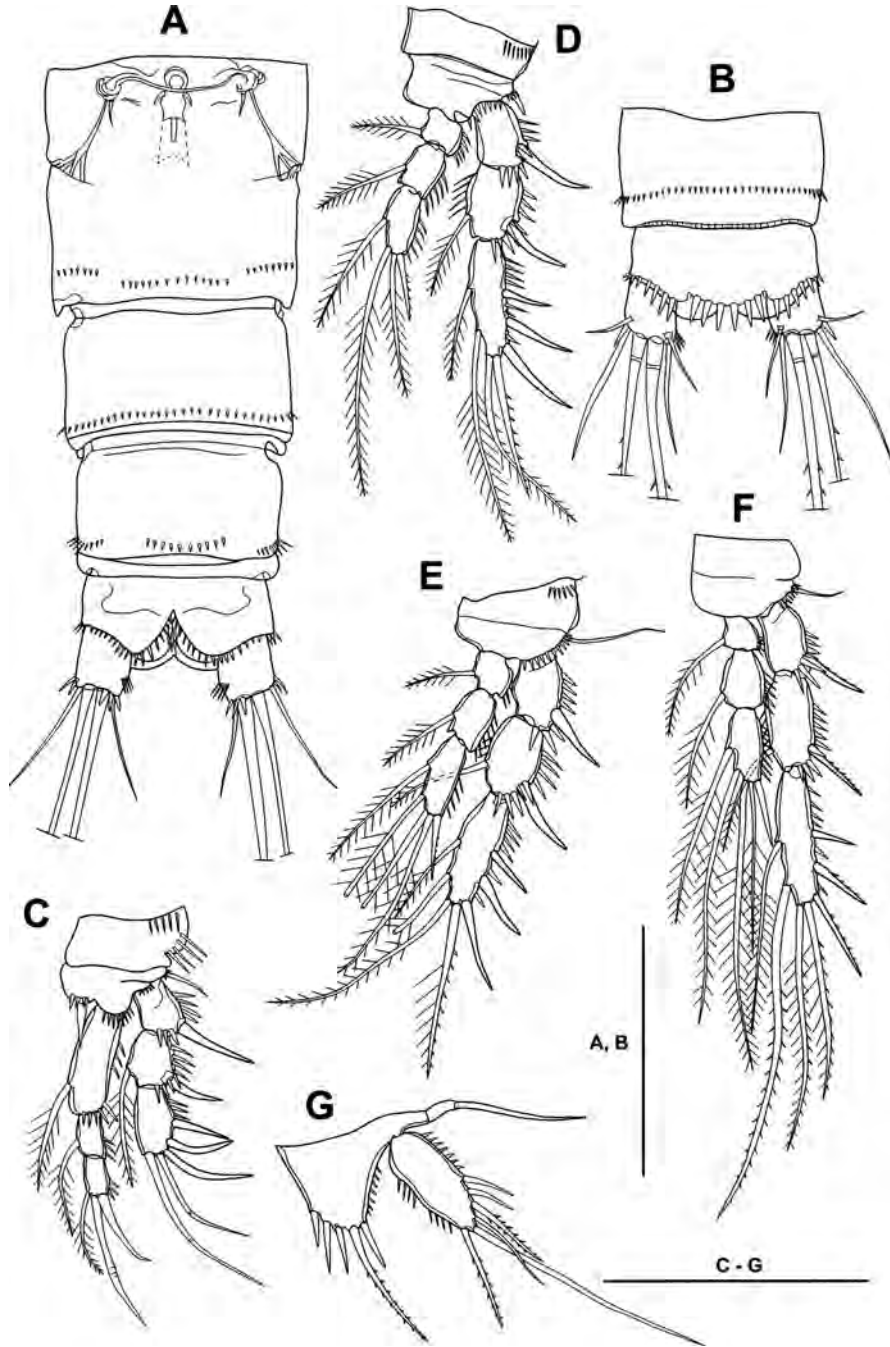


Figure 5. *Nitokra vietnamensis* n. sp., female. A, abdomen, ventral; B, anal somite and caudal rami, dorsal; C–G, P1–P5. Scale bars = 100  $\mu$ m (A–G).

bearing one small naked seta, and one long plumose seta.

Maxilla (Figure 4G). Syncoxa with one seta, spiniform, and one endite with two slender and one strong apical setae; allobasis forming one strong claw; endopodite one-segmented, small, with two apical setae.

Maxilliped (Figure 4H) subchelate; syncoxa with one pinnate seta distomedially, one arched row of spinules proximally, and one spinule row on outer distal

corner; basis with one arched row of spinules on outer distal corner; endopod one-segmented, armed with one strong curved apical claw and one minute seta as accessory armature.

P1–P4 with 3-segmented endopods and exopods. P1 (Figure 5C) basis armed with one spiniform seta inner distally, with spinule row near its base, and one seta outer distally; enp-1 elongated, slightly longer than sum of exp-1 and exp-2, not quite reaching to middle

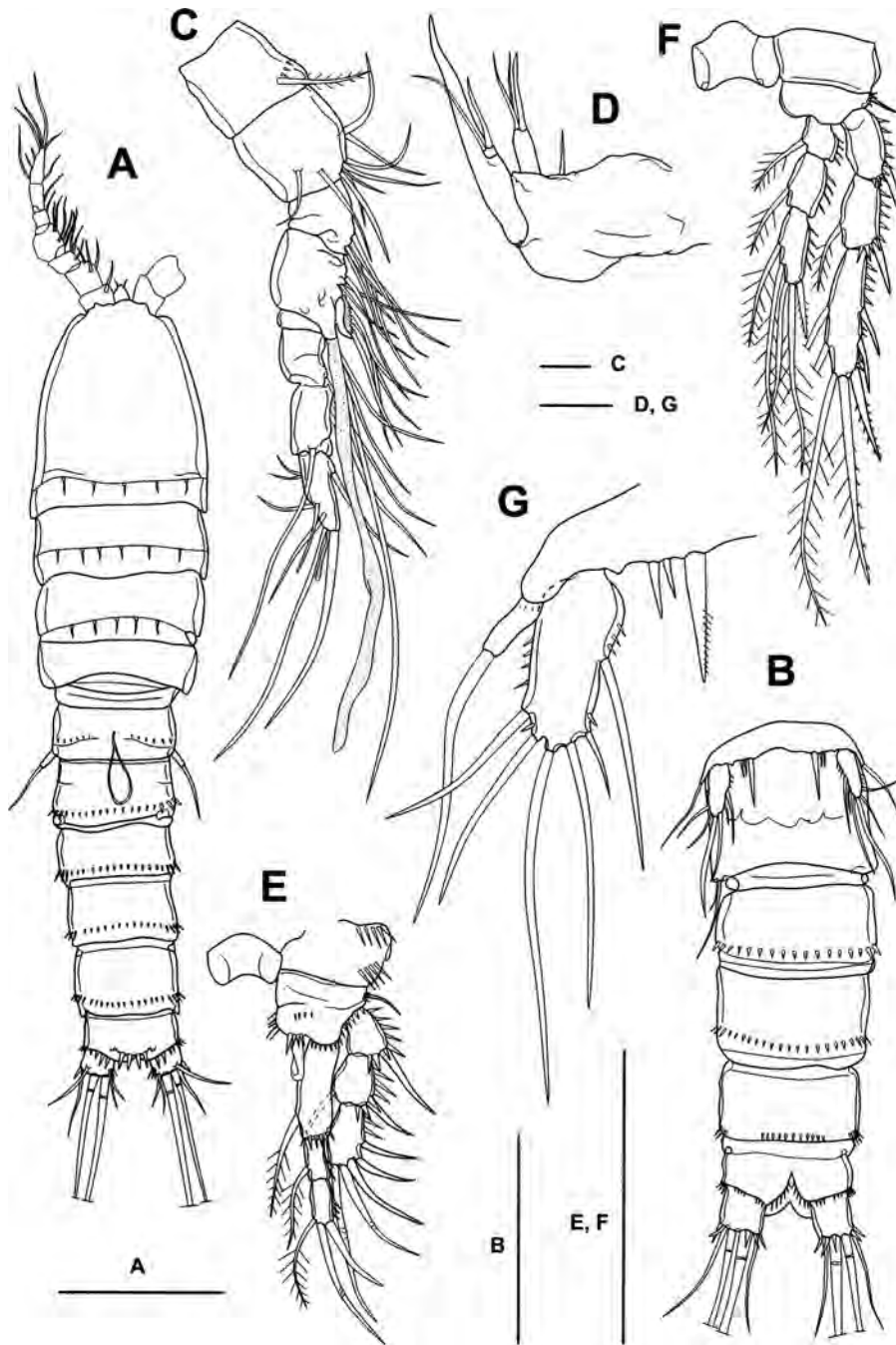


Figure 6. *Nitokra vietnamensis* n. sp., male. A, habitus, dorsal; B, P5, P6 and urosome, ventral; C, antennule; D, maxilla; E, P1; F, P3; G, P5. Scale bars = 100 μm (A, B, E, F) and 10 μm (C, D, G).

of exp-3, with one stout inner seta on distal quarter of inner margin; enp-3 with one outer spine, one geniculate apical seta, and one plumose inner seta; exp-2 with one inner seta; exp-3 with three outer spines and two geniculate apical setae. P2–P4 (Figure 5D–F) enp-1 with inner seta; both exopods and endopods of P2–P4 with spinule rows on outer margins. P2 enp-3 with four elements of one outer distal spine, two apical and one

inner plumose setae. P4 exp-3 with one elongate, stout, spiniform inner distal seta for supporting egg sac. Seta and spine armature of P1–P4 as follows:

P1	basis 1-I	exp I-0; I-1; III,2,0	enp 0-1; 0-1; I,1,1
P2	basis I-0	exp I-0; I-1; III,2,2	enp 0-1; 0-1; I,2,1
P3	basis 1-0	exp I-0; I-1; III,2,2	enp 0-1; 0-1; I,2,2
P4	basis 1-0	exp I-0; I-1; III,2,2	enp 0-1; 0-1; I,2,2

P5 (Figure 5G) baseoendopod triangular, nearly reaching to middle of exopod, bearing total of five setae, with spinule row on outer margin; exopod ellipsoidal, tapering distally, 2.45 times longer than wide (ranging 2.35–2.52,  $m = 2.42$ ,  $N = 6$ ) with total of six setae (one inner distal, one apical, and 4 outer setae), with spinule rows along both inner and outer margins; apical seta longest, more than two times as long as subapical ones.

**Male.** Body (Figure 6A-B) similar to female in general appearance, but smaller and more slender than female, 510  $\mu\text{m}$  in length (ranging 482–525  $\mu\text{m}$ ,  $m = 515$ ,  $N = 4$ ). Spinule ornamentation of urosomites including anal somite, nearly same as in female. Anal operculum convex with four to five strong marginal spinules. Spermatophore as shown in Figure 6A. Sexual dimorphism shown in A1, inner distal seta on P1 basis, P5, and P6. A1 (Figure 6C) eight-segmented; geniculated between segments 2 and 3, and between segments 6 and 7; segment 4 with hyaline, denticulate chitinous process along distal inner margin and with one very long aesthetasc; segment 5 small, without seta; aesthetasc on last segment slender; armature formula: 1-[1], 2-[9], 3-[6], 4-[10 + aesthetasc], 5-[0], 6-[3], 7-[1], 8-[8 + aesthetasc].

Seta on inner distal margin of P1 basis modified as hook-like protrusion with hyaline membrane, flanking four spinules around its base (Figure 6E). P3 enp-3 (Figure 6F) not modified.

P5 (Figure 6G), inner lobe of baseoendopod not protruding; posterior margin rather straight, with one innermost swollen seta and two slender naked setae; exopod ellipsoidal, 2.30 times longer than broad (ranging 1.95–2.35,  $m = 2.28$ ,  $N = 4$ ), with spinule row on outer margin, bearing six setae including two outer, three inner, and one apical setae. P6 (Figure 6B)

armed with two setae, inner one about 2.5 times as long as outer one.

### Specimens examined

**Holotype:** female (IEBR-COP2325); **allotype:** male (IEBR-COP2326), dissected and mounted on slide, Son Duong cave (20°14'46.34''N, 105°55'39.94''E), Ninh Binh Province, North Vietnam, 24 May 2010, leg. DL Tran. **Paratypes:** 6f (IEBR-COP2327-2332), 3m (IEBR-COP2333-2335), dissected, collection details same as in holotype. **Other materials:** 2f, 1m, 1 copepodite, preserved in 70% alcohol (IEBR-COP-AED0182004), collection details same as in holotype.

### Etymology

The specific epithet, *vietnamensis*, is a modified version of the word “Vietnam”.

### Remarks

Genus *Nitocra* currently comprises 61 species or subspecies (Pesce, 2011), of which *N. vietnamensis* n. sp. is allied with the species possessing elongated P1 enp-1, but shorter than whole exopod, and eight seta/spine elements on P4 exp-3 (Table 3).

*N. vietnamensis* n. sp. is clearly distinguished from *N. spinipes* Boek, 1864, *N. fragilis* Sars, 1905, *N. sewelli* Gurney, 1927, *N. uenoi* Miura, 1962, and *N. intermedia* Pesca, 1983 by the elongated, ellipsoidal P5 exopod in both sexes: about 2.3–2.5 times longer than wide in the female, and 1.95–2.35 times in the male, while being rather oval in both sexes of the congeners (in the female, about 1.3–2.0 times in *N. spinipes*, 1.7 times in *N. sewelli*, about 1.5 times in *N. uenoi*, and 1.4–1.6 times in *N. fragilis* and *N. intermedia*).

Table 3. Character comparison of *Nitocra vietnamensis* n. sp. with its allied congeneric species.

	Anal operculum, spinules	Caudal ramus, L/W	P1, enp-1 vs. exopod	P2-P4 enp-1, inner setae	P2 enp-3, setae + spine	P5 exopod setae, ♀/♂	♀, P5 exopod
<i>arctolongus</i>	6	~1	<<	0:0:0	3	6/6	ellipsoidal
<i>bdelluriae</i>	0	~2	<	0:0:0	4	6/5(6)	ellipsoidal
<i>fragilis</i>	10–14	<1	<	1:1:1	4	5/6	oval
<i>intermedia</i>	7–8	~1	<	1:1:1	4	5/6	oval
<i>koreanus</i>	4–6	~1	<<	1:1:1	4	6/6	ellipsoidal
<i>lacustris</i>	7–8	~1	<<	0:0:0	3	6/6	ellipsoidal
<i>malaica</i>	7–8	>1	<	0:0:0	4	6/6	ellipsoidal
<i>pietschmanni</i>	4–6	~1	<<	1:1:1	4	6/6	round
<i>platypus</i>	4	>1	<<	1:0:0	4	6/5	round
<i>sewelli</i>	?	<1	<	1:1:1	4	6/6	oval
<i>spinipes</i>	8–14	~1	<	1:1:1	4	5/6	oval
<i>uenoi</i>	11	~1	<	0:0:0	4	5/-	oval
<i>vietnamensis</i>	5–6	~1	<	1:1:1	4	6/6	ellipsoidal

Moreover, *N. vietnamensis* differs from *N. spinipes*, *N. fragilis*, and *N. intermedia* in bearing six setae on the P5 exopod in females, while there are only five setae in the other three species. Furthermore, *N. vietnamensis* differs from *N. spinipes* by 5–6 spinules of anal operculum (versus 8–14 in *N. spinipes*), and differs from *N. spinipes* and *N. fragilis* by the armature maxillary syncoxa (proximal endite reduced to a small seta in the new species, while two endites not reduced in *N. spinipes* and *N. fragilis*).

*N. vietnamensis* differs from *N. sewelli* by relatively longer caudal rami (slightly longer than wide, while a little shorter than wide in *N. sewelli*), the triangular basoendopod of P5 (versus round in *N. sewelli*) and by the spine/seta armature of P1 exp-3 in the female (five elements in the new species, versus four in *N. sewelli*).

*N. bdelluræ* Liddell, 1912 apparently differs from the other species as well as from the new species by an elongated caudal rami (nearly 2 times longer than wide), smooth anal operculum, and elongated enp-3 of P1 (about 2 times longer than enp-2 in *N. bdelluræ*, while nearly equal or slightly longer than enp-2 in the other species).

*N. uenoi* was described on the basis of a female specimen collected from a coastal well in the Okinawa Islands, Japan. The specimen showed a reduction of the armature of the female P5 baseoendopod (four instead of five) as in the case in *N. humphreysi* Karanovic and Pesce, 2002 from anchialine ground waters of a karst area in northwestern Australia (Karanovic and Pesce 2002). *N. uenoi* also shows some reduction in the setal armature of the antennary exopod (two setae instead of three), and deficiency of inner seta on P2–P4 enp-1 and 4 elements on P2 enp-3. However, the new species does not show the reduction in the characteristics above, and has even more setae on the P5 exopod in both sexes (six instead of five). The reduction in the new species can be seen only in the armature of the maxillary syncoxa with a distal endite and a small seta representing the proximal endite.

*N. vietnamensis* resembles *N. malaica* Kiefer, 1929 in the structure of the P1 endopod and the shape of P5 in the female. However, *N. malaica* is distinguished by the absence of inner seta on P2–P4 enp-1, three seta/spine elements on P3 enp-3 in the male (versus five in *N. vietnamensis*), and two spiniform setae on the P5 basoendopod in the male (versus three in *N. vietnamensis*).

Among the congeners from the northeastern Pacific, *N. vietnamensis* is distinguished from *N. platypus* Daday, 1906 by the inner seta of P3–P4 enp-1, and from *N. arctolongus* Shen and Tai, 1973 and *N. lacustris* (Schmankevitch, 1875) by the inner seta of P2–P4 enp-1 and four seta/spine elements on P2 enp-3. *N. vietnamensis* is also discernible from *N. pietschmanni*

(Chappuis, 1934) by the ellipsoidal P5 exopod in both sexes (versus round in *N. pietschmanni*).

*N. vietnamensis* is very similar to *N. koreanus* Chang, 2007 in sharing the characteristics of only 4–6 spinules along the posterior margin of anal operculum, four seta/spine elements on P2 enp-3, the presence of inner seta of P2–P4 enp-1, an ellipsoidal P5 exopod in both sexes, and three spiniform setae on the P5 basoendopod in the male. However, besides some difference of relative length of P1 enp-1, it differs from *N. koreanus* by the reduced maxillary syncoxa, with only an endite and a seta (versus two endites in *N. koreanus*).

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### References

- Apostolov A. 2007. Notes sur les harpacticoides cavernicoles (Crustacea: Copepoda) de Vietnam du nord. *Hist Nat Bulg.* 18:65–73.
- Borutzky EW. 1967. Copépodes harpacticoides d'eaux douces de Vietnam du Nord. *Arch Zool Mus Univ. Moscou.* 46(7):1015–1023.
- Boxshall GO, Halsey SH. 2004. An introduction to copepod diversity. Vols. 1 and 2. London: Ray Society. p. 1–966.
- Chang CY. 2007. Two harpacticoid species of genera *Nitokra* and *Ameira* (Harpacticoida: Ameiridae) from brackish waters in Korea. *Integr Biosc.* 11(2):247–253.
- Chertoprud ES, Gómez S, Gheerardyn H. 2009. Harpacticoida (Copepoda) fauna and the taxocene diversity of the South China Sea. *Oceanology.* 49(4):488–498.
- Ho TH, Tran DL. 2007. To add six new species freshwater crustaceans (Cyclopoida, Harpacticoida-Copepoda) to the fauna of inland freshwater zooplankton of Vietnam. *J Biol.* 29(2):9–16.
- Huys R, Ohtsuka S, Conroy-Dalton S, Kikuchi Y. 2005. Description of two new species of *Neotachidius* Shen and Tai (Copepoda, Harpacticoida, Tachidiidae) from Korean brackish waters and proposal of a new genus for *Tachidius* (*Tachidius*) *vicinospinalis* Shen and Tai, 1964. *Zool J Linn Soc.* 143:133–159.
- Karanovic T, Pesce GL. 2002. Copepods from ground waters of Western Australia, VII. *Nitokra humphreysi* sp. nov. (Crustacea: Copepoda: Harpacticoida). *Hydrobiol.* 470:5–12.
- Kihara TC, Rocha CEF. 2007. *Microarthridion corbisierae* sp. nov. (Harpacticoida, Tachidiidae), um novo copépode da meiofauna dolitoral norte do litoral norte estado de São Paulo, Brasil. *Rev Bras Zool.* 24(4):907–918.
- Pesce GL. 2009. Harpacticoida: *Nitokra* Boeck, 1864. [Internet] Copepod Web Portal [cited 2011 March 21]. Available from: <http://www.lucioesce.net/copepods/arpa/nitokra.htm>.