Two species of *Tortanus* (*Eutortanus*) (Copepoda: Calanoida: Tortanidae) new to Korea

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Two species of *Tortanus* (*Eutortanus*) are newly recorded from shallow Korean waters: *T.* (*E.*) *vermiculus* Shen, 1955 and *T.* (*E.*) *komachi* Itoh, Ohtsuka and Sato, 2001. As a result of this study, five species are reported in the subgenus of the family Tortanidae in Korea. The sequence of cytochrome *c* oxidase subunit 1 (CO1) is also provided as a molecular characteristic.

Keywords: cytochrome c oxidase subunit 1, Eutortanus, Korean waters, Tortanidae

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Introduction

The planktonic calanoid copepod genus *Tortanus* consists of five subgenera: *Tortanus* Giesbrecht, 1892, *Eutortanus* Smirnov, 1935, *Acutanus* Ohtsuka, 1992, *Atortus* Ohtsuka, 1992 and *Boreotortanus* Ohtsuka and Reid, 1998.

Among them, the subgenus *Eutortanus* Smirnov, 1935 is distributed exclusively in subtropical/temperate brackish waters in East Asia (Ohtsuka *et al.*, 1992; 1995) and comprises seven species: *T. (E.) derjugini* Smirnov, 1935, *T. (E.) dextrilobatus* Chen and Zhang, 1965, *T. (E.) komachi* Itoh, Ohtsuka and Sato, 2001, *T. (E.) sheni* Hulsemann, 1988, *T. (E.) spinicaudatus* Shen and Bai, 1956, *T. (E.) terminalis* Ohtsuka and Reid, 1998 and *T. (E.) vermiculus* Shen, 1955. Although these species are likely to occur in Korean waters due to geographical proximity, only three species, *T. (E.) derjugini*, *T. (E.) dextrilobatus* and *T. (E.) spinicaudatus* have so far been reported (Kim, 1985; Ohtsuka *et al.*, 1992; 1995; Soh, 2010).

In this study, we collected two tortanid copepods, T. (E.) vermiculus and T. (E.) komachi, which are reported for the first time to the Korean copepod fauna. In addition, sequences of the mitochondrial cytochrome c oxidase subunit 1 (CO1) were also obtained for their molecular characteristics.

MATERIALS AND METHODS

Morphological taxonomy

T. (E.) vermiculus and T. (E.) komachi were collected

from Ganghwado Island (Incheon) and three localities on the east coast of Korea, respectively. The specimens were preserved in 95% ethanol immediately after collection. The appendages of the specimens were dissected in glycerol on a slide glass under a stereomicroscope (Olympus SZX-7; Olympus, Tokyo, Japan) and observed using a light microscope (Leica DM 2500; Leica Microsystems, Wetzlar, Germany). All drawings and measurements were made with the aid of a drawing tube.

The Roman and Arabic numerals in the armature formula represent spines and setae, respectively. All voucher specimens are deposited in the National Institute of Biological Resources (NIBR), Incheon, Korea.

Molecular taxonomy

Single antennule was removed from each specimen for genomic DNA extraction. The other body parts were used for the morphological observation. The genomic DNA was extracted using a RED Extract-N-Amp Tissue PCR kit (Sigma-Aldrich, St. Louis, MO, USA) according to the manufacturer's instructions. Mitochondrial CO1 was amplified by polymerase chain reaction (PCR) using universal primers of LCO1490 and HCO2198 (Folmer et al., 1994). PCR amplification was carried out under the following conditions: 3 min at 94°C, 35 cycles of 95°C for 15 sec, 48°C for 30 sec, and 72°C for 90 sec, with a final 72°C extension reaction for 7 min. The PCR products were gel-purified using a QIAquick® Gel Extraction Kit (Qiagen, Valencia, CA, USA) and sequenced with an ABI PRISM® 3700 DNA Analyzer using a BigDye Terminator Cycle Sequencing Ready Reaction Kit (Applied Biosystems, Foster City, CA, USA).

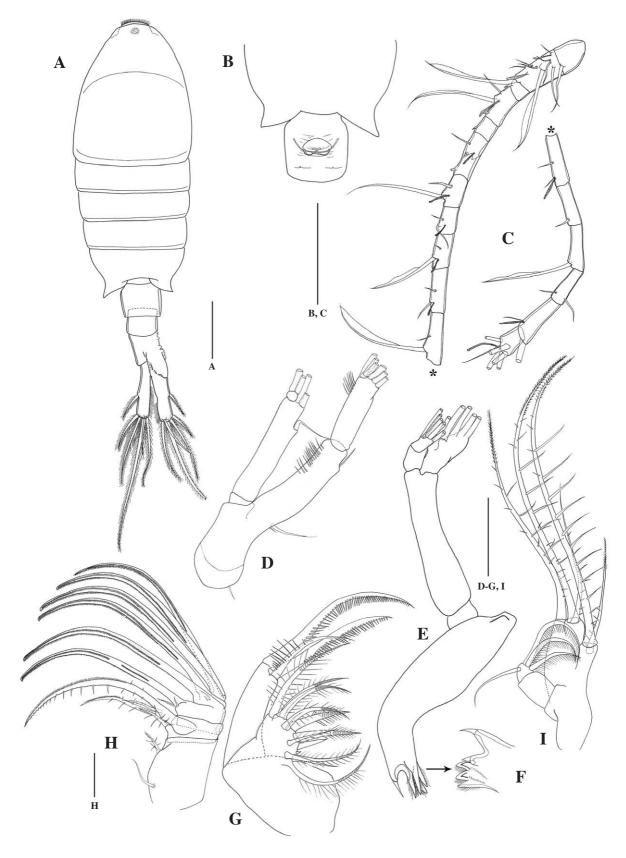


Fig. 1. *Tortanus* (*Eutortanus*) *vermiculus*, female. A. habitus, dorsal view. B. prosomal end and genital segment, ventral view. C. antennule. D. antenna. E. mandible. F. gnathobase of mandible. G. maxillule. H. maxilla. I. maxilla. Scale bars: A-C=0.25 mm, E-I=0.1 mm.

Systematic Accounts

Family Tortanidae Sars, 1902

Genus *Tortanus* Giesbrecht, 1898 (in Giesbrecht and Schmeil, 1898)

Subgenus Eutortanus Smirnov, 1935

^{1*}Tortanus (Eutortanus) vermiculus Shen, 1955 (Figs. 1-3)

Tortanus vermiculus Shen, 1955, 80, figs. 5-6; Chen and Zhang, 1965, 117, figs. 5-8.

 triangular process. Urosome 3-segmented, distal urosomite incompletely fused with caudal rami (Fig. 1A); genital double-somite as wide as long; genital operculum located ventromedially, oval with posterior margin slightly convex (Fig. 1B); anal somite incompletely fused with left caudal ramus and almost completely with right caudal ramus having uneven dorsolateral process on anterior right side. Caudal rami asymmetrical, right ramus slightly longer than left.

Antennule (Fig. 1C) 18-segmented, ancestral segments II to IX, XI to XII, and XXVI to XXVIII completely fused. Armature as follows; I, 1; II-IX, 9+2ae; X, 1; XI-XII, 3+ae; XIII, 1; XIV, 2+ae; XV, 1; XVI, 2+ae; XVII, 2+ae; XVIII, 2+ae; XIX, 2+ae; XX, 2; XXI, 2+ae; XXII, 1; XXIII, 1; XXIV, 1+1; XXV, 1+1+ae; XXVI-XXVIII, 6+ae. Antenna (Fig. 1D) coxa with a seta; basis and endopod fused; basis with 2 unequal setae; endopod 2-segmented, proximal segment with 1 outer seta and a row of spinules subterminally on inner margin, distal segment with 6 setae and a row of spinules subterminally. Mandible (Fig. 1E, F) with gnathobase bearing 3 mono-

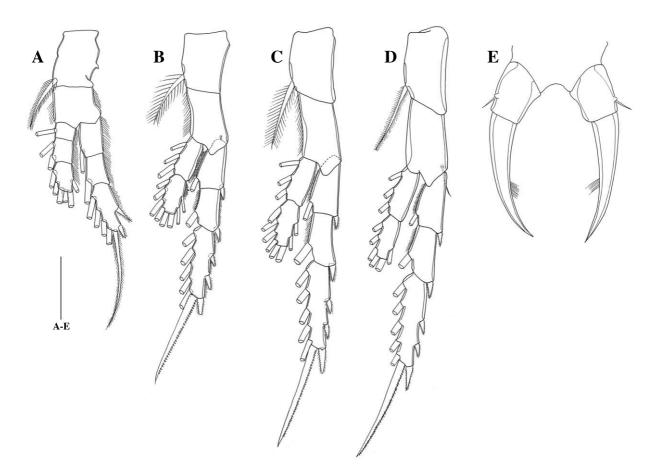


Fig. 2. Tortanus (Eutortanus) vermiculus, female. A. leg 1, anterior view. B. leg 2, anterior view. C. leg 3, anterior view. D. leg 4, anterior view. E. leg 5, anterior view. Scale bar: A-E=0.1 mm.

Korean name: 1*비대칭부푼긴노벌레(신칭)

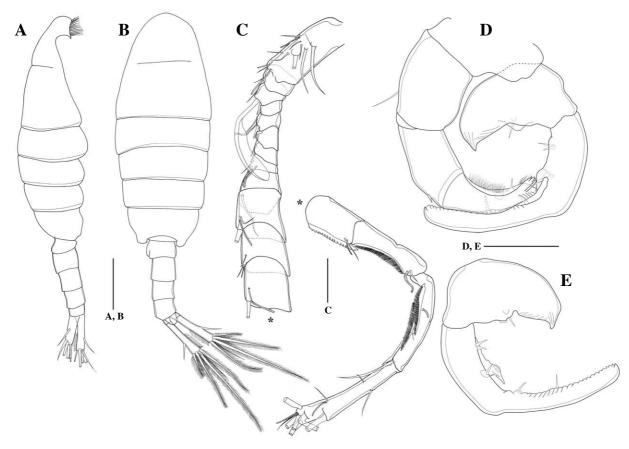


Fig. 3. Tortanus (Eutortanus) vermiculus, male. A. habitus, lateral view. B. habitus, dorsal view. C. right antennule. D. leg 5, posterior view. E. right leg 5, anterior view. Scale bars: A, B=0.25 mm, C-E=0.1 mm.

cuspid and 2 bicuspid teeth; endopod and exopod with 5 setae, respectively. Maxillule (Fig. 1G) without basis, endopod and exopod; praecoxal arthrite bearing 12 setae and 1 minute seta; coxal endite with 3 stout, spinulose setae. Maxilla (Fig. 1H) well developed; first and second praecoxal endites with 1 and 2 setae, respectively; first and second coxal endites with 1 and 3 setae, respectively; basal endite with 1 long and 2 short setae; endopod with 5 long and 2 rudimentary setae. Maxilliped (Fig. 1I) with setal formula of praecoxal and coxal endites: 0, 2, 2, 1. Basis unarmed; endopod 1-segmented, with 4 setae.

Seta and spine formula of legs 1 to 4 (Fig. 2A-D) as follows:

	coxa	basis	exopod segment			endopod segment		
			1	2	3	1	2	3
Leg 1	0-1	0-0	0-1	0-1	II, 1, 4	0-1	0-2	1, 2, 3
Leg 2	0-1	0-0	I-1	I-1	III, I, 5	0-3	2, 2, 4	
Leg 3	0-1	0-0	I-1	I-1	III, I, 5	0-3	2, 2, 4	
Leg 4	0-1	1-0	I-1	I-1	III, I, 5	0-3	1, 2, 3	

Leg 5 (Fig. 2E) 3-segmented; both coxal segments fused completely with intercoxal sclerite to form a common base; basis with a seta at point two-thirds along outer margin; exopod 1-segmented, tapering distally; both exopods with row of hairs along inner margin; left exopod slightly longer than right.

Male: Body length 1.68 mm (Fig. 3A, B). Cephalosome and first pedigerous somite separated; fourth and fifth pedigerous somites completely fused. Last pedigerous somite slightly asymmetrical, produced into blunt process. Urosome 5-segmented, first urosomite asymmetrical; second urosomite longest. Caudal rami asymmetrical, right ramus slightly longer than left.

Right antennule (Fig. 3C) 16-segmented and geniculate; ancestral segments I to VIII, XXI to XXIII, and XXIV to XXVIII completely or incompletely fused. Armature as follows; I-VII, 10+2ae; VIII, 1; IX, 2; X, 1; XI, 2+ae; XII, 1; XIII, 1; XIV, 2+ae; XV, 1; XVI, 2+ae; XVIII, 2+ae; XVIII, 2+ae; XIX, 1+ae+process; XXI-XXIII, 2+ae+2 processes; XXIV-XXVIII, 10+2ae.

Leg 5 (Fig. 3D, E) right and left coxal segments fused to form common base. Right leg chelate; basis produced

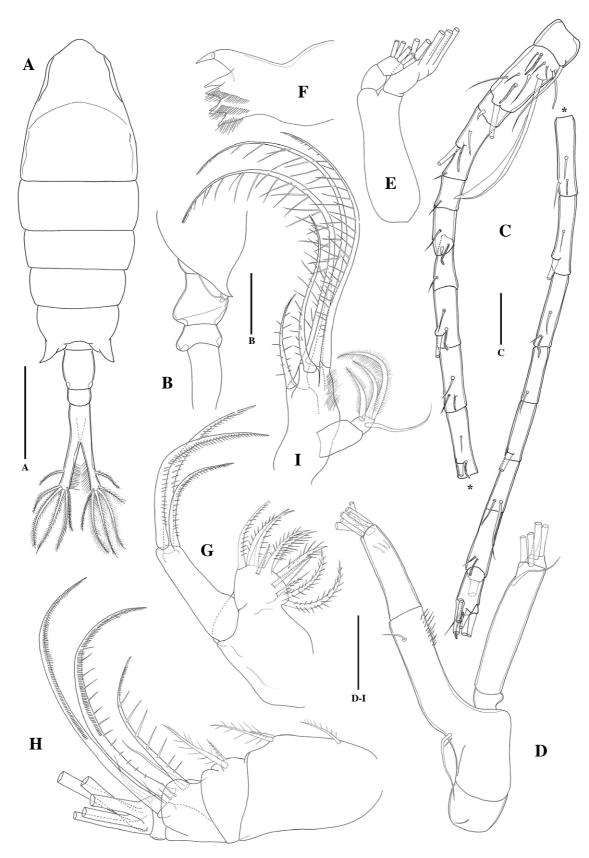


Fig. 4. *Tortanus (Eutortanus) komachi*, female. A. habitus, dorsal view. B. urosome, lateral view. C. antennule. D. antenna. E. palp of mandible. F. ganthobase of mandible. G. maxillule. H. maxilla. I. maxilliped. Scale bars: A=0.5 mm, B=0.25 mm, C-I=0.1 mm.

inwards into triangular process bearing 1 seta; exopod angularly curved inwards, bearing pointed process and 6 short spines on inner margin. Basis of left leg with outer seta subterminally; exopod 2-segmented, first segment with 1 short spine subterminally, second segment with 1 distal and 2 subdistal spines.

Distribution. China (Fenghsien), Korea (Incheon).

Remarks. Shen (1955) first reported *T. (E.) vermiculus* from the coastal water of Fenghsien, Kiangsu Province. Later, Chen and Zhang (1965) redescribed this species from the Amoy. Until now, this species has been known as showing a restrictive distribution along the southern part of the East China Sea (Ohtsuka *et al.*, 1992; Ohtsuka and Reid, 1998; Itoh *et al.*, 2001). The discovery of this species in Korean waters would expand the range of the hitherto known biogeography.

It is difficult to compare original specimen, because of poor description.

Tortanus (Eutortanus) vermiculus is readily distinguished from other congeners by: anal somite and right caudal ramus of female with uneven dorsolateral process,

and right exopod of leg 5 in male with long process along proximal inner margin.

The amplified DNA was 631 bp in length. The partial CO1 sequence of *T.* (*E.*) *vermiculus* was determined for the first time and registered at GenBank (Accession no: JN605791).

1*Tortanus (Eutortanus) komachi Itoh, Ohtsuka and Sato, 2001 (Figs. 4-6)

Tortanus (*Eutortanus*) *komachi* Itoh, Ohtsuka and Sato, 2001: 59, figs. 1-5.

Material examined. Korea: Gangwon-do: 5 ♂ ♂, Goseong-gun, Goseong port, 17 Apr 2011, BJ Lim; 208 ♂ ♂, 17 ♀ ♀, Gangneung-si, Gyeonso-dong, Gangneung port, 13 May 2011, SS Hong; 7 ♂ ♂, Cheongho-dong, Sokchosi, Cheongchoho, 12 Mar 2012, JH Song.

Description. Female: Body length 2.4 mm (Fig. 4A). Cephalosome and first pedigerous somite completely separated; fourth and fifth pedigerous somites incompletely fused. Posterior corners of last pedigerous somite asym-

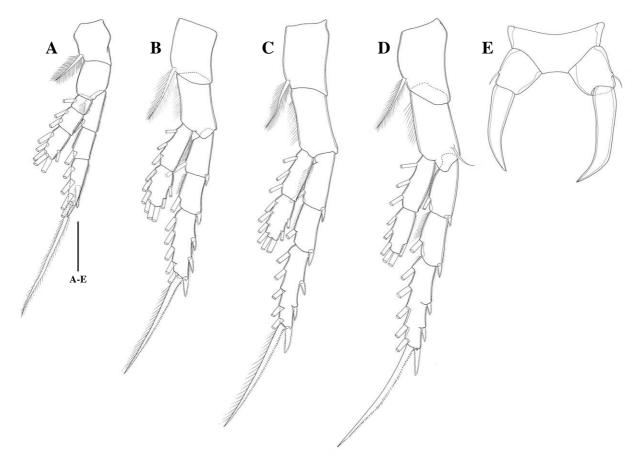


Fig. 5. *Tortanus (Eutortanus) komachi*, female. A. leg 1, posterior view. B. leg 2, posterior view. C. leg 3, posterior view. D. leg 4, posterior view. E. leg 5, anterior view. Scale bar: A-E=0.1 mm.

Korean name: 1*예쁜이부푼긴노벌레(신칭)

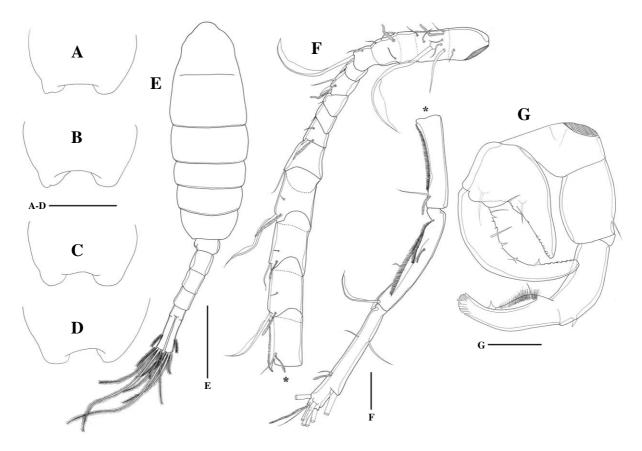


Fig. 6. *Tortanus* (*Eutortanus*) *komachi*, male. A-D. variability of prosomal end, dorsal view. E. habitus, dorsal view. F. right antennule. G. leg 5, anterior view. Scale bars: A-D=0.25 mm, E=0.5 mm, F-G=0.1 mm.

metrical, produced posterolaterally into triangular process.

Urosome 3-segmented (Fig. 4A, B), genital double somite about 2 times as long as second one; third segment completely fused with caudal rami. Caudal rami asymmetrical; both ramus almost same in length.

Antennule (Fig. 4C), antenna (Fig. 4D), mandible (Fig. 4E, F), maxillule (Fig. 4G), and maxilliped (Fig. 4I) similar to those of *T. (E.) vermiculus*. Maxilla (Fig. 4H) well developed; first and second praecoxal endites with 1 and 2 setae, respectively; first and second coxal endites with 2 and 3 setae, respectively; basal endite with 1 long and 2 short setae; endopod with 5 long and 2 rudimentary setae. Armature formula of legs 1 to 4 as in *T. (E.) vermiculus* (Fig. 5A-D).

Leg 5 (Fig. 5E) 3-segmented; basis with seta on outer margin; exopod 1-segmented, left exopod slightly longer than right; endopod absent.

Male: Body length 2.3 mm (Fig. 6E). Prosome about 1.8 times as long as urosome. Cephalosome and first pedigerous somite completely separated; fourth and fifth pedigerous somites incompletely fused. Posterior corners of last pedigerous somite asymmetrical, produced postero-

laterally into blunt process (Fig. 6A-D).

Urosome 5-segmented, second to fourth segments subequal in length; distal segment incompletely fused with caudal rami. Caudal rami asymmetrical, right ramus slightly longer than left.

Right antennule (Fig. 6F) 15-segmented and geniculate; ancestral segments I to VIII, XXI to XXIII, and XXIV to XXVIII completely or incompletely fused. Armature as follows; I-VIII, 11+2ae; IX, 2; X, 1; XI, 2+ae; XII, 1; XIII, 1; XIV, 2+ae; XV, 1; XVI, 2+ae; XVII, 2+ae; XVIII, 2+ae; XIX, 1+ae+process; XX, 1+ae+process; XXI-XXIII, 2+ae+2 processes; XXIV-XXVIII, 10+2ae.

Right leg 5 (Fig. 6G) 2-segmented; basis produced inwards into a long isosceles triangular process with 2 setae on posterior surface; exopod with 2 long and 4 short spines along inner margin. Left leg 2-segmented; first segment with a seta on outer margin; second and third segments incompletely fused and with 1 medial spine on outer margin, 3 spines on inner margin and 1 terminal short spine.

Distribution. Korea (East Sea), Japan (south coast of Akita).

Remarks. The Korean specimens coincide well with the

Japanese specimen of the original description with the following two minor differences: (1) the fifth pedigerous somite of male produced posteriorly into blunt process in the Japanese specimen, but the present specimens display a slightly sharp process; (2) right caudal ramus is slightly longer than left one in the original specimen, but the caudal rami of the present Korean specimen are almost same in length.

The amplified DNA was 614 bp in length. The partial CO1 sequence of T. (E.) komachi was determined for the first time and registered at GenBank (Accession no: JN605792).

Key to the species of subgenus Eutortanus from Korea

Korea
1. Urosome 3-segmented2 (female)
Urosome 5-segmented6 (male)
2. Female right caudal ramus unarmed on right side $\cdots 3$
Female right caudal ramus having uneven dorso-
lateral process on anterior right side
T. vermiculus
3. Female anal somite with spiniform process on right
side
Female anal somite without spiniform process4
4. Third urosomal segment and caudal rami completely
fused
Third urosomal segment and caudal rami completely
separated
5. Anal somite with anterolateral process
T. dextrilobatus
Anal somite without anterolateral process
·····T. derjugini
6. Right exopod of leg 5 with a pointed process on inner
proximal margin
Right exopod of leg 5 without a pointed process on
inner proximal margin7
7. Right exopod of leg 5 with not extending beyond in-
ner proximal process on basis T. spinicaudatus
Right exopod of leg 5 with exceeding beyond inner
proximal process on basis8
8. Basis of right leg 5 produced inwards into a long iso-
sceles triangular process
Basis of right leg 5 not produced inwards into a long
isosceles triangular process9
9. Basis of right leg 5 relatively narrower T. derjugini
Basis of right leg 5 relatively wider
T. dextrilobatus

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