

First Record of *Eucephalobus oxyuroides* (Nematoda: Rhabditida: Cephalobidae) from South Korea

Taeho Kim¹, Yeon Jae Bae¹, Joong-Ki Park^{2,*}

¹Division of Environmental Science and Ecological Engineering, College of Life Sciences and Biotechnology, Korea University, Seoul 02841, Korea

²Division of EcoScience, Ewha Womans University, Seoul 03760, Korea

ABSTRACT

Eucephalobus oxyuroides (de Man, 1876) Steiner, 1936 belonging to the family Cephalobidae Filipjev, 1934 (Cephalobomorpha) is newly reported from South Korea. This species is distinguished from other *Eucephalobus* species by its pointed tail terminus, high lips and relatively longer female tail and body. From many previous studies, intraspecific variation in this species has been reported for body length, absence/presence of probolae, and number/position of papillae in the posterior region of male, posing an obstacle for identification and phylogenetic analysis. In this study, details of the morphological characters and morphometric of *E. oxyuroides* Korean population are described and illustrated based on optical microscopy.

Keywords: Nematoda, Cephalobidae, *Eucephalobus oxyuroides*, new record, South Korea

INTRODUCTION

Members of the genus *Eucephalobus* Steiner, 1936 are bacterial feeders and widely distributed almost everywhere in the world except Antarctica. Morphological and morphometric variation (such as absence/presence of probolae, number of lips, and tail form) in this genus have been reported from many earlier studies (Anderson and Hooper, 1971; Boström, 1993a, 1993b; Abolafia and Peña-Santiago, 2002). This is an obstacle to correct identification and phylogenetic studies on this group. The genus *Eucephalobus* has been previously reported in South Korea, but has never been studied for species level taxonomy (Kim et al., 2012).

During a survey of several plots of strawberry farmland, *E. oxyuroides* (De Man, 1876) Steiner, 1936, were isolated from soil samples and identified as a new record in Korea. Here we provide detailed descriptions of the morphological characters and morphometrics for the Korean isolate of *E. oxyuroides*.

To extract nematode specimens, we sieved the soil samples and used the Baermann funnel method (Baermann, 1917) over two days. The nematode specimens were trans-

ferred to a 15 mL tube containing 2 mL water, to which was added 4 mL of 80°C TAF (2% triethanolamine and 7% formaldehyde) solution. The fixed nematodes were processed to dehydrated glycerin using Seinhorst's (1959) method and mounted in glycerin on HS-slides (Shirayama et al., 1993). Under an optical microscope (BX-51; Olympus, Tokyo, Japan) equipped with differential interference contrast (DIC), morphological characters of nematode specimens were observed and measured using a CoolSnap Photometrics color CCD digital camera (MP5.0-RTV-R-CLR-10; Photometrics, Tucson, AZ, USA) and the program QCapture Pro 5 (QImaging, Surrey, Canada).

SYSTEMATIC ACCOUNTS

Order Rhabditida Chitwood, 1933
Suborder Tylenchina Thorne, 1949
Infraorder Cephalobomorpha De Ley and Blaxter, 2002
Family Cephalobidae Filipjev, 1934
¹*Genus *Eucephalobus* Steiner, 1936

Korean name: ¹*유럽두염선충속(신칭)

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*To whom correspondence should be addressed
Tel: 82-2-3277-5948, Fax: 82-2-3277-2385
E-mail: jkpark@ewha.ac.kr

¹**Eucephalobus oxyuroides* (de Man, 1876) Steiner, 1936 (Table 1, Fig. 1)

Cephalobus oxyuroides: de Man, 1876: 140, fig. 26a, b.
Cephalobus (Eucephalobus) oxyuroides: Allgén, 1953: 672.
Eucephalobus oxyuroides: Steiner, 1936: 21; Goodey, 1963: 274; Andrassy, 1964: 241; 1967: 6; 1984: 172; Rashid et al., 1984: 279; Bongers, 1988: 209; Bussau, 1991: 119; Boström, 1993b: 296; Abolafia and Peña Santiago, 2002: 69.

Material examined. 10♀♀ and 10♂♂, South Korea: Chungcheongnam-do, Nonsan-si, Eunjin-myeon (GPS coordinates: 36°09'10.9"N, 127°06'55.7"E), 28 Apr 2016, extracted by sieving and the Baermann funnel method from strawberry farm soil. Two specimens (slide Nos. ZIIYIV0000001576 [female] and ZIIYIV0000001577 [male]) are deposited at the National Institute of Biological Resources, Republic of Korea. Eighteen specimens (slide Nos. 01010101001-01010101009 [females] and 01010101011-01010101019 [males]) are deposited in the Animal Phylogenomics Laboratory, Ewha Womans University, Republic of Korea.

Measurements. See Table 1.

Description. Adult: Body cylindrical, ventrally curved after fixation (Fig. 1A). Posterior body region of male more ventrally curved than female (Fig. 1B). Cuticle annulated; annuli 1.4–2.1 µm wide and 1.1–1.5 µm thick at mid-body. Lateral field occupying 12.5–36.8% of width of body at mid-body, with three incisures, fading out at phasmid region in female, outer two extending to tail terminus in male (Fig. 1D, F, G). Six lips, each with pointed terminus; lateral lips with labial papillae; subdorsal and subventral lips with cephalic papillae and a labial papillae (Fig. 1C). Transversal, oval-shaped amphidial apertures present. Stoma cephaloboid, 1.6–2.1 times lip region diameter; cheilorhabdions bar-shaped; gymnostom and stegostoma narrower than cheilostom (Fig. 1D). Pharyngeal corpus cylindrical, 2.7–3.6 times length of isthmus. Isthmus narrower than corpus. Basal bulb oval-shaped with developed valves; basal bulb 1.2–1.5 times as long as its width. Cardia conoid, not conspicuous, surrounded by intestinal tissue. Nerve ring located at posterior corpus, 45–60 annuli from head, at 58.7–65.6% of pharynx length. Excretory pore located at posterior extremity of corpus or corpus-isthmus junction region, 46–62 annuli from head, at 61.1–71.3% of pharynx length. Position of deirids in lateral field at isthmus level or corpus-isthmus junction (n = 1), 51–69 annuli from head, at 67.4–78.9% of pharynx length.

Female: Reproductive system monodelphic-prodelphic, dextral (Fig. 1E). Vulval lips slightly protruding in some individuals (n = 2). Post-vulval sac distinct, 0.8–1.3 body

diam. long. Uterus differentiated into two sections, together 1.7–2.8 times width of body. Spermatheca obvious, about 1.6–2.2 body width in length. Oviduct short. Ovary straight to posterior, with a single row of oocytes. Rectum 1.3–1.8 times as long as anal body diameter (Fig. 1F). Tail elongated-conoid, with 24–29 annuli. Phasmids at 23.3–33.8% of tail length.

Male: Reproductive system monorchic, dextral. Testis reflexed ventrad anteriorly (n = 9), dorsally in one individual. Spicules curved ventrad, 18.7–22.4 µm long (Fig. 1G). Gubernaculum wedge-shape, 42.4–53.4% of spicules length. Three pairs of precloacal subventral papillae present (one pair very close to anus level), two pairs papillae (one lateral and one subventral) at mid-tail, two pairs (one dorsosublateral and one subventral) close to tail terminus. Tail conoid, arcuate ventrad, with mucro (1.2–3.2 µm long). Phasmid openings at 22.3–40.3% of tail length.

Distribution. Korea, Arctic, Armenia, Australia, Austria, Azerbaijan, Belgium, Brazil, Bulgaria, China, Czechoslovakia, Denmark, Estonia, Great Britain, Hungary, Iceland, Israel, Italy, Kyrgyzstan, Latvia, Lithuania, Moldova, The Netherlands, Norway, Poland, Russia, Spain, Sweden, Switzerland, Tajikistan, Turkey, Turkmenistan, USA, Ukraine, Uzbekistan, Venezuela, Yugoslavia, Zaire.

Habitat. Soil from a strawberry farm.

Remarks. Morphological characters of the specimens described in this report generally agree with the original description of *E. oxyuroides* (de Man, 1876) Steiner, 1936 except for the body width (male a = 22.6–28.2 vs. 19.5–22.1). Nevertheless, the body width of the specimens are within the range of intraspecific variation reported in previous studies (Table 2). The body length of our specimens accords with the range of intraspecific variation of the materials described by some earlier studies (de Man, 1876; Andrassy, 1964, 1967, 1984; Rashid et al., 1984; Bongers, 1988; Abolafia and Peña-Santiago, 2002), but is considerably shorter than the specimens described by Allgén (1953), Boström (1993b), Bussau (1991) and Goodey (1963) (Table 2). The specimens observed in this study have six lips, which corresponds with majority of earlier descriptions of *E. oxyuroides* (Andrassy, 1967; Rashid et al., 1984; Boström, 1993b; Abolafia and Peña-Santiago, 2002). However, Boström (1984) described three bifurcate labial probolae as well as three pairs of lips (cephalic probolae) for *E. oxyuroides*. Our male specimens have a total of seven pairs of papillae (three pre-anal and four post-anal) in the posterior region (Fig. 1F). By contrast, males from described from Turkey had six pairs of papillae, and a Brazil population was depicted with nine pairs of papillae (Rashid et al., 1984; Boström,

Korean name: ¹*뽕족꼬리유럽두염선충(신칭)

Table 1. Morphometrics of *Eucephalobus oxyuroides*

	<i>Eucephalobus oxyuroides</i>	
	Females (n = 10)	Males (n = 10)
L	506.6±32.1 (472.8–579.4)	494.6±39.0 (443.2–547.0)
Body width	19.7±1.7 (17.6–23.6)	18.9±1.7 (16.6–21.6)
Lateral field width	3.9±0.7 (2.4–5.1)	4.6±1.1 (3.6–7.5)
Lateral field width/body width (%)	20.0±3.1 (12.5–22.6)	24.1±4.8 (20.7–36.8)
Pharynx length	140.5±5.1 (133.5–153.0)	137.8±5.9 (127.4–145.2)
Tail length ^a	50.7±2.4 (47.3–53.6)	39.0±3.2 (35.1–46.9)
Anal width	11.8±1.0 (10–13.6)	15.6±1.4 (13.2–17.3)
Tail annuli	25.9±1.9 (24–29)	–
a	25.7±1.0 (24.6–27.6)	26.3±1.7 (22.6–28.2)
b	3.6±0.2 (3.3–3.8)	3.6±0.2 (3.3–4.0)
c	10.0±0.6 (9.3–11.0)	12.8±1.6 (9.7–15.0)
c'	4.3±0.3 (3.8–4.8)	2.5±0.4 (2.1–3.5)
Lips region width	6.2±0.4 (5.9–7.1)	5.9±0.3 (5.5–6.5)
Stoma	11.2±0.3 (10.7–11.8)	10.9±0.9 (9.2–11.9)
Stoma diameter	2.4±0.1 (2.2–2.6)	2.4±0.4 (1.7–2.8)
Stoma/lips region width	1.8±0.1 (1.6–1.9)	1.9±0.1 (1.7–2.1)
Stoma/stoma diameter	4.8±0.2 (4.5–5.3)	4.7±0.8 (3.7–6.1)
Corpus	82.3±4.0 (78.0–91.9)	80.6±4.7 (72.5–87.7)
Isthmus	26.0±1.6 (23.1–27.9)	26.5±1.9 (23.3–30.6)
Basal bulb	16.7±0.6 (15.7–17.6)	15.7±1.1 (13.9–17.4)
Basal bulb width	12.4±0.6 (11.3–13.3)	11.9±1.0 (10.6–13.7)
Basal bulb/basal bulb width	1.4±0.1 (1.2–1.5)	1.3±0.1 (1.2–1.5)
Corpus:isthmus	3.2±0.2 (2.9–3.6)	3.1±0.3 (2.7–3.6)
Nerve ring	87.7±4.7 (81.3–99.0)	85.4±3.8 (80.3–92.0)
Excretory pore	90.5±4.8 (85.0–101.1)	90.1±5.1 (82–97.5)
Deirid	103.7±6.0 (96.1–117.0)	102.2±7.1 (89.7–111.0)
Nerve ring (% pharynx)	62.4±2.1 (58.7–65.6)	62.0±1.7 (59.7–64.4)
Excretory pore (% pharynx)	64.4±3.0 (61.1–71.3)	65.4±2.4 (61.3–68.2)
Deirid (% pharynx)	73.8±3.1 (69.2–77.7)	73.8±3.7 (67.4–78.9)
Rnr ^b	50.5±4.1 (45–57)	51.0±4.9 (47–60)
Rep ^c	51.7±4.8 (46–60)	53.2±4.8 (48–62)
Rdei ^d	58.5±5.2 (51–67)	59.9±5.5 (53–69)
Phasmid	14.1±1.8 (11.2–16.0)	12.2±1.8 (8.9–14.7)
Phasmid (% tail)	27.8±3.7 (23.3–33.8)	31.5±6.0 (22.3–40.3)
Cuticle thickness	1.3±0.1 (1.1–1.5)	1.3±0.1 (1.1–1.5)
Annuli width	1.8±0.1 (1.7–2.0)	1.8±0.3 (1.4–2.1)
Vulva from anterior end	316.4±19.9 (296.6–361.2)	–
V (%)	62.5±0.4 (62.0–63.2)	–
Vulva-anus	137.4±10.9 (126.2–163.3)	–
Vulva-anus/tail	2.7±0.2 (2.5–3.1)	–
Reproductive tract length	164.9±10.3 (153.3–182.1)	–
G (%)	32.6±2.2 (30.6–37.1)	–
Vagina	7.7±1.0 (6.5–9.9)	–
Post-vulval sac	22.8±2.7 (16.7–26.4)	–
Uterus	41.4±8.3 (30.5–60.1)	–
Oviduct	10.7±1.5 (8.1–12.9)	–
Spermatheca	35.6±3.3 (30.6–41.2)	–
Ovary	121.8±14.2 (101.4–150.5)	–
Vagina/body width	0.4±0.0 (0.3–0.5)	–
Post-vulval sac/body width	1.2±0.2 (0.8–1.3)	–
Uterus/body width	2.1±0.3 (1.7–2.8)	–
Oviduct/body width	0.5±0.1 (0.4–0.7)	–
Spermatheca/body width	1.8±0.2 (1.6–2.2)	–
Ovary/body width	6.2±0.6 (5.1–7.1)	–
Rectum	18.2±1.6 (15.8–21.4)	23.4±1.8 (19.7–25.7)
Rectum/anal width	1.5±0.2 (1.3–1.8)	1.5±0.2 (1.1–1.8)
Spicule	–	20.5±1.1 (18.7–22.4)
Gubernaculum	–	9.8±0.9 (8.4–10.9)
Gubernaculum/spicules (%)	–	47.7±3.7 (42.4–53.4)
Spicule/anal body width	–	1.3±0.1 (1.2–1.5)
Spicule/tail length	–	0.5±0.1 (0.4–0.6)
Mucro	–	2.2±0.7 (1.2–3.2)

All measurements are in μm and in the form: mean±SD (range).

^aNumber of annuli from anus to the tail end.

^bNumber of annuli from anterior end to the nerve ring.

^cNumber of annuli from anterior end to the excretory pore.

^dNumber of annuli from anterior end to the deirid.

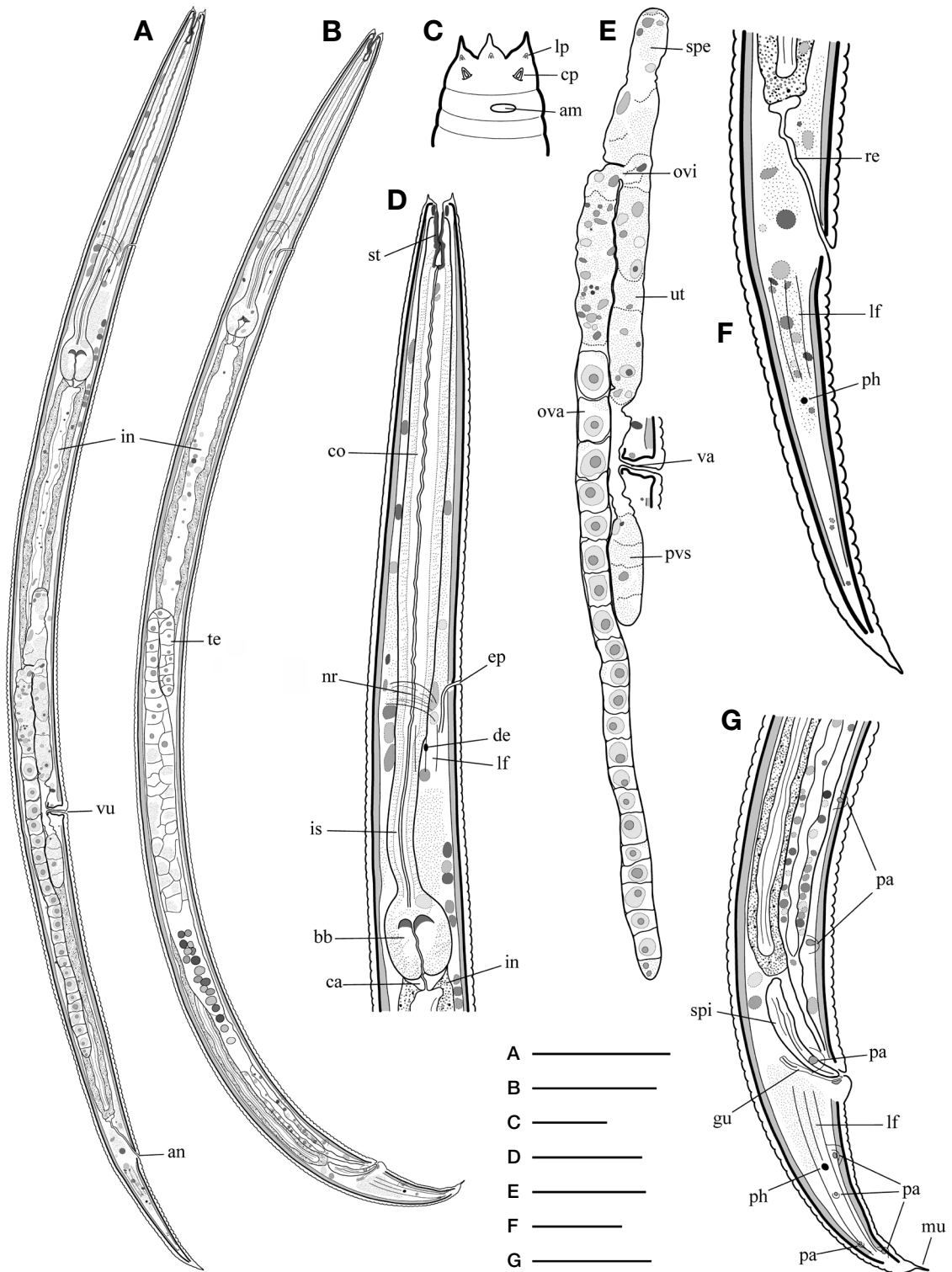


Fig. 1. *Eucephalobus oxyuroides* (de Man, 1876) Steiner, 1936. A, Entire female; B, Entire male; C, Female head region; D, Female neck region; E, Female reproductive system; F, Female posterior region; G, Male posterior region. am, amphid; an, anus; bb, basal bulb; ca, cardia; co, corpus; cp, cephalic papilla; de, deirid; ep, excretory pore; gu, gubernaculum; in, intestine; is, isthmus; lf, lateral field; lp, labial papilla; mu, mucro; nr, nerve ring; ova, ovary; ovi, oviduct; pa, papilla; ph, phasmid; pvs, post-vulval sac; re, rectum; spe, spermatheca; spi, spicule; st, stoma; te, testis; ut, uterus; va, vagina; vu, vulva. Scale bars: A, B=50 μ m, C=5 μ m, D, E, G=20 μ m, F=10 μ m.

Table 2. Morphometrics variability among *Eucephalobus oxyuroides* (de Man, 1876) Steiner, 1936

	L	a	b	c	c'	V (%)	Corpus: isthmus	Nerve ring (% pharynx)	Excretory pore (% pharynx)	Deirid (% pharynx)	Phasmid (% tail)
This study	♀ 472-580	24.6-27.6	3.3-3.8	9.3-11.0	3.8-4.8	62-64	2.9-3.6	81.3-99.0 (58.7-65.6)	85.0-101.1 (61.1-71.3)	96.1-117.0 (69.1-77.7)	11.2-16.0 (23.3-33.8)
	♂ 443-547	22.6-28.2	3.3-4.0	9.7-15.0	2.1-3.5	-	2.7-3.6	80.3-92.0 (59.7-64.4)	82.0-97.5 (61.3-68.2)	89.7-111.0 (67.4-78.9)	8.9-14.7 (22.3-40.3)
Abolafia and Peña-Santiago (2002)	♀ 524-535	21.4-27.6	1.7-3.5	8.4-8.9	4.2-5.3	60-64	2.6-3.3	82-92 (54-64)	81-97 (54-67)	100 (66)	17-18 (26-28)
Allgén (1953)	♀ 800	38.1	3.8	12.3	-	-	-	- (-)	- (-)	- (-)	- (-)
	♂ 787	35.8	4.2	15.7	-	-	-	- (-)	- (-)	- (-)	- (-)
Andrássy (1964)	♀ 540	20	3.4	10.7	3.6	60.3	-	- (-)	- (-)	- (-)	- (-)
	♂ 490	20	2.6	13.5	2.2	-	-	- (-)	- (-)	- (-)	- (-)
Andrássy (1967)	♀ 450-500	20-23	4.0-4.2	8.5-9.5	4-5	60-62	3.5-4	- (-)	- (74-77)	- (-)	- (35-40)
Andrássy (1984)	♀ 450-700	18-25	3-5	7-11	4-5	56-66	3.5-4	- (-)	- (-)	- (-)	- (-)
	♂ 500-600	18-25	3-4	12-14	-	-	3.5-4	- (-)	- (-)	- (-)	- (-)
Bongers (1988)	♀ 450-700	18-25	3-5	7-11	4-5	56-66	-	- (-)	- (-)	- (-)	- (-)
	♂ 500-600	18-25	3-4	12-14	-	-	-	- (-)	- (-)	- (-)	- (-)
Boström (1993b)	♀ 609	23	4	8.7	4.4	63	3.5	102 (-)	104 (-)	121 (-)	- (28)
	♂ 525	28	3.9	16.4	1.9	-	3.1	90 (-)	94 (-)	- (-)	- (58)
Bussau (1991)	♀ 745	24	4	9	-	61	-	- (-)	- (-)	- (-)	- (-)
	♂ 643	23	3.6	13	-	-	-	- (-)	- (-)	- (-)	- (-)
de Man (1876)	♂ 447-514	19.5-22.1	3.4-3.7	10.4-11.9	2.5-2.7	-	-	- (-)	82.4-86.3 (64.7-65.6)	- (-)	- (-)
Goodey (1963)	♀ 650	19	3.9	8.3	-	60	-	- (-)	- (-)	- (-)	- (-)
	♂ 550	21	3.5	12	-	-	-	- (-)	- (-)	- (-)	- (-)
Rashid et al. (1984)	♀ 370-650	18.0-32.0	3.3-5.0	5.0-8.5	-	55-63	-	- (-)	67-108 (-)	- (-)	23-30 (-)
	♂ 390-600	21.0-35.0	3.2-4.2	14.6-19.8	-	-	-	- (-)	- (-)	- (-)	- (-)

1993b). Thorne (1937, 1961) described seven papillae in males; however, the positions of those papillae were different, when compared to our specimens: two pairs of pre-anal papillae and five post-anal papillae.

Vast intraspecific variability in morphological characters has been reported in *Eucephalobus striatus* (Bastian, 1865) Thorne, 1937, specifically in the cephalic and labial proboscis structure, tail shapes and several measurements (Anderson and Hooper, 1971). This variability may lead taxonomic error in species identification and phylogenetic studies, and careful reevaluation for morphological features in the genus *Eucephalobus* is further required.

ACKNOWLEDGMENTS

This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR201601201) and the Marine Biotechnology Program (20170431) funded by the Ministry of Oceans and Fisheries, Korea.

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Received August 29, 2017
Revised September 21, 2017
Accepted September 26, 2017