First report and morphological description of two *Acrobeloides* species (Nematoda: Rhabditida: Cephalobidae) in South Korea

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The genus Acrobeloides (Cobb, 1924) Thorne, 1937 are bacterial feeders and are one of the most abundant and widely distributed nematode groups in various terrestrial environments. Based on morphological and morphometric analyses, we found two Acrobeloides species reported in Korea for the first time: A. bodenheimeri (Steiner, 1936) Thorne, 1937 and A. tricornis (Throne, 1925) Thorne, 1937. These species exhibit morphological characters concordant with typical features of the genus Acrobeloides, such as a fusiform pharyngeal corpus with swollen metacorpus and lateral incisures extending to the tail terminus. However, A. bodenheimeri is distinguished from other acrobeloids by having its low and rounded labial probolae, distinct post-uterine sac and five lateral incisures. Acrobeloides tricornis is distinguished from its congeners by the following characteristics: its high labial probolae with acuate termini, inconspicuous post-uterine sac and five lateral incisures. Morphological characters and their measurements, and illustrations of A. bodenheimeri and A. tricornis are described in this study.

Keywords: Acrobeloides bodenheimeri, Acrobeloides tricornis, Cephalobidae, Nematoda, South Korea

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Introduction

Members of the genus Acrobeloides (Cobb, 1924) Thorne, 1937, belonging to the family Cephalobidae, are bacterial feeders and are one of the most abundant and widely distributed nematode groups in various terrestrial environments, such as agricultural land (Pervez, 2011), forest (Háněl, 1999), highlands (Boström, 1993) and sand dunes (Yeates, 1967). Morphological and morphometric variations (such as body size, nerve ring and excretory position and tail shape) in this group have been reported from previous studies (Anderson, 1965; 1968; De Ley et al., 1999; Abolafia and Peña-Santiago, 2003). These morphological variations often hinder species delimitation and identification that mislead species-level taxonomy. To date, 40 nominal species have been reported in this genus; however, only 29 species may be accepted as valid (Andrássy, 2005), including two species previously reported in Korea (Kim et al., 2016; 2017): A. nanus (de Man, 1880) Anderson, 1968 and A. varius Kim, Kim and Park, 2017.

Following a survey of plots of farmland and overgrown fields, *A. bodenheimeri* (Steiner, 1936) Thorne, 1937 and *A. tricornis* (Thorne, 1925) Thorne, 1937 were isolated

from soil samples. In this paper, we provide a detailed description of the morphological characters and morphometrics, and illustrations of these two species.

MATERIALS AND METHODS

Nematode isolation

Soil samples were collected from a potato farm (Bukmyeon, Uichang-gu, Changwon-si, Gyeongsangnam-do, South Korea [GPS coordinates: 35°22′22.5″N, 128°36′47.3″E]) and overgrown fields (Mechuri Island, Daenam-ro, Danwon-gu, Ansan-si, Gyeonggi-do, South Korea [GPS coordinates: 37°12′02.9″N, 126°32′24.6″E]). Nematode specimens were extracted from soil samples using the Baermann funnel method (Baermann, 1917).

Sample processing and morphological observations

For fixation, the nematode specimens were transferred 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 dehydra-

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Table 1. Morphometric measurements of *Acrobeloides bodenheimeri* and *A. tricornis*.

Character	Acrobeloides bodenheimeri ${\varphi, n=1}$	Acrobeloides tricornis ♀, n = 1	Character	Acrobeloides bodenheimeri	Acrobeloides tricornis
				♀, n = 1	♀, n = 1
L	529.5	332.0	Excretory pore position (% pharynx)	63.8	79.7
a	27.2	17.5	Deirid position (% pharynx)	71.5	86.9
b	3.4	3.3	Vulva from anterior end	369.6	220.2
c	19.3	15.9	Vulva to anus	133.6	87.8
c'	2.0	1.9	Vulva to anus/tail length	4.9	4.2
V	69.8	66.3	Reproductive tract length	207.0	95.2
G	39.1	28.7	Vagina	12.2	5.1
Body diameter	19.5	19.0	Post-uterine sac	50.3	_
Pharynx length	156.3	99.8	Uterus	62.1	34.4
Tail length	27.4	20.8	Spermatheca	36.9	17.6
Anal body diameter	13.7	10.8	Oviduct	11.0	13.8
Tail annuli ^a	14	15	Ovary	155.4	85.9
Lips region diameter	7.5	6.1	Vagina/body diameter	0.6	0.3
Stoma	12.2	10.7	Post-uterine sac/body diameter	2.6	_
Stoma diameter	4.3	4.2	Uterus/body diameter	3.2	1.8
Stoma/lips region diameter	1.6	1.8	Spermatheca/body diameter	1.9	0.9
Stoma/stoma diameter	2.8	2.6	Oviduct/body diameter	0.6	0.7
Corpus	91.3	52.0	Ovary/body diameter	8.0	4.5
Isthmus	29.3	19.5	Rectum	19.9	12.9
Basal bulb	18.7	12.8	Rectum/anal body diameter	1.5	1.2
Basal bulb diameter	13.5	10.6	Anus to phasmid	13.2	9.7
Basal bulb length/diameter	1.4	1.2	Phasmid position (% tail)	48.1	46.3
Corpus:isthmus ratio	3.1	2.7	Lateral field width	4.3	4.5
Nerve ring to anterior end	99.6	70.7	Lateral field width/body diameter (%)	22.2	23.9
Excretory pore to anterior end	99.7	79.5	Cuticle thickness	1.0	0.7
Deirid to anterior end	111.8	86.7	Annuli width	2.0	1.4
Nerve ring position (% pharynx)	63.7	70.9			

All measurements are in µm.

ted using Seinhorst's (1959) method and mounted in pure glycerin on HS slides (Shirayama *et al.*, 1993). Under an optical microscope (BX-51; Olympus, Tokyo, Japan) equipped with differential interference contrast (DIC), morphological and morphometric characters of the nematode specimens were observed and measured using a Cool Snap 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 Genus *Acrobeloides* (Cobb, 1924) Thorne, 1937

Acrobeloides bodenheimeri (Steiner, 1936) Thorne, 1937 (Table 1, Fig. 1)

뭉뚝꼬리뾰족입술선충(신칭)

Acrobeles bodenheimeri: Steiner, 1936: 77, fig. 24. Cephalobus bodenheimeri: Andrássy, 1984: 170. Rafiqius bodenheimeri: Khan and Hussain, 1997: 140. Acrobeloides rotundifolius: Bussau, 1991: 122, abb. 7. Acrobeloides bodenheimeri: Thorn, 1937: 11.

Material examined. 1♀, Buk-myeon, Uichang-gu, Chang-won-si, Gyeongsangnam-do, South Korea (GPS coordi-

^aNumber of annuli from anus to the tail end.

L, body length/body diameter; b, body length/Pharynx length; c, body length/tail length; c', tail length/diameter at anus region; V, % distance of vulva from anterior end/body length; G, % reproductive tract length/body length.

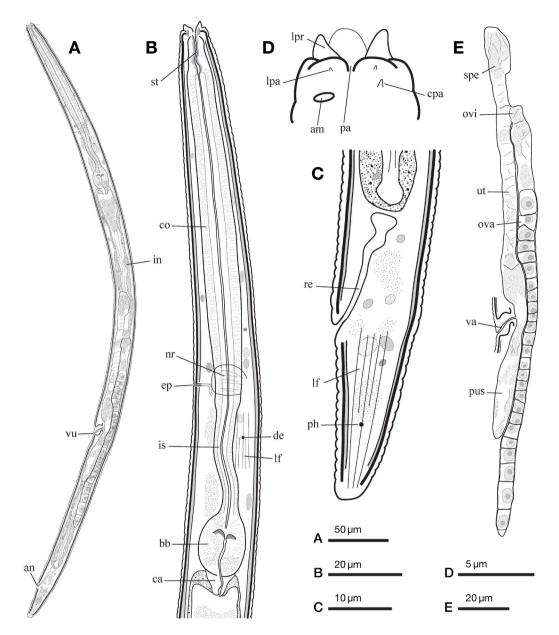


Fig. 1. Acrobeloides bodenheimeri (Steiner, 1936) Thorne, 1937. A, Entire female; B, Female neck region; C, Female posterior region; D, Female head region; E, Female reproductive system. am, amphid; an, anus; bb, basal bulb; ca, cardia; co, corpus; cpa, cephalic papilla; de, deirid; ep, excretory pore; in, intestine; is, isthmus; lf, lateral field; lpa, labial papilla; lpr, labial probolae; nr, nerve ring; ovi, oviduct; ova, ovary; pa, primary axil; ph, phasmid; pus, post-uterine sac; re, rectum; spe, spermatheca; st, stoma; ut, uterus; va, vagina; vu, vulva.

nates: 35°22′22.5″N, 128°36′47.3″E), extracted by sieving and the Baermann funnel method from potato farm soil. The specimen (slide No. NIBRIV0000862892) is deposited at the National Institute of Biological Resources, South Korea.

Measurements. See Table 1.

Description. Female: Body cylindrical, length 529.5 μ m long, ventrally curved after fixation (Fig. 1A). Cuticle annulated; annuli 2.0 μ m wide and 1.0 μ m thick at midbody. Lateral field occupying 22.2% of width of body at

mid-body. Lateral incisures varying in number along body length: three incisures between procorpus region and deirid, branching off from deirid into five incisures (Fig. 1B); two incisures fading out between anus and phasmid region; three incisures extending to tail terminus (Fig. 1C). Head region continuous with neck. Lip region 7.5 µm in diameter, with triradiate symmetry with 6+4 papillae (Fig. 1D). Three pairs of asymmetrical lips; pairs of lips separated by U-shaped primary axils. Guarding process absent. Cephalic probolae absent. Three low and rounded

labial probolae present. Stoma cephaloboid; length 1.6 times the lip region diameter (Fig. 1B). Cheilorhabdions oval-shaped. Small dorsal denticle on metastom. Pharyngeal corpus fusiform with swollen metacorpus, 3.1 times isthmus length. Isthmus narrower than corpus, distinctly demarcated from metacorpus. Basal bulb oval-shaped with well-developed valves; 1.4 times as long as its width. Cardia inconspicuous, surrounded by intestinal tissue. Nerve ring located in posterior corpus, at 63.7% of pharynx length. Excretory pore position at posterior corpus, at 63.8% of pharynx length. Position of deirids in lateral field at anterior isthmus, at 71.5% of total neck length. Reproductive system monodelphic-prodelphic (Fig. 1E). Vulva lips not protruding. Vagina length 0.6 times body diameter. Post-uterine sac 2.6 times body width. Uterus length 3.2 times body diameter. Spermatheca 1.9 times body width. Oviduct short. Ovary straight, with a single row of oocytes. Rectum length 1.5 times anal body diameter (Fig. 1C). Tail conoid, with truncated terminus. Phasmids located middle of tail, at 48.1% of tail length.

Male: Unknown.

Distribution. Denmark (Bussau, 1991), Israel (Steiner, 1936), Mongolia (Andrássy, 1967), Malawi (Siddiqi *et al.*, 1992), South Korea (this study), Spain (Abolafia and Peña-Santiago, 2003), and USA (De Ley *et al.*, 1999).

Habitat. Soil sample from a potato farm.

Remarks. Morphological characters of the specimen described in this study generally agree with previous studies (Steiner, 1936; Siddiqi et al., 1992; De Ley et al., 1999; Abolafia and Peña-Santiago, 2003), except for total body length (529 vs 613–1,530 μm), the ratio of body length to body width (a = 27.2 vs 15.0-24.3), the ratio of total body length to pharynx length (b = 3.4 vs 4.0-7.8) and corpus to isthmus ratio (3.1 vs 3.3-7.2). Although these morphometric measurements of the specimen examined from a single individual differ from A. bodenheimeri, they are considered intraspecific variations as previously reported from many other Acrobeloides species (Anderson, 1965; 1968; Boström and Gydemo, 1983; De Ley et al., 1999; Abolafia and Peña-Santiago, 2003). Morphometric analysis using multiple nematode individuals is further required to delimitate morphological variation of the species.

Acrobeloides tricornis (Thorne, 1925) Thorne, 1937 (Table 1, Fig. 2)

삼각뾰족입술선충(신칭)

Acrobeles tricornis: Thorne, 1925: 189, figs. 13. Acrobeloides setosus: Brzeski, 1962: 337, figs. 7–9. Acrobeloides uberrinus: Anderson, 1965: 232, fig. 1. Acrobeloides syrtisus: Yeates, 1967: 530, fig. 2. Acrobeloides tricornis: Throne, 1937: 11.

Material examined. 1♀, Mechuri Island, Daenam-ro, Danwon-gu, Ansan-si, Gyeonggi-do, South Korea (GPS

coordinates: 37°12′02.9″N, 126°32′24.6″E), extracted by sieving and the Baermann funnel method from overgrown field soil. The specimen (slide No. ZCIVIV0000003207) is deposited at the National Institute of Biological Resources, South Korea.

Measurements. See Table 1.

Description. Female: Body cylindrical, length 332.0 µm long, ventrally curved after fixation (Fig. 2A). Cuticle annulated; annuli 1.4 µm wide and 0.7 µm thick at midbody. Lateral field occupying 23.9% of width of body at mid-body. Lateral incisures varying in number along body length: three incisures before deirid, branching off from deirid into five incisures (Fig. 2B); two incisures fading out at anus level; three incisures extending to tail terminus (Fig. 2C). Head region continuous with neck. Lip region $6.1 \, \mu m$ in diameter, with triradiate symmetry with 6+4papillae (Fig. 2D). Three pairs of asymmetrical lips; pairs of lips separated by U-shaped primary axils. Secondary axils shallow. Guarding process absent. Cephalic probolae absent. Three labial probolae high, with conical basal part and acuate distal part. Stoma cephaloboid, length 1.8 times lip region diameter (Fig. 2B). Cheilorhabdions ovalshaped, strongly cuticularized. Pharyngeal corpus fusiform with swollen metacorpus, 2.7 times isthmus length. Isthmus narrower than corpus, distinctly demarcated from metacorpus. Basal bulb oval-shaped with well-developed valves; 1.2 times as long as its width. Cardia surrounded by intestinal tissue. Nerve ring located at anterior isthmus, at 70.9% of pharynx length. Excretory pore position at posterior isthmus, at 79.7% of pharynx length. Position of deirids in lateral field at basal bulb level, at 86.9% of total neck length. Reproductive system monodelphic-prodelphic (Fig. 2E). Vulva lips not protruding. Vagina length 0.3 times body diameter. Post-uterine sac inconspicuous. Uterus length 1.8 times body diameter. Spermatheca 0.9 times body width. Oviduct short. Ovary straight. Rectum length 1.2 times anal body diameter (Fig. 2C). Tail conoid, with truncated terminus. Phasmids located middle of tail, at 46.3% of tail length.

Male: Unknown.

Distribution. Austria, Bulgaria, Canada, France, Georgia, Germany (Andrássy, 1984), Hungary (Andrássy, 2005), Kazakhstan, Kyrgyzstan, Lithuania, The Netherlands (Andrássy, 1984), New Zealand (Yeates, 1967; Boström and Holovachov, 2010), Norway (Spitzbergen) (Loof, 1971; Boström, 1987), Poland (Brzeski, 1962), Russia (Andrássy, 1984), Senegal (De Ley et al., 1990; Boström and Holovachov, 2010), South Korea (this study), Spain (Abolafia and Peña-Santiago, 2003), USA (Thorne, 1925; Anderson, 1965), Tajikistan (Andrássy, 1984), Turkey (Boström, 1993), Turkmenistan, and Uzbekistan (Andrássy, 1984).

Habitat. Soil from an overgrown field.

Remarks. The specimen here described generally matches

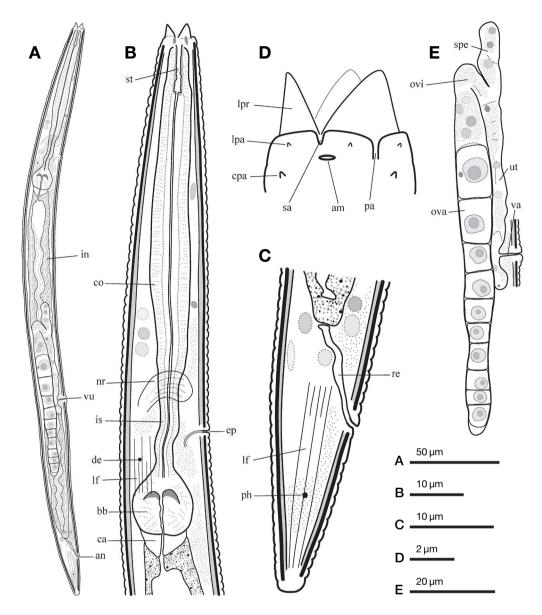


Fig. 2. Acrobeloides tricornis (Thorne, 1925) Thorne, 1937. A, Entire female; B, Female neck region; C, Female posterior region; D, Female head region; E, Female reproductive system. am, amphid; an, anus; bb, basal bulb; ca, cardia; co, corpus; cpa, cephalic papilla; de, deirid; ep, excretory pore; in, intestine; is, isthmus; If, lateral field; lpa, labial papilla; lpr, labial probolae; nr, nerve ring; ovi, oviduct; ova, ovary; pa, primary axil; ph, phasmid; re, rectum; sa, secondary axil; spe, spermatheca; st, stoma; ut, uterus; va, vagina; vu, vulva.

with morphological features that previously studied for *A. tricornis* (Thorne, 1925; Loof, 1971; Boström, 1993). *Acrobeloides setosus* Brezeski, 1962, *A. syrtisus* Yeates, 1967 and *A. uberrinus* Anderson, 1965 were treated as synonyms of *A. tricornis* by Boström and Holovachov (2010). The examined specimen in this study is morphologically very similar to *A. uberrinus*, a junior synonym of *A. tricornis*, in the longitudinal incisures in the lateral field with three incisures extending to tail terminus (Anderson, 1965; De Ley *et al.*, 1990) (Fig. 2C). Further examination of multiple nematode individuals is needed to pro-

vide a species delimitation of their morphological variations.

The two newly reported *A. bodenheimeri* and *A. tricornis* species described herein are distinguished by shape of labial probolae (low and rounded vs high with acuate distal part), post-uterine sac (distinct vs inconspicuous) and some morphometric characters (such as body length [529.5 vs 332.0] and the ratio of body length to body width [a=27.2 vs 17.5]). In this study, we were not able to find male specimens of both species and species identification of nematodes was conducted based purely on morphologi-

cal characters examined from a single female individual, which is very common in soil nematode taxonomy. Moreover, morphological characters from male representatives in many *Acrobeloides* species (such as *A. apiculatus*, *A. nanus* and *A. varius*) have not yet been documented. Although a single female specimen was examined from each of the two species, their morphological characters described herein generally agree with those of *A. bodenheimeri* and *A. tricornis*.

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