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Description of *Pseudacrobeles* (*Pseudacrobeles*) *curvatus* sp. n. (Cephalobidae: Rhabditida) in South Korea

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Abstract: Pseudacrobeles (*Pseudacrobeles*) *curvatus* sp. n. was collected from potato fields in Gyeongsangnam-do, South Korea. The new species shares morphological characters typical of the genus *Pseudacrobeles*, including three lateral incisures that fade posteriorly near the phasmid openings. The new species differs from other *Pseudacrobeles* species by its smaller body size and a comparatively shorter corpus relative to the isthmus length. In this study, we provide a comparison of morphometrics and diagnostic features of *Pseudacrobeles* species and molecular sequence data from the D2-D3 regions of the 28S ribosomal DNA (rDNA) and ITS1-5.8S-ITS2 region of rDNA from the new species, which can be used as molecular barcode sequences.

Key words: Cephalobidae, *Pseudacrobeles*, morphology, SEM, molecular sequence, new species.

The genus *Pseudacrobeles* was erected to include a single species, *Pseudacrobeles variabilis* Steiner, 1938, which was formerly *Acrobeles variabilis* Steiner, 1936. During a taxonomic revision of the genus, De Ley et al. (1993a, 1993b) considered 12 species under two subgenera (*Bunobus* and *Pseudacrobeles*). The subgenera were distinguished from each other based on the following characters: for *Pseudacrobeles*, triradiate lip region (very rarely with hexaradiate or bilateral symmetry) and labial probolae present (if absent then lips low and amalgamated); for *Bunobus*, lip region bilaterally symmetric, labial and cephalic probolae absent and subdorsal and subventral lips larger than lateral lips. Recently, Andrássy (2005) elevated the subgenus *Pseudacrobeles* (*Bunobus*) to genus level separate from the subgenus *Pseudacrobeles*. However, until further confirmation, we here treat each as a subgenus.

Pseudacrobeles species have been reported from every continent in the world except Antarctica. The distribution of *Bunobus* and *Pseudacrobeles* greatly overlap. Both are found in South America (Loof, 1964; Andrássy, 1968; Zell, 1987; De Ley et al., 1993a, 1993b), Europe (Zell, 1987; Hernández, 1990; Holovachov and De Ley, 2001; Abolafia et al., 2002; Abolafia and Peña-Santiago, 2005; Holovachov and Boström, 2006; Abolafia and Peña-Santiago, 2013), southern and western Asia (De Ley et al., 1993a, 1993b; Shokoohi and Abolafia, 2012), and Africa (Schuurmans-Stekhoven, 1951; Loof, 1964; De Ley et al., 1993a, 1993b). Thus far only *Pseudacrobeles* has been described from North America (De Ley et al., 1993a) and Oceania (Boström and Holovachov, 2011). Despite their nearly worldwide distribution, *Pseudacrobeles* is unknown from northeast Asia, possibly due to lack of survey effort. A new species, described herein as *Pseudacrobeles* (*Pseudacrobeles*) *curvatus* sp. n., was isolated from soil samples collected from potato fields in South Korea. A detailed morphological description based on morphometric analyses including observations from light microscopy (LM) and scanning electron microscopy (SEM) is provided. As molecular barcode references, we also provide sequence information for the D2-D3 regions of 28S rDNA and the ITS1-5.8S-ITS2 region of rDNA.

Material and Methods

Nematode isolation and culture: Soil samples were collected from potato farms in Changwon-si, Gyeongsangnam Province, South Korea. Nematodes were extracted by the sieving and Baermann funnel method (Baermann, 1917). An in vitro culture was established from a single female specimen and maintained at room temperature (18–20°C) on soil agar plates containing 25 mg/ml autoclaved soil, 5 μg/ml cholesterol, and 1% agar.

Morphological observations: Individual nematodes were placed in 2 ml water in a 15 ml tube and quickly fixed with the addition of 4 ml of 80°C TAF (2% triethanolamine and 7% formaldehyde), then processed to dehydrated glycerin following Seinhorst (1959) and mounted in pure glycerin on HS-slides (Shirayama et al., 1993). Morphological characters were observed under an optical microscope (Olympus BX-51) with differential interference contrast (DIC). Morphometrics were performed on images obtained with a CoolSnap Photometrics color CCD digital camera and QCapture Pro 5.

Scanning electron microscope: Prior to SEM imaging, nematodes were fixed in TAF and held for at least 24 hr at room temperature, then post-fixed in a solution

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of 4% aqueous osmium tetroxide at 4°C for 3 d. Fixed specimens were dehydrated using a series of 10%–100% absolute ethanol for 1 hr at each concentration level and then dried in a Hitachi HCP-2 critical point drier. Dried nematodes were mounted on copper/nickel tape attached to SEM stubs. The stubs with mounted nematodes were sputter-coated with gold/palladium using a sputter-coater (Eiko IB-3) and the morphology of nematodes was observed under high-vacuum conditions using a Zeiss Ultra Plus at 15 kV.

Molecular procedures and sequence analyses: Total genomic DNA from the new species was extracted using an Epicentre MasterPure DNA Purification Kit (Epicentre Co.) according to the manufacturer's protocol. The ITS1-5.8S-ITS2 region and the D2-D3 regions of 28S rDNA were PCR amplified using universal primer sets (TW81/AB28 [Joyce et al., 1994] for ITS1-5.8S-ITS2 region and D2A/D3B [De Ley et al., 1999] for the D2-D3 region in 28S). PCR reactions were performed in a total volume of 50 μl, containing 2 μl of template DNA, 10 pmol of each primer, 10× Ex Taq buffer, 0.2 mM dNTP mixture, and 1.25 U of *Taq* polymerase (TaKaRa Ex Taq). PCR amplification conditions included an initial denaturing step at 95°C for 1 min, 35 cycles with a denaturation at 95°C for 30 sec, annealing at 50°C for 30 sec, extension at 72°C for 1 min, and a final extension at 72°C for 10 min. PCR products were purified using the QIAquick Gel Extraction Kit (QIAGEN Co.) following the manufacturer's instructions. PCR-amplified fragments were then sequenced using Big Dye Terminator Cycle-Sequencing (Applied Biosystems).

Sequences of the ITS1-5.8S-ITS2 region and the D2-D3 regions of 28S rDNA were aligned to their homologous gene sequences of some other *Pseudacrobeles* species available from GenBank, in Clustal X with default options (Thompson et al., 1997). Both ends of the aligned datasets were trimmed before sequence analyses were performed using the Genious 6 program.

Results

Pseudacrobeles (*Pseudacrobeles*) *curvatus* sp. n. (Figs. 1,2; Table 1)

Description

Female: Body cylindrical, length 453.1 to 563.4 μm, usually ventrally curved after fixation but occasionally irregularly contorted. Cuticle annulated; annuli 1.6 to 2.3 μm wide and 0.7 to 1.3 μm at midbody level. Three lateral incisures, end posteriorly near phasmid opening level on the tail. Head region

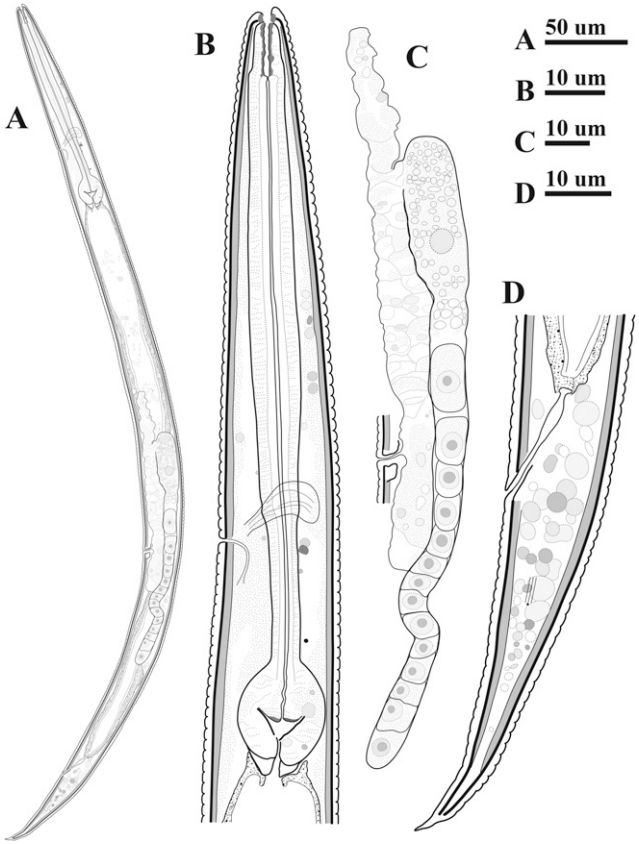

Fig. 1. *Pseudacrobeles* (*Pseudacrobeles*) *curvatus* sp. n. A. Whole female. B. Neck region. C. Female reproductive system. D: Female posterior region.

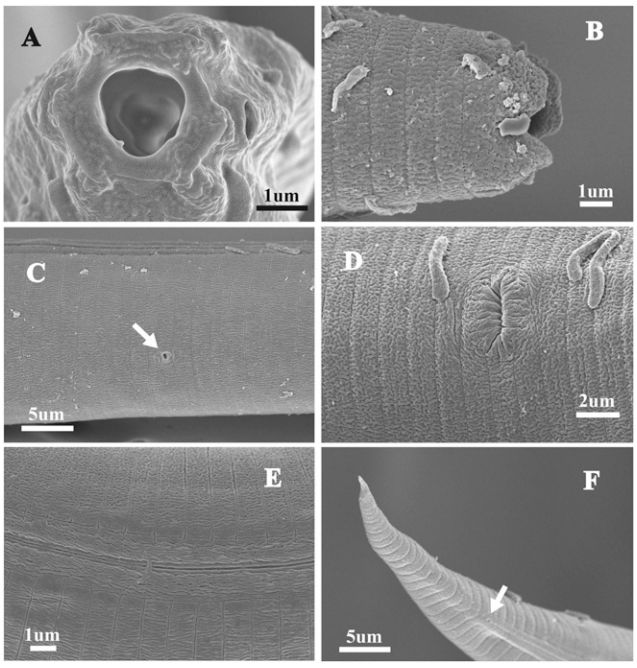

Fig. 2. *Pseudacrobeles* (*Pseudacrobeles*) *curvatus* sp. n. (scanning electron microscopy). A. Lip region. B. Anterior region. C. Excretory pore (arrow). D. Vulva. E. Lateral field. F. Female tail (arrow indicates phasmid opening near the end of the lateral lines).

TABLE 1. Morphometrics of *Pseudacrobeles* (*Pseudacrobeles*) *curvatus* sp. n.

Character	*P.* (*P.*) *curvatus* sp. n.	
	Holotype (n = 1)	Paratype (n = 8)
L	563.4	504.6 ± 38.4 (453.1–563.4)
Body width	26.3	24.3 ± 3.5 (20.6–31.3)
Pharynx length	134.3	136.0 ± 8.2 (125.8–154.6)
Tail length	54.7	50.3 ± 3.7 (43.9–54.7)
Anal region body width	15.5	12.7 ± 1.6 (11–15.5)
a	21.4	21.0 ± 2.0 (16.6–22.6)
b	4.2	3.7 ± 0.2 (3.5–4.2)
c	10.3	10.0 ± 0.3 (9.6–10.6)
c′	3.5	4.0 ± 0.3 (3.5–4.6)
Lip region width	6.9	6.4 ± 0.4 (5.4–6.9)
Stoma	12.3	11.5 ± 1.0 (10.4–13.4)
Corpus	75.8	77.5 ± 5.3 (70.1–89.2)
Isthmus	26.2	26.8 ± 2.1 (25.4–32.3)
Bulbus	20.0	19.1 ± 1.5 (16.6–21.6)
Stoma/lip region width	1.8	1.8 ± 0.2 (1.6–2.3)
Corpus/isthmus	2.9	2.9 ± 0.1 (2.8–3.1)
Nerve ring from anterior end	87.6	84.7 ± 3.3 (79–90.2)
Excretory pore from anterior end	93.5	87.7 ± 4.2 (82.1–93.5)
Deirid from anterior end	101.5	97.9 ± 3.5 (93.4–103.2)
Nerve ring (% pharynx)	65.2	62.3 ± 2.2 (58.3–65.2)
Excretory pore (% pharynx)	69.6	64.6 ± 3.2 (59.8–69.6)
Deirid (% pharynx)	75.6	72.1 ± 3.0 (65.7–75.6)
Rnr[a]	47	51.3 ± 3.5 (47–59)
Rep[b]	52	53.7 ± 3.6 (50–62)
Rdei[c]	56	59.1 ± 3.7 (55–67)
Vulva from anterior end	355.7	318.1 ± 24.5 (292.2–355.7)
V (%)	63.1	63.1 ± 1.5 (61.3–66.0)
Reproductive tract length	160.8	146.3 ± 14.6 (117–163.2)
G (%)	28.5	29.0 ± 1.6 (25.3–30.4)
Vagina	5.9	6.9 ± 0.5 (5.9–7.8)
Postvulval sac	24.1	25.1 ± 2.3 (22.1–28.9)
Uterus	50.6	48.4 ± 6.0 (39.3–59.0)
Uterus/body width	1.9	2.0 ± 0.2 (1.7–2.4)
Spermatheca	30.3	26.7 ± 4.3 (18.3–31.7)
Rectum	19.0	18.2 ± 1.3 (16.8–20.7)
Rectum/anal width	1.2	1.4 ± 0.2 (1.2–1.8)
Phasmid	20.0	18.5 ± 2.2 (15.2–21.1)
Phasmid (% tail)	36.6	36.7 ± 3.4 (30.2–41.5)
Tail annuli[d]	32	28.8 ± 2.3 (25–32)
Cuticle thickness	1.2	1.0 ± 0.2 (0.7–1.3)
Annuli width	2.2	2.0 ± 0.3 (1.6–2.3)

All measurements are in μm and in the form mean ± SD (range).
[a] Number of annules from the anterior end to the nerve ring.
[b] Number of annules from the anterior end to the excretory pore.
[c] Number of annules from the anterior end to the deirid.
[d] Number of annules from the anus to the tail end.

continuous with the neck. Lip region 5.4 to 6.9 μm wide, triradiate symmetry with 6+4 papillae. Cephalic probolae absent; three ridge-shaped, low rounded labial probolae present. Transverse, oval-shaped amphidial openings. Stoma cephaloboid, about 1.6 to 2.3 times length of the lip region diameter; bar, oval or comma-shaped cheilorhabdions. Cheilostom similar in width to the gymnostom and with a dorsal denticle on metastom. Pharyngeal corpus cylindrical, 2.8 to 3.1 times isthmus length. Isthmus narrower than corpus, and distinctly demarcated from corpus. Basal bulb oval-shaped with well-developed valve; cardia conoid, surrounded by intestinal tissue. Nerve ring at posterior end of corpus or boundary between corpus and isthmus, 47 to 59 annuli from the anterior end, at 58.3% to 65.2% of total neck length. Excretory pore at the level of posterior end of corpus to anterior end of isthmus, 50 to 62 annuli from anterior end, at 59.8% to 69.6% of total neck length. Deirid in the lateral field at level of isthmus, 55 to 67 annuli from anterior end, at 65.7% to 75.1% of pharynx length. Female reproductive system monodelpic-prodelpic. Vulva either protruding or not, postvulval sac 0.8 to 1.3 times the body width at the level of the vulva. Uterus tubular, 1.7 to 2.4 times the body diameter. Spermatheca 18.3 to 31.7 μm long. Oviduct very short. Ovary straight to posterior, sometimes with double flexure posterior to vulva, with a single row of oocytes. Rectum 1.2 to 1.8 times the anal body diameter. Tail ventrally curved and sigmoid elongated conoid, with 25 to 32 annules. Phasmid openings at 30% to 41.5% of tail length.

Male: Unknown.

Diagnosis and relationships: The new species *P. curvatus* sp. n. is characterized by having a small body (453.1–563.4 μm), three pairs of symmetrical lips, no cephalic probolae, pharyngeal corpus 2.8 to 3.1 times the isthmus length, nerve ring lying between corpus and isthmus, vulva position at 61.3% to 66.0% of total body length, uterus 39.3 to 59.0 μm long, spermatheca 18.3 to 31.7 μm long, postvulval uterine sac 0.8 to 1.3 times the body diameter in length, tail sigmoid elongated conoid (43.9–54.7 μm tail length, c = 9.6–10.6, c' = 3.5–4.6), and phasmid at 30.2% to 41.5% of tail length.

The new species shows morphological characters typical of the subgenus *Pseudacrobeles* by having radial symmetry of the lips and three lateral incisures that fade posteriorly near the level of phasmid openings. However, this new species is distinguished from other *Pseudacrobeles* species by morphological characters such as the absence of cephalic probolae, the similar widths of the cheilostome and gymnostom, and several measurements (Table 2). The new species resembles *P.* (*P.*) *variabilis* (Steiner, 1936) Steiner, 1938; however, it is distinguished by a shorter and wider tail (c = 9.6–10.6 *vs* 5.4–9.6 and c' = 3.5–4.6 *vs* 4.6–8.7) (De Ley et al., 1993a). Also, this new species differs from *P.* (*P.*) *baloghi* (Andrássy, 1968) De Ley, Siddiqi and Boström, 1993 by its shorter tail (c = 9.6–10.6 *vs* 7.1–9.2), the absence of cephalic probolae, and the relatively posterior position of the nerve ring (at the level of the corpus–isthmus junction *vs* three-fifths of the length of the corpus region) (De Ley et al., 1993a). Compared to *P.* (*P.*) *pauciannulatus* (Marinari-Palmisano, 1967) De Ley, Siddiqi and Boström, 1993, the new species is distinguished by 25 to 32 annuli on the tail (*vs* 20–24), the absence of cephalic probolae (*vs* present cephalic probolae), shorter cardia (2–4 *vs* 6.5 μm) and a gymnostom and cheilostome with similar widths (*vs* gymnostom narrower than cheilostome) (Rashid et al., 1984; Rashid et al., 1988). The new species resembles *P.* (*P.*)

TABLE 2. Morphometrics and diagnostic features of species of *Pseudacrobeles* (*Pseudacrobeles*) Steiner, 1938.

Species (female)	Body length (µm)	Pharynx	Tail	*a*	*b*	*c*	*c'*	Stoma	Corpus	Isthmus	Bulbus	Corpus/ isthmus	Nerve ring-anterior end	*V*	Phasmid	Phasmid (% tail)	Reference
P. curvatus sp. n.	453.1–563.5	125.8–154.6	43.9–54.7	16.6–22.6	3.5–4.2	9.6–10.6	3.5–4.6	10.4–13.4	70.1–89.2	25.4–32.3	16.6–21.6	2.8–3.1	79.0–90.2	61.3–66.0	15.2–21.1	30.2–41.5	This study
P. variabilis	440–795	120–185	55–100	17–30	3.4–4.3	5.4–9.6	4.6–8.7	9–16	74–122	20–31	14–24	2.7–4.5	78–112	57–64	8–24	12–32	De Ley et al. (1993a)
P. baloghi	400–620	123–162	49–79	16–24	3.0–3.7	7.1–9.2	3.5–6.4	11–14	63–105	21–30	15–21	2.9–4.2	72–99	59–64	10–21	18–27	De Ley et al. (1993a)
P. tabacum	535–690	121–148	60–75	21–29	4.2–5.0	7.6–10	4.1–4.8	10–13	80–98	16–19	15–16	4.5–5.1	90–107	58–64	16–27	27–36	De Ley et al. (1993a)
P. macrocystis	525–785	153–205	57–89	17–29	3.1–4.2	7.1–9.9	3.7–5.8	12–16	91–135	23–33	18–24	3.0–4.7	79–131	58–66	9–22	11–31	De Ley et al. (1993a)
	625–793	137–195	71–89	18.0–27.7	3.9–4.7	7.0–10.1	3.5–7.0	12–16	95–109	21–56	20–28	2.0–2.4	97–136	59–71	-	17–24	Shokoohi and Abolafia (2012)
P. laevis	595–705	159–182	46–57	24–29	3.4–3.9	11–14	3.1–3.8	12–15	97–122	29–33	18–21	3.3–4.2	96–111	61–66	12–20	34–42	De Ley et al. (1993a)
P. teres	740	-	-	25	4.1	11	4	-	-	-	-	4.5	-	62	-	27	De Ley et al. (1993a)
P. pauciannulatus	320–540	91–157	25–77	16.6–26.4	3.0–4.5	6.0–14.0	2.9–5.9	11.5–14.5	-	-	-	-	-	61–67	-	28–33	Rashid et al. (1984, 1988)
P. unguicolis	520–665	139–171	54–75	21.0–27.9	3.6–4.4	7.2–10.6	3.9–5.5	10–15	78–99	24–33	14–18	2.4–3.3	83–106	58–63	11–24	26–35	Abolafia et al. (2002)
P. eurystoma	524–644	135–172	37–48	25.8–31.5	3.3–4.3	12.8–16.3	2.7–4.1	11–15	85–99	25–44	15–18	2.3–3.1	92–103	62–67	13–18	38–40	Abolafia et al. (2002)
P. iranicus	479–560	97–123	64–75	21.9–25.2	4.4–5.0	7.3–8.0	5.0–5.6	12–13	62–72	18–33	16–18	2.2–3.4	73–86	57–61	-	36–38	Shokoohi and Abolafia (2012)
P. maungatautariensis	585–768	153–202	52–63	28.2–36.7	3.5–3.9	10.8–13.2	3.7–5.0	15–16	120–144	28–30	17–19	4.1–4.8	78–103	62–67	12.5–19.0	23–34	Boström and Holovachov (2011)
P. elongatus	603–780	161–198	42–54	24.0–29.0	3.3–4.5	11.3–15.8	2.8–4.2	13–18	97–119	24–35	15–20	3.4–4.6	84–127	59.7–63.5	13–18	35–47	Abolafia and Peña-Santiago (2005)

All morphometrics except *P. curvatus* sp. n. calculated from original descriptions.

TABLE 3. Uncorrected pairwise (*p*) distance among species of the genus *Pseudacrobeles* Steiner, 1938 for the D2-D3 regions of 28S rDNA.

		1	2	3	4	5	6	GenBank accession no.	Collection location
1	*Pseudacrobeles* (*Pseudacrobeles*) *curvatus* sp. n.							KX889090	South Korea
2	*Pseudacrobeles* (*Bunobus*) *bostromi*	16.2						HM439772	Ukraine
3	*Pseudacrobeles* sp. CR-2010	15.2	18					HM055403	United States
4	*Pseudacrobeles* sp. JB-56	15.7	12.1	16.7				DQ145653	United States
5	*Pseudacrobeles* sp. JB-85	14.1	11.8	16.7	9.8			DQ145654	Mexico
6	*Pseudacrobeles* sp. OH-2016[a]	14.1	11.8	16.7	9.8	0		KU180684	Mexico
7	*Pseudacrobeles* (*Pseudacrobeles*) *variabilis*[b]	15.7	12.1	16.7	0	9.8	9.8	AF143368	United States

[a] Same strain as JB-85.
[b] Same strain as JB-56.

unguicolis Abolafia, Liébanas and Peña-Santiago, 2002, but it is distinguished by a shorter tail (43.9–54.7 *vs* 54–75 μm), tail with 25 to 32 annuli (*vs* 38–41), uterus 1.7 to 2.4 times the body diameter (*vs* 3 times) and shorter vagina (0.2–0.3 *vs* 0.5 times the body width). A detailed comparison of the morphometrics and diagnostic features of *Pseudacrobeles* species is shown in Table 2.

Type habitat and locality: Specimens of *P.* (*P.*) *curvatus* sp. n. were collected from a small potato field in Bukmyeon, Uichang-gu, Changwon-si, Gyeongsangnam Province, South Korea (GPS coordinates: N 35° 22′ 22.5′′, E 128° 36′ 47.3′′). The soil was taken from the potato farm during a period of rest after the harvest.

Type material: Holotype (slide no. KOSPIV0000243269) and two paratypes (slide no. KOSPIV0000234931 and KOSPIV0000234932) are deposited in the National Institute of Biological Resources, Republic of Korea. Six paratypes (slide no. 01010401004-01010401009) are deposited in the Animal Phylogenomics Laboratory, Ewha Womans University, South Korea.

Etymology: The specific epithet refers to having a bent tail.

Molecular characterization: Sequences of the D2-D3 region of 28S and the ITS1-5.8S-ITS2 region of rDNA were obtained from *P.* (*P.*) *curvatus* sp. n. (GenBank accession no. KX889090 and KX889083, respectively). Comparison with some other *Pseudacrobeles* species shows that the D2-D3 region sequence differs by 14.1% (*Pseudacrobeles* sp. JB-85, DQ145654 and *Pseudacrobeles* sp. OH-2016) and 16.2% (*P.* (*b.*) *bostromi*, HM439772) (Table 3) and the ITS1-5S-ITS2 region shows a 25.3% difference from *Pseudacrobeles* sp. JB056 (DQ146429). However, molecular sequence information of D2-D3 and ITS1-5S-ITS2 for other species belonging to the genus *Pseudacrobeles* is very limited: only six D2-D3 of 28S rDNA sequences (four of which are from unidentified species) and one ITS1-5S-ITS2 region sequence from an unidentified species are available on GenBank. Further molecular analyses from diverse *Pseudacrobeles* species will help to better understand the sequence diversity and genetic divergence level among the species of this genus.

LITERATURE CITED

Abolafia, J., Liébanas, G., and Peña-Santiago, R. 2002. Nematodes of the order Rhabditida from Andalucía Oriental, Spain. The subgenus *Pseudacrobeles* Steiner, 1938, with description of a new species. Journal of Nematode Morphology and Systematics 4:137–154.

Abolafia, J., and Peña-Santiago, R. 2005. Nematodes of the order Rhabditida from Andalucía Oriental: *Pseudacrobeles elongatus* (de Man, 1880) comb. n. Nematology 7:917–926.

Abolafia, J., and Peña-Santiago, R. 2013. Iberian species of the subgenus *Bunobus* De Ley, Siddiqi and Boström, 1993 (Nematoda, Rhabditida, Cephalobidae), with description of *Pseudacrobeles (Bunobus) rotundilabiatus* sp. n. and comments on the subgenus. Zootaxa 3640:200–212.

Andrássy, I. 1968. Fauna paraguayensis. 2. Nematoden aus den Galeriewäldern des Acaray-Flusses. Opuscula Zoologica 8:167–315.

Andrássy, I. 2005. Free-living nematodes of Hungary (Nematoda errantia), vol. 1. Budapest, Hungary: Hungarian Natural History Museum.

Baermann, G. 1917. Eine einfache methode zur auffindung von ankylostomum (Nematoden) larven in erdproben. Geneeskunding Tijdschrift voor Nederlandsch-Indië 57:131–137.

Boström, S., and Holovachov, O. 2011. Two new species of *Eucephalobus* Steiner, 1936 and one new species of *Pseudacrobeles* Steiner, 1938 (Rhabditida: Cephalobidae) with mucronate tail. Journal of nematode morphology and systematics 14:91–104.

De Ley, P., Félix, M. A., Frisse, L. M., Nadler, S. A., Sternberg, P. W., and Thomas, W. K. 1999. Molecular and morphological characterisation of two reproductively isolated species with mirror-image anatomy (Nematoda: Cephalobidae). Nematology 1:591–612.

De Ley, P., Siddiqi, M. R., and Boström, S. 1993a. A revision of the genus *Pseudacrobeles* Steiner, 1938 (Nematoda: Cephalobidae). Part 1. subgenus *Pseudacrobeles* grad. n. Fundamental and Applied Nematology 16:219–238.

De Ley, P., Siddiqi, M. R., and Boström, S. 1993b. A revision of the genus *Pseudacrobeles* Steiner, 1938 (Nematoda: Cephalobidae). Part 2. Subgenus *Bunobus* subgen. n., problematical species, discussion and key. Fundamental and applied Nematology 16:289–308.

Hernández, M. A. 1990. *Heterocephalobus pseudolatus* n. sp. encontrada en Navarra, norte de España (Nematoda, Cephalobidae). Boletín de la Real Sociedad Española de Historia Natural. Biología 85:101–106.

Holovachov, O., and De Ley, P. 2001. Description of *Pseudacrobeles (Bunobus) bostromi* sp. n. (Rhabditida: Cephalobidae) from rotting wood in Roztochya, Ukraine. Journal of Nematode Morphology and Systematics 4:21–29.

Holovachov, O., and Boström, S. 2006. Description of *Pseudacrobeles (Bunobus) arboricola* sp. n. (Rhabditida: Cephalobidae) from rotting wood in Roztochya, Ukraine. Journal of Nematode Morphology and Systematics 9:49–54.

Joyce, S. A., Reid, A. P., Driver, F., and Curran, J. 1994. Application of polymerase chain reaction (PCR) methods to identification of entomopathogenic nematodes. Pp. 178–187 *in* A. M. Burnell, R. U. Ehlers, and J. P. Masson, eds. Biotechnology: Genetics of entomopathogenic nematodes-bacterium complexes. Luxembourg: European Commission, DG-XII.

Loof, P. A. A. 1964. Free-living and plant-parasitic nematodes from Venezuela. Nematologica 10:201–300.

Marinari-Palmisano, A. 1967. Contributo alla conoscenza di alcuni nematode dei generi *Rhabditoides, Eucephalobus, Heterocephalobus*. Redia 50:289–308.

Rashid, F., Geraert, E., and Sharma, R. D. 1984. Morphology, taxonomy and morphometry of some Cephalobidae (Nematoda: Rhabditida) from Brazil, with descriptions of two new genera and four new species. Nematologica 30:251–298.

Rashid, F., Geraert, E., Coomans, A., and Suatmadji, W. 1988. Cephalobidae from the Krakatau region (Nematoda: Rhabditida). Nematologica 34:125–143.

Schuurmans-Stekhoven, J. J. 1951. Nématodes saprozoaires et libres du Congo Belge. Mémoires de l'Institut Royal des Sciences Naturelles de Belgique 2:3–79.

Seinhorst, J. W. 1959. A rapid method for the transfer of nematodes from fixative to anhydrous glycerin. Nematologica 4:67–69.

Shirayama, Y., Kaku, T., and Higgins, R. P. 1993. Double-sided microscopic observation of meiofauna using an HS-slide. Benthos Research 44:41–44.

Shokoohi, E., and Abolafia, J. 2012. Nematodes of the order Rhabditida from Tehran province, Iran. The genus *Pseudacrobeles* Steiner, 1938. Annales Zoologici 62:331–340.

Steiner, G. 1936. Opuscula miscellanea nematologica. IV. Proceedings of the helminthological Society of Washington 2:74–80.

Steiner, G. 1938. Opuscula miscellanea nematologica. VII. Proceedings of the helminthological Society of Washington 5:35–40.

Thompson, J. D., Gibson, T. J., Plewniak, F., Jeanmougin, F., and Higgins, D. G. 1997. The CLUSTAL_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. Nucleic Acids Research 25:2876–4882.

Zell, H. 1987. Nematoden eines Buchenwaldbodens 9. Die Cephaloben (Nematoda, Rhabditida). Carolinea 45:121–134.