

# *Cervidellus sonorensis* n. sp. (Nematoda: Cephalobidae) from the desert of Anza-Borrego, CA, USA

Erik J. RAGSDALE<sup>1,2</sup>, Manuel MUNDO-OCAMPO<sup>1,3</sup>,  
Daniel J. BUMBARGER<sup>1,2</sup> and James G. BALDWIN<sup>1,\*</sup>

<sup>1</sup>Department of Nematology, University of California, Riverside, CA 92521, USA

<sup>2</sup>Department of Evolutionary Biology, Max Planck Institute for Developmental Biology, Spemannstraße 37,  
72076 Tübingen, Germany

<sup>3</sup>CIIDIR-IPN, Unidad Sinaloa, Sinaloa, Mexico

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**Summary** – *Cervidellus sonorensis* n. sp. is described from sand near the roots of creosote bush (*Larrea tridentata*) from Anza-Borrego Desert State Park, CA, USA. It is distinguished from others in the genus by the combination of a greatly distended, guitar-shaped corpus and the morphology of the lips and labial probolae. Distinctive features include a large pharyngeal metacarpus with a double swelling, a knob-like ledge at the base of each labial probola and lips with five filamentous tines, the most apical of which are those closest to the primary axil. One male individual was discovered in this parthenogenetic species. Characters used to diagnose *C. sonorensis* n. sp. are not definitive with respect to other genera and are probably plesiomorphic or convergent in light of DNA-based phylogenetic hypotheses. The position of *C. sonorensis* n. sp. and morphologically close congeners in relation to species of *Nothacrobeles* and *Paracrobeles* is discussed.

**Keywords** – Cephalobomorpha, description, morphology, morphometrics, new species, phylogeny, scanning electron microscopy, taxonomy.

Cephalobidae Filipjev, 1934 are bacterial-feeding, terrestrial nematodes that are worldwide in distribution. Habitats include dry, desert soils, such as in the southwestern North American deserts, from which many species have already been described or recorded. Taxonomy of the family is problematic (De Ley, 1997) and the most important diagnostic characters of genera and species, namely of the lip region, are highly plastic and convergent when mapped on DNA-based phylogeny (Nadler *et al.*, 2006; Smythe & Nadler, 2006; Holovachov *et al.*, 2009a, b, 2011; Boström *et al.*, 2010). In spite of taxonomic difficulties, including the probable paraphyly of several genera, a huge diversity of cephalob species remains to be documented and described. Cephalob taxonomy increasingly has made use of characters in addition to those understood from light microscopy (LM), such as those resolved at the level of scanning electron microscopy (SEM) and from DNA sequences, allowing better delineation of species. A broader sampling of morphological and molecular characters, as well as ad-

ditional species in comprehensive phylogenetic analyses will allow much-needed monographic revision. The genus *Cervidellus* Thorne, 1937, in particular, is central to the question of independent evolution of complex labial structures. Its species are found in many habitats but are especially well known from arid regions. Several species of *Cervidellus* have been recorded from the southwestern and southern parts of North America (Thorne, 1925, 1937; Holovachov *et al.*, 2007), including the Mojave and Sonoran deserts of southern California. Herein a new species from the region, *Cervidellus sonorensis* n. sp., is described.

## Materials and methods

Nematodes were extracted from soil samples using sieving and Baermann funnel extraction. Nematodes collected from the type locality in April 2003 were processed for permanent slide mounts and isolated into culture

\* Corresponding author, e-mail: james.baldwin@ucr.edu

*in vitro*. For slide mounts, nematodes were heat-killed at 65°C, fixed in 4% aqueous formalin and then dehydrated and transferred to glycerin (Seinhorst, 1959). Slide mounts were examined by LM for morphometrics and morphological description. Cultures of *C. sonorensis* n. sp. (strain code JB-138) were maintained on an unidentified bacterial strain (code 272 PEET) on 2% water agar. Nadler *et al.* (2006) used nematodes from these cultures to sequence a partial 28S rRNA gene and included the sequence (as taxon '*Cervidellus* sp. JB-138') in their phylogeny of Cephalobidae. Waceke *et al.* (2005) sequenced the ITS1, 5.8S and ITS2 of rRNA gene fragments from cultured nematodes and included the taxon (likewise as '*Cervidellus* sp. JB-138') in a phylogeny of several cephalob species. Nematodes in culture JB-138 were ultimately lost, but *C. sonorensis* n. sp. was re-collected from the type locality in June 2010. Re-isolated nematodes were processed for gene sequencing as well as for SEM.

## SEM

Nematode specimens were concentrated in a 12.5 ml volume of distilled water, transferred to a 25 ml glass vial, and then immediately fixed in 5.0% buffered formalin solution (pH 7.0) at 55–60°C for a minimum of 24 h. To minimise distortion and loss of rigidity of fragile structures such as probolae, nematodes were rinsed with several changes of 0.1 M phosphate buffer and post-fixed for 4 h in 2.0% osmium tetroxide solution. Post-fixed specimens were rinsed with several changes of cold 0.1 M phosphate buffer during a period of 15 min and dehydrated through a series of aqueous dilutions of ethanol from 20% through 100%. Dehydrated specimens were critical-point dried in a Tousimis® (Rockville, MD, USA) Autosamdri-810. Specimens were mounted in several positions on top of double-sticking copper tape attached to aluminium stubs, coated for 1–3 min with a 25 nm layer of gold palladium in a Cressington® (Watford, UK) 108 Auto sputter coater, and then observed with a Phillips® XL 30-FEG 35 scanning electron microscope operating at 10 kV.

## DNA EXTRACTION

To test the coherence of individuals isolated at different times as belonging to a single species, the D2-D3 domain (*ca* 780 base pairs) of the 28S rRNA gene was obtained from a nematode picked from a sample taken 7 years after the original material was collected. A single, live,

female nematode was prepared for sequencing by cutting, lysing, and amplification of DNA by the polymerase chain reaction (PCR) using the primers D2Ab (forward) and D3B (reverse) by the methods described in De Ley *et al.* (2005). Examination of the sequence in Clustal X (Jeanmougin *et al.*, 1998) showed an exact match with the homologous sequence published for *Cervidellus* sp. JB-138 (Nadler *et al.*, 2006). Because a more complete sequence of the 28S gene of this species has already been published, the sequence obtained in the present study is not shown herein but has been deposited in the GenBank database under accession number HM776994.

Morphometric characters presented are as defined for Cephalobidae by Tandingan De Ley *et al.* (1999). Terminology of cephalic structures is congruent with Rashid *et al.* (1989). Stoma terminology is based on De Ley *et al.* (1995). For morphological comparison, type material for *Cervidellus capraeolus* (De Ley, Geraert & Coomans, 1990) Boström & De Ley, 1996 was obtained courtesy of Paul De Ley.

### *Cervidellus sonorensis*\* n. sp.

= *Cervidellus* sp. JB-138 *apud* Waceke *et al.*  
(2005) and Nadler *et al.* (2006)  
(Figs 1–3)

## MEASUREMENTS

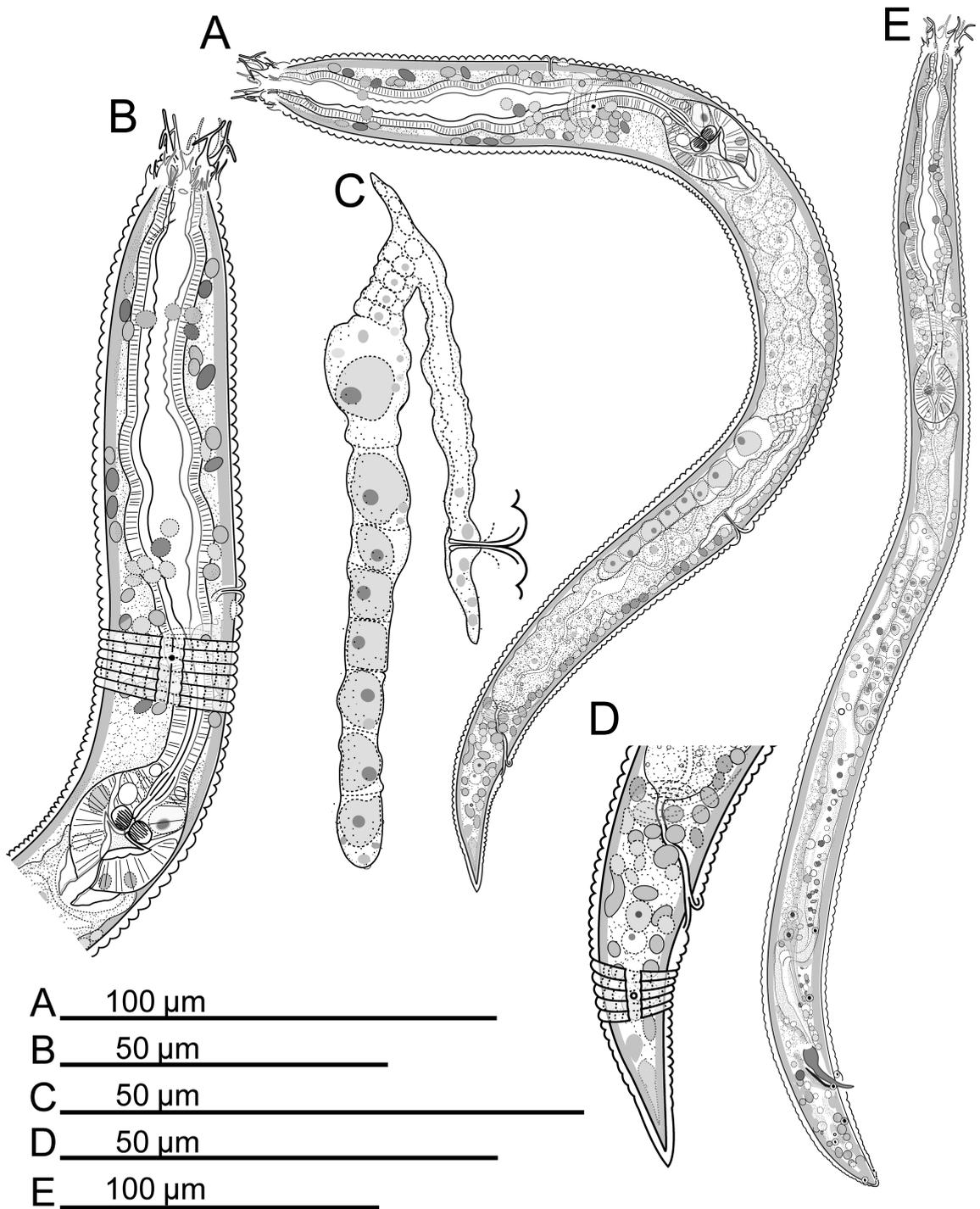
See Table 1.

## DESCRIPTION

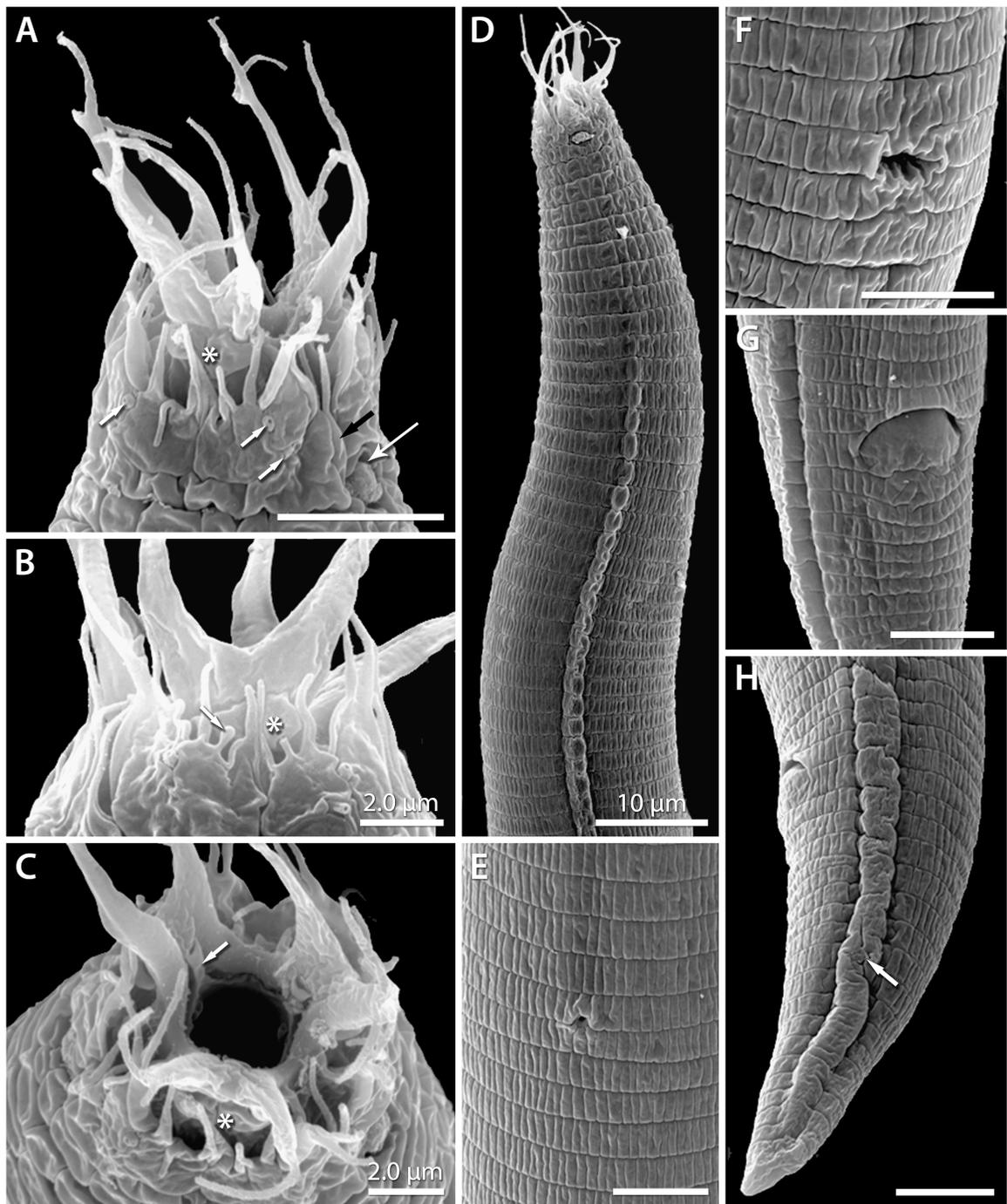
### *Female*

Body straight to arcuate dorsally or ventrally after heat relaxation. Body diam. greatest at neck, specifically in region of pharyngeal metacarpus and postcorpus. Cuticle thickness averaging 1.6 µm at region of max. body diam. Cuticle annulated throughout body except at tail tip and tessellated into longitudinal blocks with striae discontinuous across annulations. Lateral field with three incisures (two wings or ridges), extending from middle of pharynx to anus, anteriorly, field narrowing briefly to two incisures before also disappearing at level of middle pharynx, and, posteriorly, to two incisures extending to tail tip. Lip region slightly offset. Six lips each with five tines. Lips asymmetrical, such that pairs of adjacent lips are mirrored in symmetry. Labial tines generally filamentous, although

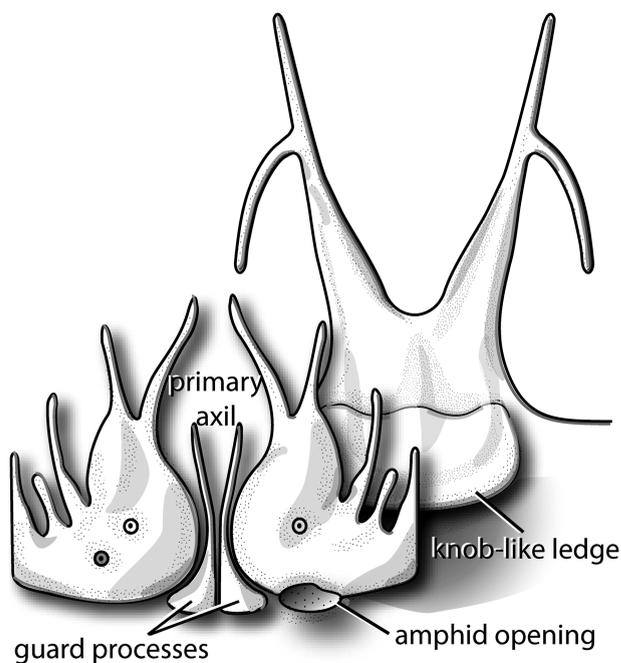
\* The species epithet denotes the type locality, which is in the northern part of the Sonoran desert.



**Fig. 1.** *Cervidellus sonorensis* n. sp., female (A-D) and male (E). All are right lateral views. A: Entire specimen; B: Anterior end; C: Reproductive tract; D: Tail region; E: Entire specimen (tail papillae represented by ringed black dots).



**Fig. 2.** Scanning electron micrographs of the female of *Cervidellus sonorensis* n. sp. Asterisks indicate abaxial basal ledge of probola. A: Oblique dorsal view of lip region (short white arrows indicate labial and cephalic papillae; long white arrow indicates amphidial aperture; black arrow indicates guard process); B: Right subventral view of lip details, including a shorter length variant of fourth labial tine (arrow) from primary axil; C: Dorsal, en face view of lip region (arrow indicates thin, adaxial ridge of probola); D: Lateral view of neck, showing tapering of lateral field; E: Ventral view of excretory pore; F: Oblique ventral view of vulva; G: Oblique ventral view of anus; H: Lateral view of tail (arrow indicates phasmid). (Scale bars = 5  $\mu\text{m}$  unless otherwise labelled.)



**Fig. 3.** Diagrammatic representation of lip region of *Cervidellus sonorensis* n. sp. From left to right: right subdorsal lip, guard processes, right lateral lip and right subventral probola.

fourth tines (counting from the primary axil) sometimes shorter and rounded (clavate). First and second tines longest, most prominent, extending from apex of lip. At primary axils, lips interrupted by arcuate guard processes that are triangular at their bases. Labial papillae at base of apex of each lip, cephalic papillae located where submedial (quadrant) lips meet main body cuticle, elliptical amphidial apertures on lateral sectors. Labial and cephalic papillae each punctuated by a pore. Probolae doubly forked. Primary forks at one-third of length of probolae, secondary forks at two-thirds of length of probola (half length of each primary branch of probola), giving impression of a harpoon shape, although terminal branches apparently flimsy as found in all SEM preparations. Posterior to primary fork of each probola is a basal, abaxial, knob-like ledge, appearing cup-shaped under LM. Basal ledge as wide as base of probola, edges of ledge discontinuous with primary branches of probola. On main stem of each probola is a thin, offset, adaxial ridge forking anteriorly and widening posteriorly, except for discontinuities between the three probolae, posterior spreading of these ridges giving impression of a thin collar within stoma. Rhabdion of cheilostom, at base of each probola stem, clearly visible under LM. Gymnostom short, less than one-fourth of stegostom. Four prominent, continuous

rhabdions in stegostom. Dorsal part of telostegostom with thin, anteriorly directed projection. Corpus 2.5-5.5 times longer than isthmus. Corpus guitar-shaped, namely with two serial swellings in metacarpus. Pharyngeal corpus tending to be greatly distended in dying or dead nematodes and upon fixation. Dilatation of procorpus widest anteriorly, namely at transition to stegostom, cuticle of posterior part of procorpus wavy or 'ribbed'. Metacarpus forming most distended part of pharynx with widest points usually extending to body wall. Double swelling in metacarpus exaggerated when pharynx is distended but also present in live nematodes when pharyngeal lumen is closed. Basal bulb pyriform with transverse, triradiate grinder. Plates of pharyngo-intestinal junction arc-shaped. Excretory pore usually located at level of nerve ring but variable. Excretory gland cell often visible by LM. Hemizonid immediately posterior to excretory pore. Deirid visible by LM and generally four or five annules posterior to excretory pore. Gonad monodelphic, prodelphic, reflexed, compact, on right side of intestine. Generally seven to nine but occasionally up to 11 oocytes visible in ovary. Oviduct with four layers of cells. Spermatheca offset, short, thin, obscure. Vagina straight, occupying *ca* one-third of body diam. Uterus generally narrow, with thin epithelium. Vulva narrow, elliptical. Post-vulval sac thin, short, little more than 0.5 vulval body diam. long. Anus arcuate. Phasmid opening a transverse slit in lateral field. Tail narrowly conoid, tip subacute.

#### Male

Only a single male found in all samples examined. Limited LM resolution of this one specimen is acknowledged and detail was only resolvable to level shown in Figure 1E. Males not observed in live culture. Male generally similar in morphology to female, although body diam. more uniform in male. Body size similar to that of female. Gonad monorchic, testis long, reflexed ventrally. Only early spermatocytes, occurring in testis flexure, entirely visible in specimen. Spicules slightly arcuate, gubernaculum slender. Two unicellular 'ejaculatory glands' with dorsal cell bodies tapering and extending to cloacal aperture. Eight pairs of genital papillae present plus single, faintly resolved, precloacal papilla (should be confirmed with additional specimens and SEM). Tail wider than in female, conical tail tip broader, more abruptly tapering.

#### TYPE LOCALITY AND HABITAT

*Cervidellus sonorensis* n. sp. was isolated from sand near the roots of creosote bush (*Larrea tridentata*) on

**Table 1.** Morphometrics of *Cervidellus sonorensis* n. sp. All measurements are in  $\mu\text{m}$  and in the form: mean  $\pm$  s.d. (range).

Character	Female		Male
	Holotype	Paratypes	Paratype
n	–	18	1
L	375	381 $\pm$ 27 (324-434)	379
a	16	15 $\pm$ 1.5 (12-18)	17
b	2.9	2.9 $\pm$ 0.2 (2.6-3.2)	3.0
c	13	11 $\pm$ 0.8 (9.0-13)	10
c'	2.2	2.5 $\pm$ 0.3 (2.0-3.0)	2.3
V	67	65 $\pm$ 1.4 (63-68)	–
Reproductive tract length (RTL)	78	80 $\pm$ 14 (62-108)	187
Dist. from ant. end to:			
vulva	251	249 $\pm$ 21 (205-291)	–
anus/cloaca	337	341 $\pm$ 27 (289-397)	342
nerve ring	88	91 $\pm$ 8.0 (73-103)	95
excretory pore	90	93 $\pm$ 12 (68-113)	95
deirid	98	100 $\pm$ 11 (77-121)	106
Labial probolae length (LPL)	13	13 $\pm$ 1.4 (11-16)	13
Lip region diam. (LRD)	15	14 $\pm$ 0.9 (13-16)	15
Stoma length (StL)	10	9 $\pm$ 0.8 (7-10)	9
Neck length (NL)	130	131 $\pm$ 7.4 (110-143)	128
Pharynx length	104	107 $\pm$ 6.7 (91-120)	114
Corpus length (CL)	67	66 $\pm$ 4.0 (59-75)	68
Isthmus length (IL)	14	19 $\pm$ 3.9 (11-27)	24
Basal bulb length	23	22 $\pm$ 2.4 (19-28)	22
Nerve ring/neck (%)	68	69 $\pm$ 5.3 (60-80)	74
Excretory pore/neck (%)	69	71 $\pm$ 7.7 (61-85)	74
Deirid/neck (%)	75	77 $\pm$ 8.2 (65-94)	83

**Table 1.** (Continued.)

Character	Female		Male
	Holotype	Paratypes	Paratype
Max. body diam. (MBD)	24	25 $\pm$ 1.7 (22-30)	22
Annule width at MBD	2.5	2.3 $\pm$ 0.6 (1-3)	2.4
Lateral field width at MBD	3.9	4.0 $\pm$ 0.3 (3.2-4.4)	3.9
Body diam. at:			
vulva (VBD)	20	22 $\pm$ 1.7 (19-26)	–
spermatheca (SPD)	21	22 $\pm$ 1.6 (19-25)	–
anus/cloaca (ABD)	13	15 $\pm$ 1.2 (13-18)	18
Spermatheca length (SpL)	12	16 $\pm$ 2.7 (10-20)	–
Vagina length	7	8 $\pm$ 1.1 (6-9)	–
Post-vulval sac length (PVS)	10	13 $\pm$ 1.8 (10-15)	–
Rectum length (RL)	12	14 $\pm$ 2.1 (10-18)	–
Vulva to anus dist. (VAD)	86	92 $\pm$ 7.5 (81-106)	–
Number of oocytes	7	8 $\pm$ 1 (7-11)	–
Spicule length (SpL)	–	–	20
Gubernaculum length (GL)	–	–	12
Testis flexure length	–	–	70
Tail length (TL)	29	36 $\pm$ 2.1 (30-39)	37
Phasmid to anus/cloaca dist.	13	16 $\pm$ 2.2 (13-22)	16
Annules from ant. end to:			
nerve ring	29	30 $\pm$ 3 (24-37)	35
excretory pore	29	31 $\pm$ 3 (26-35)	35
deirid	33	34 $\pm$ 3 (28-41)	39
Number of tail annules	18	18 $\pm$ 1 (15-20)	16
LPL/LRD	0.9	1.0 $\pm$ 0.1 (0.8-1.2)	0.9
StL/LRW	0.7	0.6 $\pm$ 0.1 (0.5-0.7)	0.6
CL/IL	4.8	3.5 $\pm$ 0.8 (2.5-5.5)	2.8
SpL/SBD	0.6	0.7 $\pm$ 0.1 (0.5-1.0)	–

**Table 1.** (Continued.)

Character	Female		Male
	Holotype	Paratypes	Paratype
VL/VBD	0.4	0.3 ± 0.1 (0.3-0.4)	–
PVS/VBD	0.5	0.6 ± 0.1 (0.4-0.8)	–
RL/ABD	0.9	1.0 ± 0.2 (0.7-1.2)	–
G (RTL/L (%))	21	21 ± 3.0 (17-27)	49
Phasmid/TL (%)	45	45 ± 6.7 (36-57)	43

a small sand dune at Hawks Canyon in Anza-Borrego Desert State Park, San Diego County, CA, USA, at 33°10'49"N, 116°12'20"W by D. Bumbarger on 4 April 2003 and by E. Ragsdale on 15 June 2010.

#### TYPE MATERIAL

Holotype female (slide accession number 30608), ten paratype females (30609-30618), one paratype male (30619), and several juvenile specimens deposited in the University of California Riverside Nematode Collection, Riverside, CA, USA. Four paratype females deposited at the University of California Davis Nematode Collection, Davis, CA, USA. Four paratype females deposited at the Swedish Museum of Natural History, Stockholm, Sweden.

#### DIAGNOSIS AND RELATIONSHIPS

*Cervidellus sonorensis* n. sp. is small (325-435 µm long) with the body broadest at the neck. The probolae are doubly forked and harpoon-shaped. The base of each probola has a knob-like ledge. The lips are asymmetrical with five tines each, the most apical (anterior) tines being closest to the primary axil. The pharyngeal metacarpus has a double swelling and the entire corpus is usually distended in dying and fixed nematodes. The corpus is at least 2.5 times longer than the isthmus. Males present but rare. The combination of characters given for the probolae, lips, and pharynx, is unique for the species.

Comparatively limited observations of *C. rarus* (Nesterov, 1969) Boström & De Ley, 1996 indicate that it is close to *C. sonorensis* n. sp. Distinguishing *C. rarus* is a fusiform vs doubly swollen metacarpus and, if adequately interpreted from LM in the original description, a basal

ledge wider than the base of the probolae. Although the position of the vulva in *C. rarus* is given as ca 66% of the body length, the drawing shows a more posterior vulva (ca 70%). Such a posterior vulva position would also set the species apart from *C. sonorensis* n. sp., although the drawing is likely to be a misrepresentation. The lack of SEM of *C. rarus* prohibits comparison of labial morphology, which would very likely yield the characters necessary to separate it from or synonymise it with another species of *Cervidellus*, including *C. sonorensis* n. sp. Because closer examination of labial and probolae characters could just as easily reject as support synonymisation of *C. rarus* with *C. sonorensis* n. sp., we contend that presently they are best considered as two distinct species.

*Cervidellus sonorensis* n. sp. is morphologically close to *C. capraeolus*. The two species are unique in having a guitar-shaped pharynx, although this character is more pronounced in *C. sonorensis* n. sp. and is not always present in *C. capraeolus* (Boström, 2000; Holovachov *et al.*, 2007). *Cervidellus sonorensis* n. sp. differs from the latter by the following characters: probolae with two vs three forks, lips with five vs eight labial tines, lips with no tines vs one tine between apex of lip and primary axils, labial tines that are filamentous vs blunt and rounded, pharyngeal corpus at least 2.5 times (usually three to four times) vs 1.5-2.5 times longer than isthmus, generally greater body length, at 325-435 vs 240-330 µm and ovary always straight vs usually with two (but sometimes no) flexures posterior to the vulva.

*Cervidellus cancellatus* (Thorne, 1925) Boström & De Ley, 1996 and *C. sonorensis* n. sp. are similar in the structure of their probolae, although the former species differs by having lips with only three vs five tines, absence vs presence of a ledge or protuberance at the base of the probolae and a weakly expanded vs robust metacarpus. *Cervidellus doorselaeri* (De Clerk & De Ley, 1990) Boström & De Ley, 1996 and *C. sonorensis* n. sp. are putatively disparate species with respect to phylogeny (Nadler *et al.*, 2006) but share some morphological features. Characters distinguishing *C. doorselaeri* from *C. sonorensis* n. sp. include rounded vs filamentous tines on the probolae, probolae without vs with a basal ledge, three vs two levels of probola forking, probolae with primary forks at two-thirds vs one-third of length of probolae, secondary forks shallow vs at two-thirds of length of probolae, secondary axils separated by small and lobate vs filamentous tines and stoma weakly vs strongly sclerotised. Other *Cervidellus* species clearly differ from *C. sonorensis* n. sp. by the numbers and positions of forks

in the probolae or by having symmetrical and often lobate lips.

## Discussion

*Cervidellus* is poorly defined as an increasing number of species descriptions have caused its boundaries to overlap with those of other cephalob genera, particularly *Acrobeles* von Linstow, 1877 and *Nothacrobeles* Allen & Noffsinger, 1971. For example, frequently cited characters circumscribing the genus are probolae with no, or up to three, forks, primary axils with or without guard processes and symmetrical or asymmetrical lips. All of these characters also apply to some species of *Nothacrobeles* and *Paracrobeles* Heyns, 1968. The lack of a basal ridge, namely an abaxial ridge continuous with the primary branches of each probola, seems to distinguish *Cervidellus* from *Nothacrobeles*, although homology of the various basal protuberances found in species of both genera is certainly not clear cut. *Cervidellus* was split into two morphological groups (*vexilliger* and *hamatus* groups) based on: *i*) symmetry vs asymmetry of the lips, respectively; *ii*) three or four vs four to nine labial tines, respectively; and *iii*) the presence vs absence, respectively, of guard processes in the secondary axils (Abolafia *et al.*, 2003). In this division, *C. sonorensis* n. sp. clearly falls into the *hamatus* group. However, these groups are incongruent with phylogeny based on at least a large segment (*ca* 77%) of the partial 28S rRNA gene, which rejects monophyly the genus (Nadler *et al.*, 2006; Smythe & Nadler, 2006; Holovachov *et al.*, 2009a, b, 2011; Boström *et al.*, 2010). In all of these phylogenetic analyses, two of which include species morphologically close to *C. sonorensis* n. sp., *i.e.*, *C. capraeolus* and *C. cancellatus* (Boström *et al.*, 2010; Holovachov *et al.*, 2011), *Nothacrobeles borregi* Poiras, Baldwin, Mundo-Ocampo & Bumbarger, 2002 is the sister taxon of *C. sonorensis* n. sp. A key character that may unite *C. sonorensis* n. sp. and *N. borregi* is the presence of an abaxial ledge or swelling at the base of the probolae. This basal ‘ledge’ is a protuberance proximal to the primary fork of the probola and, unlike a basal ‘ridge’, is not continuous with the branches of the probola. A basal (‘knob-like abaxial’) ledge is prominent in *N. borregi* (Poiras *et al.*, 2002) and is present, but not as prominent, in *Nothacrobeles laticollaris* De Ley & Vandebroek, 1992. A similar swelling in the same position is found in *C. capraeolus* (Boström, 2000). The possible, albeit dubious, homology of this structure in *N. borregi* was mentioned specifically with reference to the basal

swelling found in *C. capraeolus* (Poiras *et al.*, 2002). A ‘cup-shaped process’ is described for the probola of *C. rarus* (Nesterov, 1969) and appears to be similar to the basal ledge of *C. sonorensis* n. sp., which resembles an elliptical ring when observed by LM. The description of *C. rarus* shows the process to be wider than the bases of the probola, in contrast to the narrower process of *C. sonorensis* n. sp. This ledge is instead reminiscent of the large ‘knob-like abaxial ledge’ described for *N. borregi*, although comparison is limited by the lack of SEM in *C. rarus*. Homology of a basal ridge, typically a diagnostic character of the genus *Nothacrobeles*, is uncertain with respect to similar ridges in other genera although *Cervidellus* spp. generally lack such a structure. The presence of both a basal ridge and an offset and more posterior ledge in *N. borregi* and *N. laticollaris* indicates that the ridge and ledge represent at least two distinct homologues when considering the possible convergence of a ridge, which is found in species of both *Nothacrobeles* and *Paracrobeles*. The possibility of intermediates between these two confounds hypotheses of homology. For example, a basal ridge that dips proximally but is still continuous with the probola branches is found in *Nothacrobeles capricornis* (Holovachov, Boström, Susulovsky & Nevo, 2001) Abolafia & Peña-Santiago, 2003 and was described as a character intermediate between *Cervidellus* and *Nothacrobeles* (Holovachov *et al.*, 2001). It is this basal ridge that for the most part led to the transfer of this species from the former to the latter genus (Abolafia & Peña-Santiago, 2003). Because *C. sonorensis* n. sp. lacks a basal ridge, and because it has elevated lips and elongate, rod-like probolae typical of *Cervidellus* spp., the species is assigned to its present genus.

The probolae of *C. rarus* and *C. sonorensis* n. sp. are similar, in both cases being doubly forked and harpoon-shaped. The probolae of *C. rarus* as drawn are more robust than those of *C. sonorensis* n. sp., an interpretation that is supported by the depression drawn between the bases of each tine posterior to their forking. The lips of *C. sonorensis* n. sp. are acute and elevated, consistent with other *Cervidellus* species that are otherwise morphologically close, but in contrast to species of *Nothacrobeles* as diagnosed (Abolafia *et al.*, 2003). The number and details of the tines are unknown in *C. rarus*, in which the lips have been described simply as “six wedges” (Nesterov, 1969). Because of the increasing importance of SEM-informed lip characters in the diagnosis of cephalob species, the phylogenetic position or even positive identification of *C. rarus* may never be realised. Re-examination of type

material for *C. rarus* is considered impossible as slides have probably been discarded since the original description (L. Poiras, Academy of Sciences of Moldova, pers. commun.). Nevertheless, to allow for the possibility that a similar nematode can be again isolated from the type locality (Chişinău, Moldova) and positively identified, it is retained as a valid species that may later be more clearly described.

Long, thin labial tines are characteristic of both *C. sonorensis* n. sp. and *Paracrobeles* spp., in contrast to the rounded tines characteristic of most other species of *Cervidellus*. In *C. sonorensis* n. sp., several species of *Nothacrobeles* (Allen & Noffsinger, 1971; Tandingan De Ley *et al.*, 1999; Holovachov *et al.*, 2001; Poiras *et al.*, 2002; Abolafia & Peña-Santiago, 2003), and *Paracrobeles mojavicus* Taylor, Baldwin & Mundo-Ocampo, 2004, the most apical labial tines are closest to the primary axils. In contrast, other *Cervidellus* species with asymmetrical lips, such as *C. capraeolus* and *C. doorsselaeri*, have one or more tines that separate the apices of the lips and the guard processes (De Clerck & De Ley, 1990; De Ley *et al.*, 1990). Although homologies between some individual tines are doubtful, the number of labial tines seems to be highly variable among species and will probably still continue to be useful in diagnostics. However, the inflated number of tine numbers in *Acrobeles* spp. (Thorne, 1925) could confound hypotheses of their homology and their use in phylogenetics.

In addition to its affinity to species of *Cervidellus* and *Nothacrobeles*, *C. sonorensis* n. sp. shows some similarities with *Paracrobeles* spp. One similarity is the tendency for the metacarpus to be greatly distended, especially upon fixation. In *Paracrobeles* spp. the procorpus is apparently closed, whereas in *C. sonorensis* n. sp. the entire corpus is distended. An open procorpus can occur in other species of *Cervidellus*, including *C. capraeolus* (De Ley *et al.*, 1990) and *C. baujardi* Mounport, Boström & Villenave, 2002, and in some species of *Acrobeles* (Rashid *et al.*, 1990; Shahina & De Ley, 1997), raising doubts of the homology of this feature as distinct from artefact. In spite of the wider presence of a distended pharyngeal corpus in other species, the metacarpus of *C. sonorensis* n. sp. is most similar to that of *C. capraeolus* in being guitar-shaped, a character which is distinct in the latter when the corpus is distended. Similar between *C. sonorensis* n. sp. and *P. mojavicus* is the presence of unicellular 'ejaculatory glands' associated with the cloaca in the male, although we do not rule out the possibility of this feature being present but not described in other close species as

glands associated with the *vas deferens* have also been found in *Acrobeles crossotus* Steiner, 1929 and *A. complexus* Thorne, 1925 (Steiner, 1929; Thomas, 1965). In addition to morphological features, preliminary 28S D2-D3 evidence supports *P. mojavicus* as part of a clade including *N. borregi* and *C. sonorensis* n. sp., likewise to the exclusion of other species of *Cervidellus* and *Nothacrobeles* (Pereira *et al.*, unpubl.).

In contrast to its morphologically close congeners *C. capraeolus*, *C. rarus*, and *C. cancellatus*, as well as to *N. borregi*, *C. sonorensis* n. sp. has at least one known male individual. Interestingly, the species is at least facultatively parthenogenetic as strains of only females were successfully maintained in culture. It is unknown whether males function in reproduction of the species or are simply vestigial. A better knowledge of the phylogeny of the group could test the possibility that the limited presence of males is a transitional state in the evolution of parthenogenesis in Cephalobidae.

In spite of limited resolution offered by analyses of one gene, molecular phylogenetic hypotheses predict that the genera *Cervidellus*, *Nothacrobeles* and *Acrobeles* are paraphyletic and so the elaborate probolae characteristic of all of these genera probably comprises several cases of convergence (Nadler *et al.*, 2006). We emphasise that a combination of molecular and broad scale morphological analysis is essential for revising generic characters and reassigning species to genera. Consideration of molecular characters may require synonymy of some or all of these genera with *Acrobeles*. However, because establishment of adequately tested homologies is still preliminary, and because these genera are still undersampled in phylogeny, *C. sonorensis* n. sp. has been presented in accordance with traditional diagnostic characters and is thus ascribed to the genus *Cervidellus*.

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