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Four new species of Iponematinae (Drilonematidae, Drilonematoidea, Cephalobomorpha) parasitic in earthworms: description and molecular affiliations

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Summary – Three new species of *Filiponema*, *Filiponema* sp., and a new species of *Iponema* are described from earthworms. *Filiponema champa* sp. n., from the coelomic cavity of *Pheretima* sp. from Vietnam, is closest to *F. cylindropharyngatum* and *F. javanicum* but differs in having a wider body, longer spicules and more conspicuous excretory pore. *Filiponema yaoense* sp. n., from the coelomic cavity of *Pheretima* sp. from Vietnam, is differentiated from all valid species of *Filiponema* by its long, conspicuous excretory duct and a nerve ring situated more closely to the anterior extremity. It can be differentiated from *F. champa* sp. n. by a slimmer body, the more anterior position of amphids, circular *vs* transversely elliptical amphids and caudal organs, the wider tail tip, a shorter mail tail, longer eggs, smaller, straighter, spicules and a smaller gubernaculum. *Filiponema suifunense* sp. n., from the coelomic cavity of *Drawida ghilarovi* from Russia, is distinguished from all known species of the genus due to the presence of narrow, projecting lateral fields. *Filiponema* sp., from the coelomic cavity of *Lumbricus friendi* from France, is described but not named due to lack of material. *Iponema visayanum* sp. n., from the coelomic cavity of Megascolecidae gen. sp. from the Philippines, can be distinguished from *I. tonkinense* by its longer spicules and pharynx and its larger amphids situated further from the anterior extremity than in *I. tonkinense*. D2-D3 LSU sequences were obtained for *I. visayanum* sp. n. and *F. suifunense* sp. n. and compared with homologous sequences of other Drilonematoidea nematodes.

Keywords – Annelida, *Drawida ghilarovi*, *Filiponema*, *Filiponema champa* sp. n., *Filiponema suifunense* sp. n., *Filiponema yaoense* sp. n., France, *Iponema*, *Iponema visayanum* sp. n., *Lumbricus friendi*, Megascolecidae gen. sp., morphology, morphometrics, new species, *Pheretima*, Philippines, phylogeny, Russia, SEM, taxonomy, Vietnam.

In this paper we present taxonomic descriptions for several new nematode species belonging to the genera *Filiponema* Timm & Maggenti, 1966 and *Iponema* Timm & Maggenti, 1966 of the subfamily Iponematinae (Drilonematidae, Drilonematoidea, Cephalobomorpha), and also provide molecular characterisation for the three species.

Iponematinae Spiridonov & Ivanova, 2005 is a group of nematodes parasitising a coelomic cavity of earthworms. All members of the subfamily are characterised in having large amphidial apertures. After the revision of Iponematinae (Ivanova & Neuhaus, 2011), the subfamily accommodates five genera, *i.e.*, *Iponema* (type genus), *Filiponema*, *Tonoscolecinema* Timm, 1967, *Globocephalonema* Anand & Rao, 1986 and *Plutellonema* Timm & Maggenti, 1966.

The distribution of Iponematinae is based mainly on the earthworms from the vast subfamily Megascolecoidea, although a host from Lumbricidae and another from Glossoscolecidae have also been recorded (both serving hosts for the members of *Filiponema*). *Filiponema* is the only taxon of Drilonematoidea recorded in lumbricid hosts apart from *Dicelis* Dujardin, 1845 (Dicelinae, Drilonematidae). Although, as yet, only a single species of *Filiponema* has been described from a lumbricid, a greater diversity can be expected. Several species of drilonematid nematodes featuring large amphids have been found in lumbricids but the descriptions are rather inadequate by current standards (Kakulia & Kvavadze, 1974; Kvavadze & Eliava, 1975; Kvavadze *et al.*, 1985, 1986) and were later considered as invalid (Ivanova, 1994). Occasional findings of similar forms have been recorded by authors from lumbricids in Scotland, North Caucasus, and the Russian Far East (unpubl.).

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Three new species of *Filiponema* and a new species of *Iponema* described herein were recovered from earthworms collected in Eastern Asia. Yet another species of *Filiponema* parasitising a lumbricid in France is presented as *Filiponema* sp. and not formally named as the description is based on only one specimen. The new 'Asian' species of *Filiponema* were found in pheretimoid earthworms in Vietnam (two species) and in a moniligastrid host in the Russian Far East (one species; the first record). The new species of *Iponema* was recovered from a pheretimoid host in the Philippines. The molecular data were obtained for a species of *Filiponema* and a species of *Iponema*, are the first for the subfamily and therefore are important for the clarification of relationships within the Drilonematidae. The taxonomy of Iponematinae was discussed by Ivanova & Neuhaus (2011) who pointed out the necessity for molecular characterisation in discriminating between its two genera, *Iponema* and *Filiponema*, which contain a number of intermediate forms.

Materials and methods

Parasitological procedures

The specimen of *Filiponema* sp. was recovered from the formaldehyde-preserved *Lumbricus friendi* Cognetti, 1904 collected in the French Pyrenees and deposited in the collection of the Natural History Museum in Geneva. The rest of the nematodes described were recovered from live earthworms. Juvenile specimens of *Pheretima* sp. 1 were collected in the Chu Yang Sin National Park, Dak Lak Province, Vietnam (12°52′37″N 108°26′17″E) in June 2008 from the floor of a semi-evergreen forest. Seven adult and juvenile specimens of *Pheretima* sp. 2 were collected at the foothills of Mount Mau Son, Loc Binh District, Lang Son Province, Vietnam (21°52′37.6″N 107°0′15.34″E), in April 2010. Specimens of a black morph of *Drawida ghilarovi* Gates, 1969 were collected in water meadows (*Carex* + *Calamagrostis*) along the River Razdolnaya in Primorsky Krai, Russia (43°33′16″N 131°52′15″E) in 2013-2014. A specimen of Megascolicidae gen. sp. was collected at Initao, Misamis Oriental Prov., the Philippines (08°30′N 124°19′E).

Nematodes were washed with Ringer's solution from the coelom into a watch glass and then picked with a needle. Two or three individuals of each species were frozen for DNA extraction and the rest were fixed by adding hot 4-5% formaldehyde for morphological studies. Formaldehyde-fixed nematodes were then processed to glycerin following the method of Seinhorst (1959) and mounted on permanent slides using the wax ring method. Measurements and drawings were obtained with Zeiss Jenaval and Nikon Eclipse E200 microscopes with drawing attachments. Illustrations were finalised with a WACOM Intuos A4 USB drawing tablet and Adobe Illustrator CS5 following Coleman (2003).

For scanning electron microscopy (SEM), material was re-hydrated after formaldehyde fixation, dehydrated in a graded ethanol series, critical-point dried using a HCP-2 HITACHI dryer, mounted on aluminium stubs and coated with gold in a BIO-RAD SC502 sputter coater. Specimens were studied in a JCM-6380 LA SEM and CamScan S2 (Cambridge Instruments, UK).

Molecular characterisation and DNA analysis

DNA was extracted from a single nematode of the three species studied using proteinase K digestions as proposed by Holterman *et al.* (2006). The D2-D3 expansion segment of LSU rDNA was amplified with primers proposed by Nadler *et al.* (2007), *i.e.*, LSU 391 and LSU 501.

The LSU rDNA sequence of *Iponema visayanum* sp. n. was deposited in the NCBI GenBank under the accession number KT160020 and that of *Filiponema suifunense* sp. n. under KT160021. For the phylogenetic analysis the sequences similar to those obtained were found in NCBI GenBank with BLAST (Altschul *et al.*, 1990). The sequences were aligned using Clustal X with default values for gap opening and gap extension penalties. Alignments were analysed with MEGA5 (Tamura *et al.*, 2011) for maximum parsimony (MP), maximum likelihood (ML) and neighbour joining (NJ) and MrBayes 3.1 (Huelsenbeck & Ronquist, 2001) for Bayesian inference (BI). GTR + G + I model was selected for ML and BI analyses with ModelTest 3.5. according to Akaike Information Criterion.

Results

Filiponema champa[*] sp. n.
(Figs 1-3)

Measurements

See Table 1.

[*] The species name refers to the historical name of the area (Dak Lak province) where the earthworm hosts were collected.

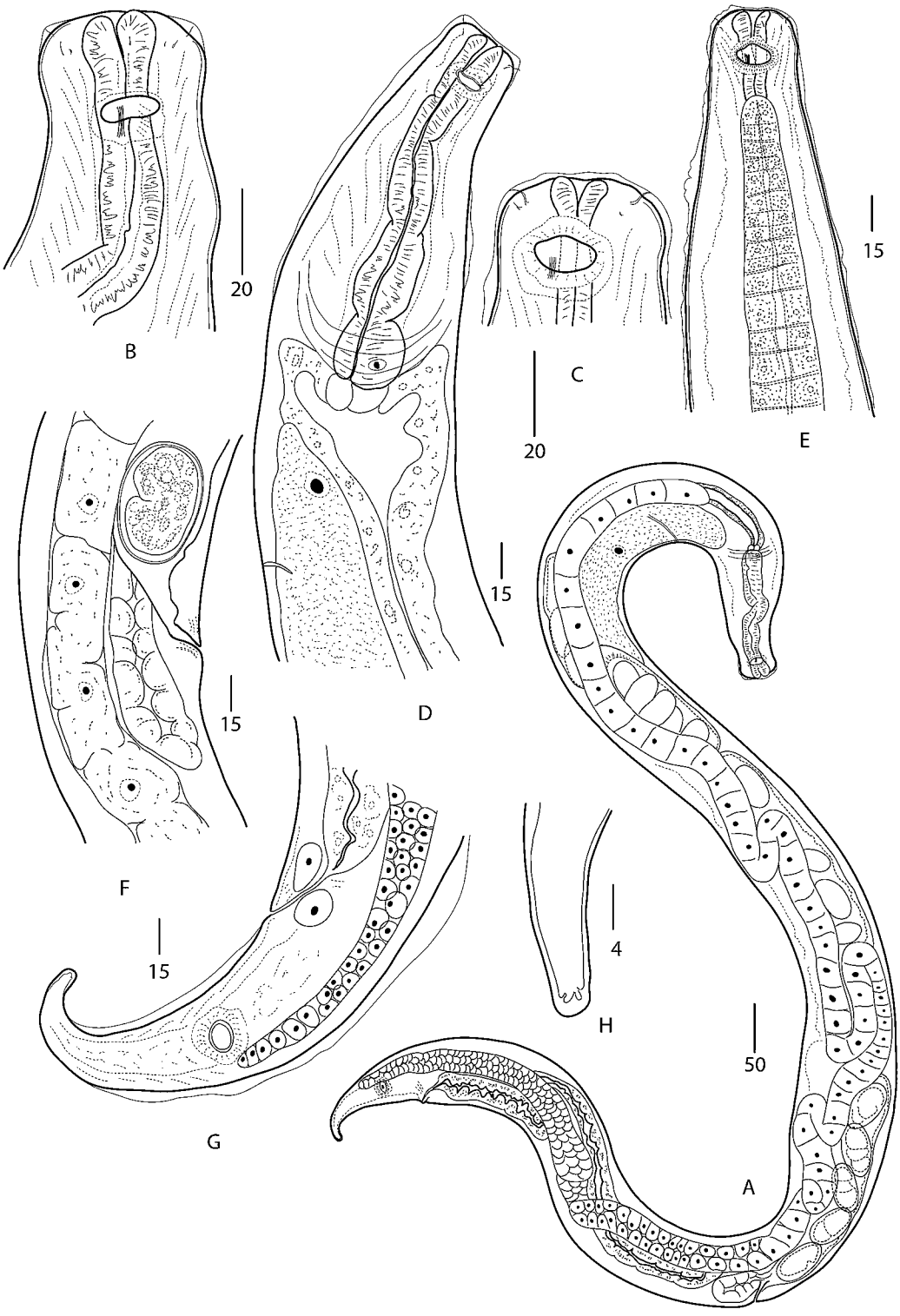

Fig. 1. *Filiponema champa* sp. n. Female. A: Entire worm; B: Head, left lateral view; C: Head, right lateral view; D: Anterior part, lateral view; E: Lateral field at anterior; F: Vulval region, lateral view; G: Tail, lateral view; H: Tail tip, lateral view. (Scale bars in μm.)

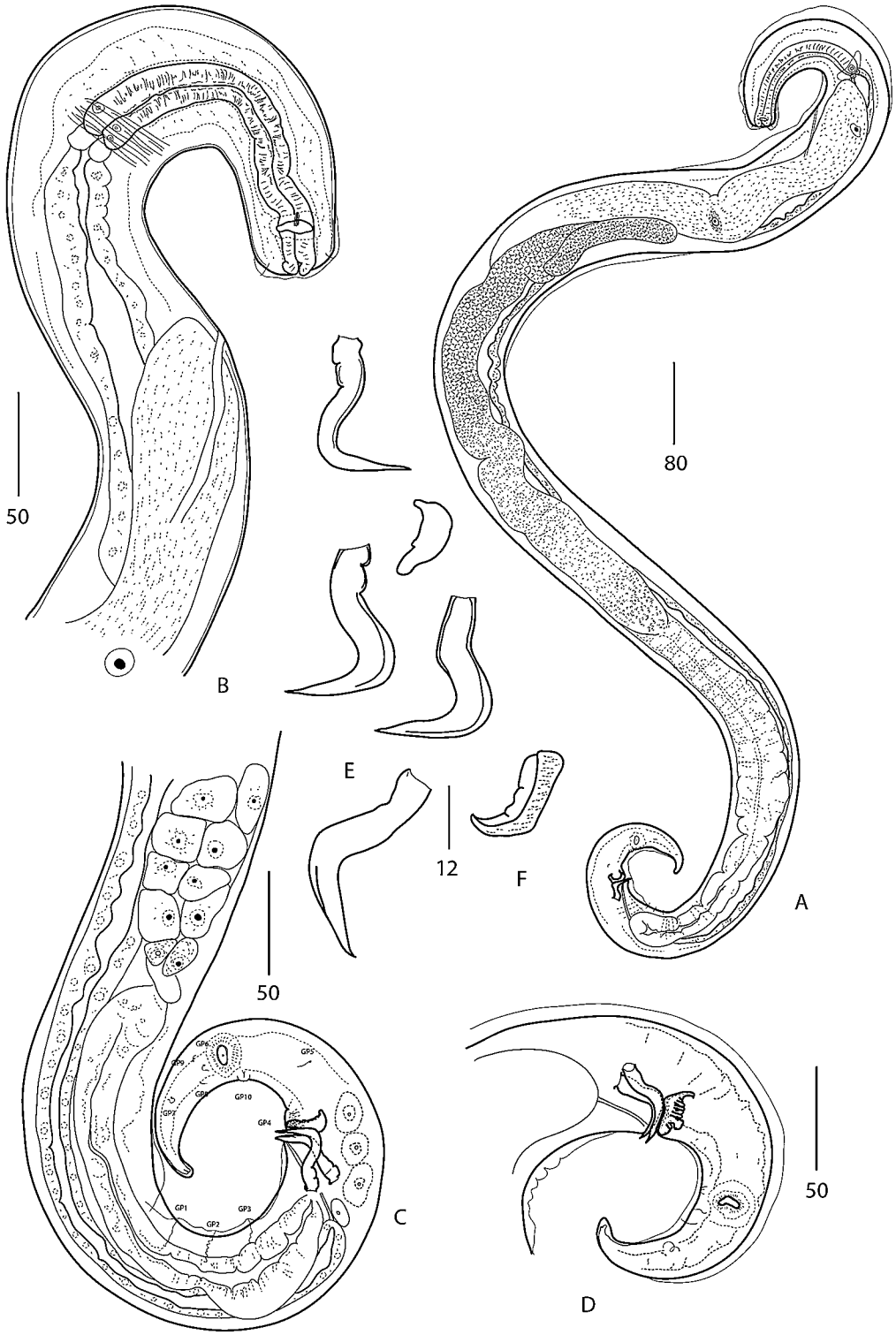

Fig. 2. *Filiponema champa* sp. n. Male. A: Entire worm; B: Anterior region, lateral view; C: Posterior region, lateral view; D: Tail, lateral view; E: Spicules, lateral view; F: Gubernaculum, lateral view. Abbreviations: GP = genital papillae. (Scale bars in μm.)

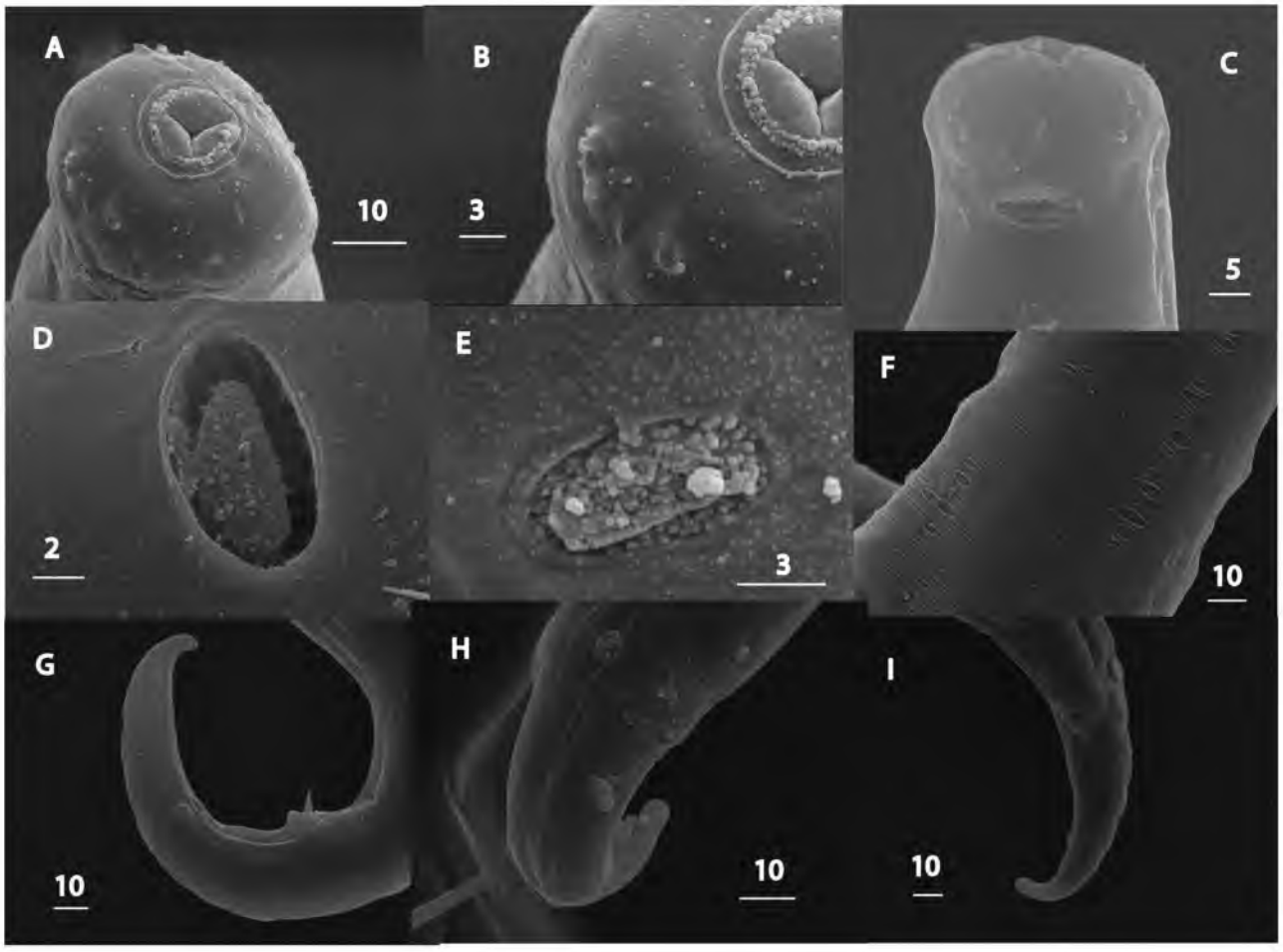

Fig. 3. *Filiponema champa* sp. n. SEM images. A: Female head; B: The same, group of subdorsal papillae; C: Male head; D: Amphid (female); E: Caudal organ (female); F: Cuticular membrane at mid-body (female); G: Male tail, lateral view; H: Male tail, dorsal view; I: Female tail, lateral view. (Scale bars in μm.)

Description

Adults

No marked sexual dimorphism except in sexual characters. Body wide, cylindroid in mid-region, slightly tapering towards anterior extremity. Thin, irregular, cuticular membrane covering entire body present. Cuticle transversely striated, 1 μm thick. Lateral fields faint, 10-28 μm wide, extending from short distance posterior to amphids to mid-tail, marked by widely set apart transverse striations. Head bluntly rounded, with slightest constriction at level of amphids (stump-like). No apparent lips. Area around mouth aperture marked by thin border. Cephalic sensilla papilliform with protruding bristles, located some distance from mouth aperture, ten (6 + 4), subventral and subdorsal sensilla arranged in groups of two. Amphids without obvious pouch. Amphidial apertures large, at least one-third of corresponding diam. wide, transversely elliptical, located 0.5-0.7 corresponding diam. from apex, aperture surrounded by thin rim. Outer rim observed in some specimens as a wide, radially striated band without distinct margins. Group of *ca* 8 μm long sensilla visible inside amphidial aperture. Mouth aperture small, triangular, leading to small funnel-shaped stoma. Pharynx relatively short, rather cylindroid, moderately expanded at anterior and at base. Small anterior bulb separated from corpus by slight constriction. Dorsal sector of pharynx always slightly protruding at apex. Isthmus not pronounced. Nerve ring surrounding pharynx base. Cardia and intestine well developed. Intestinal lining mildly sclerotised.

Table 1. Morphometrics of *Filiponema champa* sp. n. All measurements are in μm and in the form: mean ± s.d. (range).

Character	Male		Female
	Holotype	Paratypes	Paratypes
n	–	4	8
L	1585	1641 ± 58 (1600-1723)	2124 ± 167 (1844-2292)
a	19.8	20.6 ± 3.5 (15.5-23.4)	24.6 ± 4 (20.3-31.7)
b	9.6	10.8 ± 0.9 (9.4-11.5)	14.8 ± 1.7 (13.1-18.7)
c	11.3	12.9 ± 0.7 (11.9-13.4)	12.9 ± 0.8 (11.2-14.0)
V	–	–	60.8 ± 4.4 (53.8-69.1)
Mid-body diam.	80	82 ± 15 (70-103)	88 ± 11 (72-98)
Pharynx	165	153 ± 12 (143-170)	145 ± 17 (115-174)
Head to excretory pore	245	248 ± 27 (215-280)	224 ± 27 (179-257)
Head to nerve ring	160	148 ± 11 (140-165)	139 ± 17 (115-170)
Tail	140	128 ± 6 (120-136)	166 ± 20 (132-193)
Spicule (arc)	36	38 ± 4 (32-41)	–
Spicule (chord)	26	29 ± 3 (25-32)	–
Gubernaculum	15	17 ± 3 (14-21)	–
Egg length	–	–	53 ± 3 (50-58)
Egg diam.	–	–	32 ± 4 (27-36)

Excretory pore situated well posterior to pharynx base, *ca* 2 μm wide; excretory duct 20-30 μm long, weakly sclerotised, reaching body wall at acute angle. Excretory gland enormous, with milky, non-transparent content. Pair of prominent nuclei, one opposite excretory pore and another at mid-length of gland present. Tail curved, broadly conical with blunt, relatively wide (5-6 μm), attenuated, nearly cylindrical tip *ca* 20 μm long. Caudal organs at mid-tail, symmetrically positioned, similar in shape to amphids but with a slightly wider rim and two sensilla, apertures *ca* one third corresponding diam. wide.

Female

Monodelphic, prodelphic. Ovary tip located in tail region. Gonadal tube wide, running anteriad meanderingly then turning posteriad at 218 ± 46 (125-257) μm from pharynx base. Developing oocytes initially in three, then two, and finally one row. Spermatheca not distinctly offset, situated at some distance from flexure and usually obscured by ascending branch of gonadal tube. Oviduct and uterus not morphologically distinguished. Vulva postmedian. Vagina short, oblique. Vulval lips small. Up to five fully developed eggs in uterus; eggshells ovoid, rather small, smooth, length:diam. ratio = 1.5:1. Short, nonfunctional post-uterine sac (*ca* 1-1.5 egg lengths long) present. Tail ventrally or, rarely, dorsally curved, short, *ca* two anal body diam. long. Rectum and anus present. Caudal organs located anterior to mid-tail, its apertures 7 ± 2 (6-11) μm long and 11 ± 3 (8-18) μm wide, outer structures 21 ± 4 (15-28) μm long and 18 ± 7 (11-27) μm wide, rim *ca* 3 μm thick.

Male

Slightly shorter and slimmer than female. Anterior end as in female. Monorchic. Testis reflexed at 386 ± 48 (322-439) μm from anterior end; flexure 166 ± 40 (120-190) μm long. Growth zone wide, very long, with uniform small numerous spermatocytes, *vas deferens* and ejaculatory duct distinct, separated by slight constriction. Spicules paired, equal, short, not distinctly cephalate, bent at right-angle at middle, distal tips pointed, slightly curved ventrally. Gubernaculum massive, boat-shaped. Tail curved ventrally. Caudal sensilla (GP) presented by three pairs of hair-like, precloacal, lateral (GP1-3) sensilla, and seven pairs of postcloacal sensilla comprising six pairs of bristle-like sensilla (GP4-9) plus a pair of prominent subventral papillae located anterior to caudal organs (GP10). GP4 located subventrally just posterior to cloaca, GP5 situated laterally between cloaca and caudal organ, GP6 and GP7 laterally posterior to caudal organ at mid-line of lateral field, GP6 just posterior to caudal organ and GP7 *ca* 20 μm posterior to caudal organ, GP8 and GP9 positioned laterally at margins of lateral field just posterior to GP6. Caudal organs located at mid-tail, smaller in size than in female.

Type host and locality

Coelomic cavity of *Pheretima* sp. (*sensu* Kinberg, 1867), identified by Dr Thai Tran Bai, collected in the Chu Yang Sin National Park, Dak Lak Province, Vietnam (12°52′37″N 108°26′17″E), in June 2008 from the floor of a semi-evergreen forest.

Type material

Holotype male (catalogue number MHNG-INVE-91828), one paratype male (MHNG-INVE-91829), two paratype females (MHNG-INVE-91831-91831) deposited in the Muséum National d'Histoire Naturelle, Geneva, Switzerland. A paratype female (catalogue number 1263) and paratype male (catalogue number 1264) are deposited in the Museum of the Helminthological collections of the Centre of Parasitology at the Severtsov Institute of Ecology and Evolution, Moscow.

Diagnosis and relationships

The present species is characterised by the presence of ten cephalic sensilla, large, transversely elliptical amphidial apertures without an apparent pouch, small funnel-shaped stoma, pharynx nearly cylindroid with the slight expansion at ends, nerve ring at the pharynx base, excretory pore positioned posterior to the pharynx base, tail widely conical with a short, wide, cylindrical tip in both sexes, symmetrically disposed caudal organs similar in structure to amphids, female with a post-median vulva, posteriorly inclined vagina and short post-uterine sac, male with equal, broad, curved spicules and a massive gubernaculum. It was assigned to the genus *Filiponema* on the basis of the presence of the large amphids, the arrangement of cephalic papillae and the symmetrical position of caudal organs. From the rest of the species of *Filiponema*, *F. champa* sp. n. differs by the presence of elliptical *vs* circular to broadly elliptical amphidial apertures and peculiar shape of head and tail ends.

Due to its similar pharynx shape and the position of a nerve ring and an excretory pore, *F. champa* sp. n. is closest to *F. cylindropharyngatum* Ivanova & Neuhaus, 2011 and *F. javanicum* Ivanova & Neuhaus, 2011 but differs in having a wider body, longer spicules and more conspicuous excretory pore. It is also related to *F. baviense* Spiridonov & Ivanova, 1997 in having similarly sized spicules and a pharynx with anterior bulb but is distinguished by more numerous cephalic (ten *vs* four) and caudal sensilla (ten *vs* four), more anterior position of a nerve ring, a different shape of spicules and a gubernaculum and a smaller gubernaculum.

Filiponema champa sp. n. differs from *F. philippinense* Timm & Maggenti, 1966, *F. burmense* Timm, 1967 and *F. sarmathicum* Spiridonov, Kozodoi & Khrustalev, 1989, by having a cylindroid *vs* clavate pharynx.

Prevalence and intensity

71.4% (15 out of 21) earthworms were infected with 4.2 (1-8) nematodes per host.

Filiponema yaoense* **sp. n.**
(Figs 4, 5)

Measurements

See Table 2.

Description

Adults

Body long and moderately slim, cylindroid in middle, very slightly tapering to anterior extremity and more distinctly so to tail. Thin cuticular membrane distinct on either end of body. Cuticle *ca* 1 μm thick, transversely striated. Lateral fields faint, *ca* 10 μm wide at mid-body. Head rounded. Lips not apparent. Mouth aperture circular, surrounded by narrow, low rim. Cephalic sensilla bristle-like, 10 (6 + 4), subventral and subdorsal sensilla arranged in groups of two. Amphids large, *ca* 0.5 corresponding head diam. wide, apertures circular with rim, situated 0.5 head diam. from apical region. Stoma tiny, funnel-shaped. Pharynx short, clavate, with small distinct anterior bulb followed by constriction, straight corpus expanding to base, no isthmus and medium-sized pyriform bulb. Nerve ring surrounding anterior of bulb. Excretory pore located well posterior to pharynx base, duct *ca* 1.5 μm diam., cuticularised. Excretory gland large, excretory channels long, convoluted, weakly cuticularised. Tail shape similar in both sexes, cylindrical anterior to caudal organs and then tapering to a broad rounded tip. Caudal organs located anterior to mid-tail, circular, salient, symmetrical, with thin membrane half-covering aperture, narrow flat rim and slightly convex outer, radially striated rim and single sensillum inside aperture.

* The species name reflects the name of the Yao ethnic group in Lang Son Province, Vietnam.

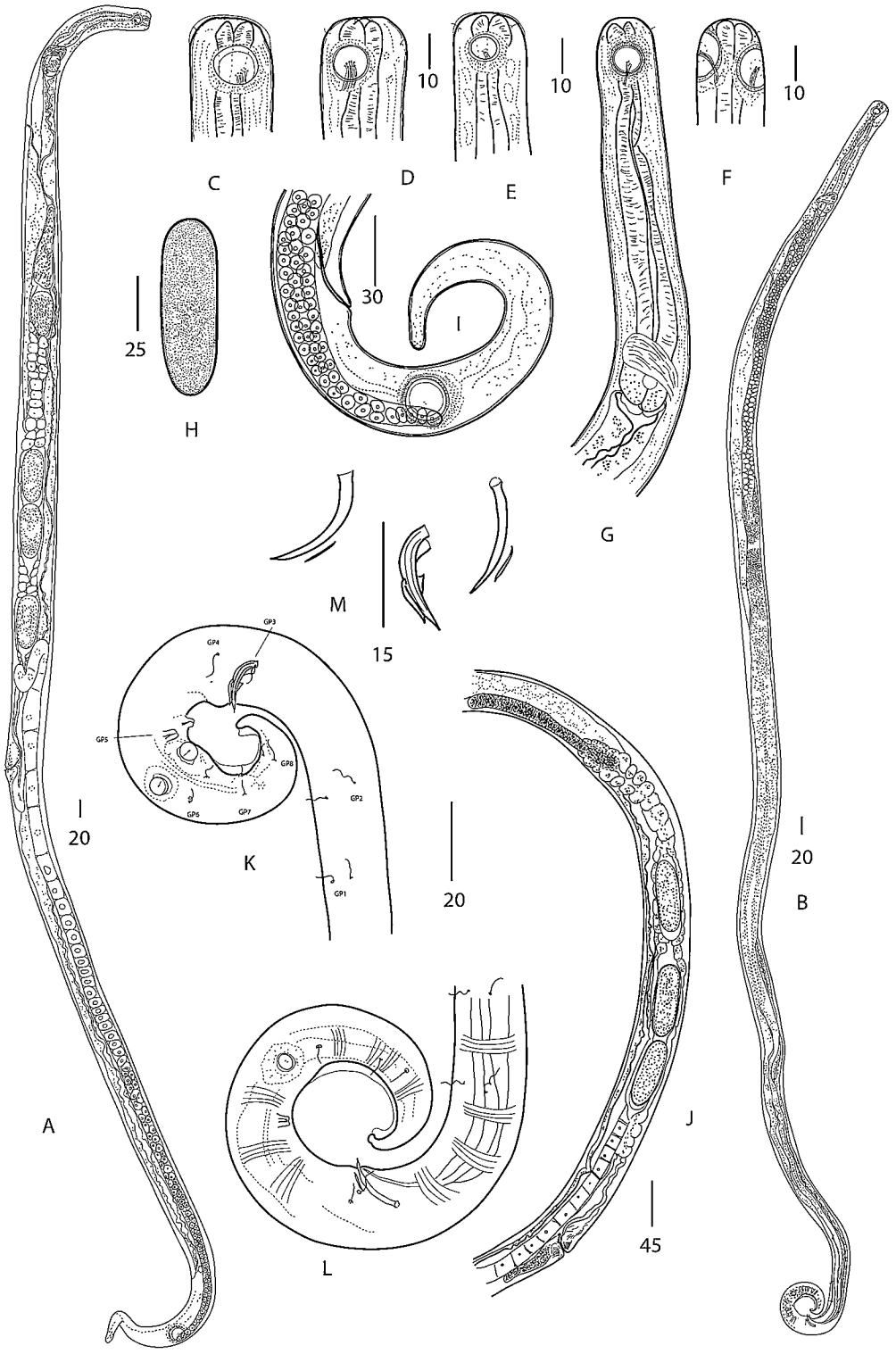

Fig. 4. *Filiponema yaoense* sp. n. Female: A, C-E, G-J. Male: B, F, K-M. A, B: Entire worm; C-E: Head, lateral view; F: Head, dorsal view; G: Anterior part, lateral view; H: Egg; I: Tail, lateral view; J: Vulval region, lateral view; K: Tail, subventral view; L: Tail, lateral view; M: Spicules and gubernaculum, lateral view. Abbreviations: GP = genital papillae. (Scale bars in μm.)

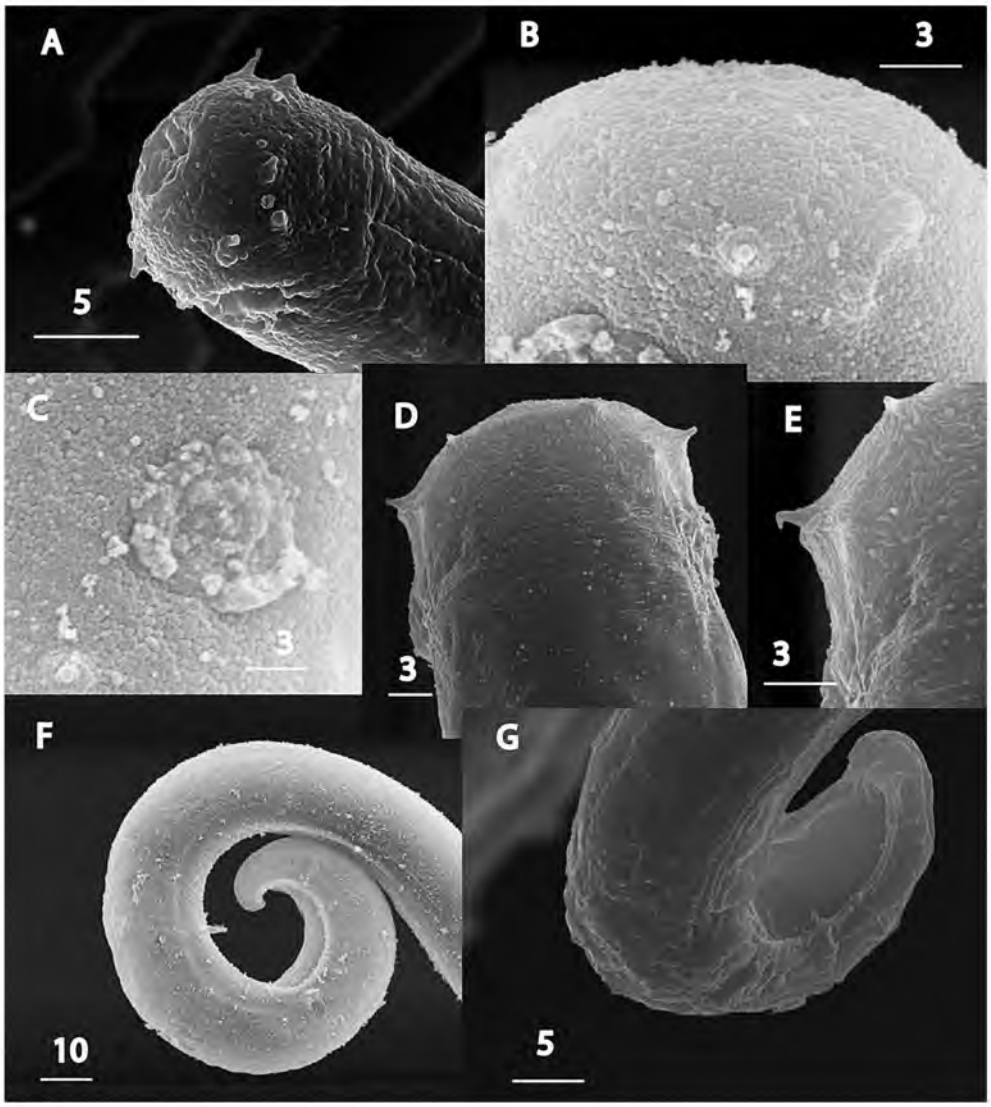

Fig. 5. *Filiponema yaoense* sp. n. SEM images. A: Female head; B: Group of subdorsal papillae (male head); C: Amphid (female); D: Male head; E: Same head, a group of lateral sensilla; F: Male tail, lateral view; G: Male tail, subdorsal view. (Scale bars in μm.)

Female

Amphidial apertures 8-12 μm diam. Monodelphic, prodelphic. Ovary tip located in tail region. Gonadal tube wide, running anteriad on dorsal side, making 1-2 loops at vulva level and reflexing at 316 $\pm$ 3 (314-319) μm from anterior extremity, forming spermatheca at this point. Spermatheca thin-walled, 116 $\pm$ 36 (85-155) μm long and 25 $\pm$ 3 (22-27) μm wide. Oviduct composed of large rounded cells. Uterus containing 1-2 mature eggs. Eggs elongated, egg shells dotted. Vulva located just posterior to mid-body. Vagina slightly oblique, short. Vulval lips small. Non-functional post-uterine sac 46 $\pm$ 2 (45-48) μm long and 14 $\pm$ 2 (12-15) μm wide present. Rectum and anus present. Caudal organs at mid-tail, apertures 14 μm diam., situated 69 $\pm$ 2 (67-70) μm from anus.

Male

Body shape and anterior end structure as in female. Amphidial apertures 10-11 μm diam. Monorchic. Testis reflexing at 174-200 μm from apical region, flexure 114-115 μm long. Tail short, ventrally curved, gradually tapering posterior to caudal organs, tip short, broadly elon-

Table 2. Morphometrics of *Filiponema yaoense* sp. n. All measurements are in μm.

Character	Female		Male
	Holotype	Paratypes	Paratypes
n	–	2	2
L	1863	1788, 1945	1293, 1649
a	41.1	44.7, 46.3	41.7, 48.5
b	11.2	12.7, 12.8	9.9, 10.8
c	9.6	8.3	12.3, 15.3
V	54.4	56, 58.4	–
Mid-body diam.	45	40, 42	31, 34
Pharynx	167	141, 152	130, 153
Head to excretory pore	221	229, 237	180, 247
Head to nerve ring	145	132, 134	110, 133
Tail	194	215	105, 108
Spicule (arc)	–	–	18, 19
Spicule (chord)	–	–	15, 17
Gubernaculum	–	–	6, 8
Egg length	68	73, 77	–
Egg diam.	26	20, 25	–

gated. Two pairs of long, hair-like precloacal sensilla (GP1, GP2), two pairs of similar adcloacal sensilla (GP3, GP4) and four pairs of postcloacal sensilla, of which GP5 are salient papilliform sensilla located anterior to caudal organs and GP6-GP8 are hair-like sensilla located posterior to caudal organs. All sensilla located in subventral position. Spicules slightly curved, thin, weak, simple, not distinctly cephalate, short, gubernaculum stick-like, thin. Caudal organs smaller than amphidial apertures, 8 μm diam., symmetrically positioned at mid-tail.

Type host and locality

Coelomic cavity of *Pheretima* sp. (*sensu* Kinberg, 1867), identified by Dr Thai Tran Bai, collected at the foothills of Mount Mau Son, Loc Binh District, Lang Son Province, Vietnam (21°52′37.6″N 107°0′15.34″E), in April 2010.

Type material

Holotype female (catalogue number MHNG-INVE-91832) and one paratype male (catalogue number MHNG-INVE-91833) deposited in the Muséum National d'Histoire Naturelle, Geneva, Switzerland. A paratype male (catalogue number 1265) and paratype female (in two pieces, catalogue number 1266) are deposited in the Museum of the Helminthological collections of the Centre of Parasitology at the Severtsov Institute of Ecology and Evolution, Moscow.

Diagnosis and relationships

Filiponema yaoense sp. n. is characterised by its long, slim body, a head bearing ten sensilla, large, circular amphids without an apparent pouch, which are situated 0.5 head diam. from the apical extremity, a pharynx with a small anterior bulb and a straight corpus expanding towards the base, a nerve ring positioned at the posterior part of the pharynx, an excretory pore located posterior to the pharynx base, long, cuticularised excretory duct, caudal organs placed symmetrically, tail of both sexes tapering posterior to caudal organs with a wide rounded tip, female with vulva located just posterior to mid-body, a short post-uterine sac and slightly inclined vagina, male with short, weak spicules and tiny gubernaculum.

Filiponema yaoense sp. n. differs from all known species of *Filiponema* by its long, conspicuous excretory duct and a nerve ring which is situated closer to the anterior extremity (*i.e.*, at the posterior part of pharynx *vs* at pharynx base or posterior to it). It is similar to *F. cylindropharyngatum*, *F. javanicum* and *F. daklakense* sp. n. by having a similar pharynx shape. From *F. cylindropharyngatum* it can be distinguished by the wider body and pharynx, a rounded *vs* pointed tail, spermatocytes arranged in two *vs* one row, symmetrically *vs* asymmetrically placed caudal organs and cephalic sensilla arranged in one *vs* two circles; from *F. javanicum* by the slimmer body, shorter spicules, longer eggs and the cephalic sensilla arranged in one *vs* two circles; from *F. champa* sp. n. by a slimmer body, the more anterior position of the amphids, circular *vs* transversely elliptical amphids and caudal organs, a wider tail tip, shorter male tail, longer eggs, smaller, straighter spicules and a smaller gubernaculum.

By having a similar pharynx with an anterior bulb it is comparable to *F. baviense* but is differentiated by more numerous cephalic (ten *vs* four) and caudal sensilla (eight *vs* four), much smaller and differently shaped spicules and a gubernaculum. From *F. philippinense* Timm & Maggenti, 1966, *F. burmense* and *F. sarmathicum*, *F. yaoense* sp. n. can be immediately differentiated by having a cylindroid *vs* clavate pharynx.

Prevalence/intensity

A single host specimen was infected by eight females and nine males.

Filiponema suifunense[*] sp. n.
(Figs 6, 7)

Measurements

See Table 3.

Description

Female

Thin cuticular membrane visible in few specimens. Body short and relatively wide, with max. diam. at middle, gradually tapering to both ends. Cuticle clearly and evenly annulated. Lateral fields prominent, *ca* 5-7 μm wide at mid-body, extending from short distance posterior to amphid to tail tip, broken only by presence of caudal organ. Head rounded, no lips present. Mouth aperture small, triangular. Six small papillae situated around mouth aperture. Four salient papillae with protruding bristles of outer circle distanced from inner circle of papillae. Amphids *ca* one head diam. from apical extremity, amphidial aperture 10 ± 2 (8-15) μm in diam., or *ca* two-thirds of corresponding diam., surrounded by rim 4 ± 1 (2-5) μm wide. No obvious pouch. Bundle of 6-7 long sensilla visible inside amphidial aperture. Stoma reduced. Pharynx muscular, cylindroid with well developed anterior bulb and insignificant expansion at base. Nerve ring located at pharynx base or anterior to it. Cardia and intestine well developed, intestine lining thickened, rectum wide, anus and rectal glands present. Excretory pore *ca* 1 μm wide, situated halfway between pharynx base and spermatheca. Excretory duct as narrow as pore, mildly cuticularised, 67 ± 4 (60-72) μm long. Excretory cell large, with conspicuous, long channels inside. Monodelphic, prodelphic. Ovary tip located in tail region, developing oocytes slightly flattened, ovary wide, running anteriad on dorsal region of body and at 275 ± 86 (210-510) μm from anterior extremity turning posteriad. Spermatheca 82 ± 19 (46-98) μm long and 34 ± 10 (20-47) μm wide, located at gonad flexure. Oviduct composed of large flattened cells. Uterus thin-walled. One to three elongated eggs in uterus, eggshell thinly and finely mamillate. Vagina muscular, short, straight to very slightly inclined. Vulva post-median, vulval lips flat. Post-uterine sac 26 ± 4 (20-30) μm long, reduced. Tail wide, truncate-conoid, slightly curved ventrad. Tail tip broad, slightly asymmetrical, sometimes with 1-2 tiny appendages. Caudal organs symmetrically located at 71 ± 7 (60-85) μm from anus, similar to amphids in appearance, apertures 16 ± 2 (13-19) μm in diam. One or two sensilla protruding from aperture.

Male

Not found.

Type host and locality

Coelomic cavity of *Drawida ghilarovi* Gates, 1969, identified by Dr G.N. Ganin, collected in water meadows (*Carex* + *Calamagrostis*) along the River Razdolnaya in Primorsky Krai, Russia (43°33′16″N 131°52′15″E), in June 2013 and July 2014.

Type material

Holotype female (catalogue number MHNG-INVE-91834) and two paratype females on the same slide (catalogue number MHNG-INVE-91835) deposited in the Muséum National d'Histoire Naturelle, Geneva, Switzerland. Two paratype females (catalogue numbers 1267-1268) deposited in the Museum of the Helminthological collections of the Centre of Parasitology at the Severtsov Institute of Ecology and Evolution, Moscow.

Diagnosis and relationships

The species is characterised by the presence of four salient cephalic papillae in an external circle and six small ones in an internal circle, amphids with large circular apertures with a rim and no pouch, reduced stoma, a cylindroid pharynx with the small anterior bulb and slight basal expansion, a nerve ring at the pharynx base or anterior to it, an excretory pore located posterior to the pharynx, a wide, truncate-conoid tail, symmetrically disposed caudal organs similar in structure to amphids, female with a post-median vulva, nearly straight vagina and short post-uterine sac. Males unknown.

Filiponema suifunense sp. n. differs from all known species of the genus by the presence of narrow, projecting lateral fields. It is closest to *F. sarmathicum* and *F. javanicum* due to its similar tail shape and the arrangement of cephalic sensilla. It can be differentiated from *F. sarmathicum* by the pharynx shape (cylindroid with an anterior bulb *vs* clavate), the more anterior position of the nerve ring and more elongated eggs with mamillated egg-shells.

[*] The species name refers to the historical name of the River Razdolnaya, the collection site.

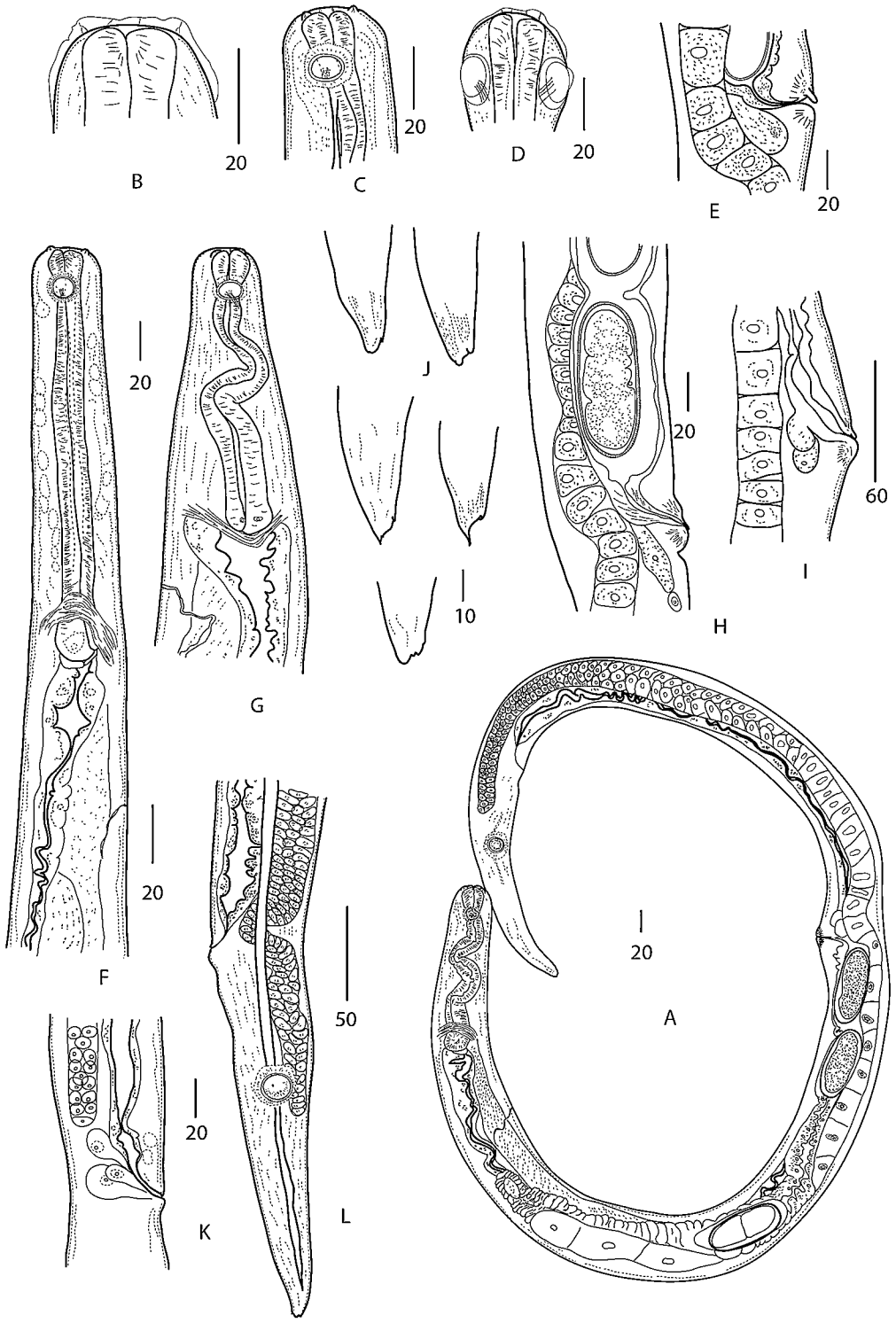

Fig. 6. *Filiponema suifunense* sp. n. Female. A: Entire worm; B: Head end, lateral view; C: Head, lateral view; D: Head, ventral view; E: Vulval region, lateral view; F, G: Anterior region, lateral view; H: Uterus with egg and straight vagina, lateral view; I: Inclined vagina, lateral view; J: Tail tips, lateral view; K: Anal region, lateral view; L: Tail, lateral view. (Scale bars in μm.)

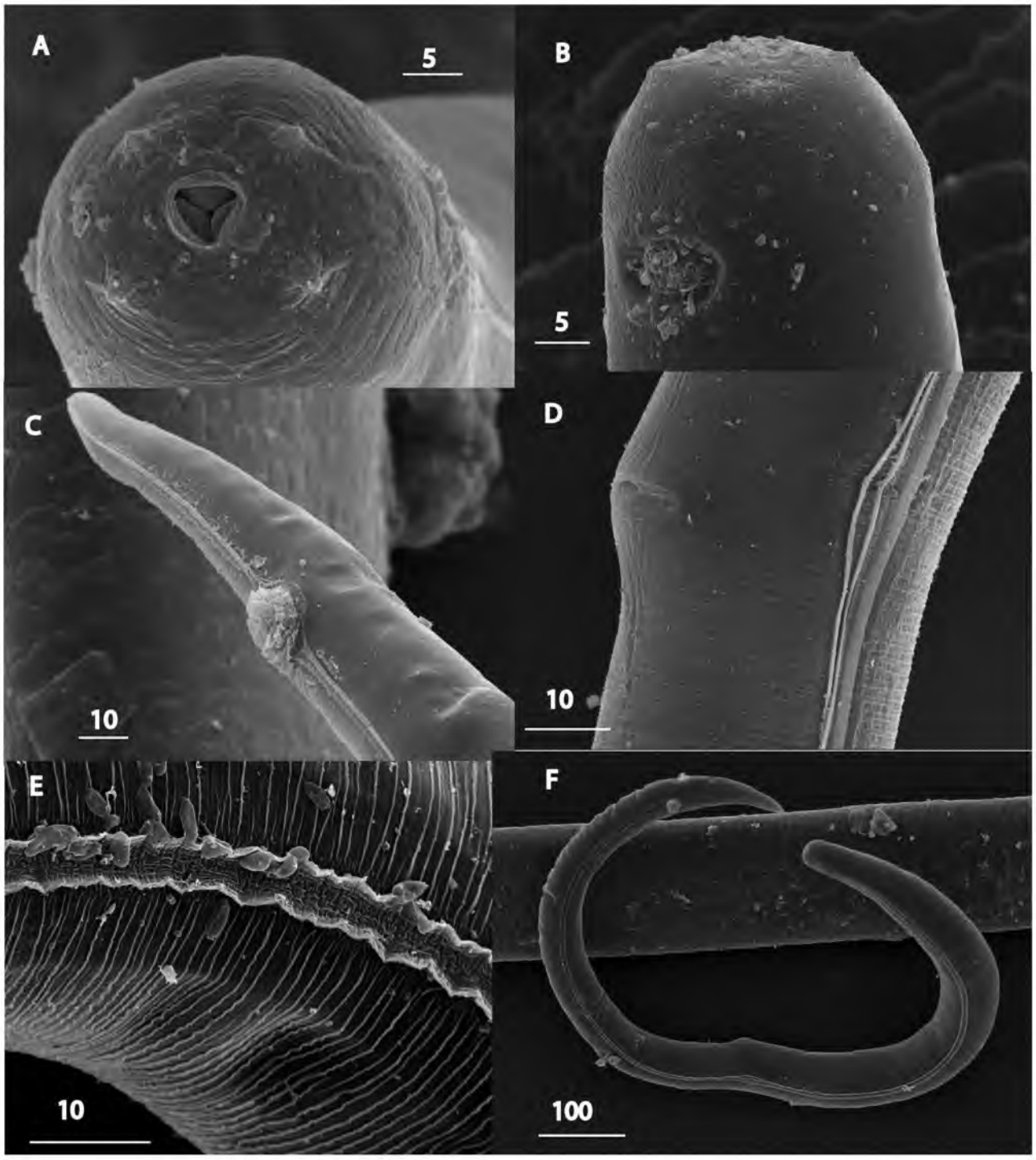

Fig. 7. *Filiponema suifunense* sp. n. SEM images. Female. A: Head, sub-apical view; B: Head, lateral view; C: Tail; D: Vulval region; E: Lateral field at mid-body; F: Entire worm. (Scale bars in μm.)

Table 3. Morphometrics of female *Filiponema suifunense* sp. n. All measurements are in μm and in the form: mean ± s.d. (range).

Character	Holotype	Paratypes
n	–	15
L	1669	1474 ± 190 (1293-2023)
a	27.8	21.9 ± 3.2 (16.8-28.9)
b	7.8	7.8 ± 0.7 (6.6-9.0)
c	7.6	7.0 ± 0.8 (6.2-9.1)
V	55.1	54.8 ± 2.6 (49.1-59.1)
Mid-body diam.	60	68 ± 5 (62-81)
Pharynx	215	189 ± 15 (171-215)
Head to excretory pore	300	221 ± 38 (156-290)
Head to nerve ring	180	159 ± 21 (130-185)
Tail	221	210 ± 13 (180-229)
Egg length	72	71 ± 4 (66-79)
Egg diam.	27	31 ± 3 (27-37)

It is comparable to *F. javanicum* in the presence of a cylindroid pharynx but can be distinguished by having an anterior bulb, the more anterior position of a nerve ring, a distinct excretory pore and duct (*vs* poorly distinguishable), significantly elongated *vs* ovoid eggs with ornamented *vs* smooth egg shells and truncated tail tip bearing 1-2 minute appendages *vs* rounded tip.

Filiponema suifunense sp. n. is similar to *F. philippinense*, *F. baviense*, *F. champa* sp. n. and *F. yaoense* sp. n. due to the presence of an anterior bulb. It can be easily differentiated from *F. baviense* by having ten *vs* four cephalic sensilla and a truncated *vs* pointed tail; from *F. philippinense* it can be differentiated by the presence of much longer and narrower eggs with patterned *vs* smooth surface and the much shorter and wider tail; from *F. champa* sp. n. by the arrangement of the cephalic sensilla in two *vs* one circle, circular *vs* elliptical amphids and caudal organs and a truncate-conoid tail *vs* broadly conical one with wide terminus; from *F. yaoense* sp. n. it differs in having a wider body, a truncated *vs* rounded tail tip, an annulated *vs* smooth cuticle and in the arrangement of cephalic sensilla in two *vs* one circle.

From *F. burmense* the present species differs in having ten *vs* four cephalic sensilla, a more anterior nerve ring position, a truncated *vs* rounded tail tip in female and less numerous, elongated eggs with ornamented shells *vs* rounded with smooth eggshells.

From *F. cylindropharyngatum*, to which *F. suifunense* sp. n. is similar in having greatly elongated, ornamented eggs, it differs by a thick, distinctly annulated cuticle *vs* thin and longitudinally striated, a wider body, the lack of an anterior bulb of a pharynx, the more anterior position of the nerve ring, distinct *vs* indistinct excretory pore and duct, a shorter, wider tail, symmetrical *vs* asymmetrical position of caudal organs and the slightly more posterior vulval position.

Prevalence/intensity

In 2013, 37.8% (14 out of 37) earthworms were infected with five (1-23) nematodes whereas in 2014, 63.6% (14 out of 22) earthworms were infected with 3.5 (1-9) nematodes.

Molecular characterisation

The 666 bp long LSU rDNA sequence of *F. suifunense* sp. n. differs by 76 nucleotides from the sequence of *Homungella tonkinense* Spiridonov & Ivanova, 1998 (JF323056), by 77 nucleotides from *I. visayanum* sp. n.; and by >97 nucleotides from all other nematodes with a known homologous sequence.

Filiponema sp.
(Fig. 8)

Description (based on a single female)

Body 2418 μm long and 70 μm wide, cylindroid, gradually tapering to tail; a = 34.5; b = 16.1; c = 8.5. Head rounded. Cuticle longitudinally striated. Cuticular membrane absent or inconspicuous. Lateral fields indistinct. No apparent lips. Six small sensilla situated around mouth aperture. Four hair-like, 4 μm long sensilla located slightly away from mouth aperture. Stoma reduced. Amphidial apertures situated 40 μm from anterior extremity, circular, 28 μm in diam., surrounded by inner, thin rim 1 μm wide and outer, convex, radially striated rim 7 μm wide. No apparent pouch. Bundle of ten long sensilla protruding from amphidial aperture. Pharynx extending to anterior extremity, clavate, 150 μm long. Corpus 108 μm long and 15 μm wide, cylindrical but with slight expansion (15 μm) at anterior, isthmus not pronounced. Basal bulb 42 μm long × 26 μm wide, pyriform. Cardia 11 μm long. Intestine well developed, with thick walls. Nerve ring encircling intestine at 135 μm from pharynx base. Excretory pore situated 135 μm posterior to nerve ring or at 420 μm from anterior extremity, 3 μm wide, followed by mildly cuticularised duct 49 μm long. Excretory channels long, straight, non-sclerotised. Monodelphic, prodelphic. Ovary tip located anterior to anus. De-

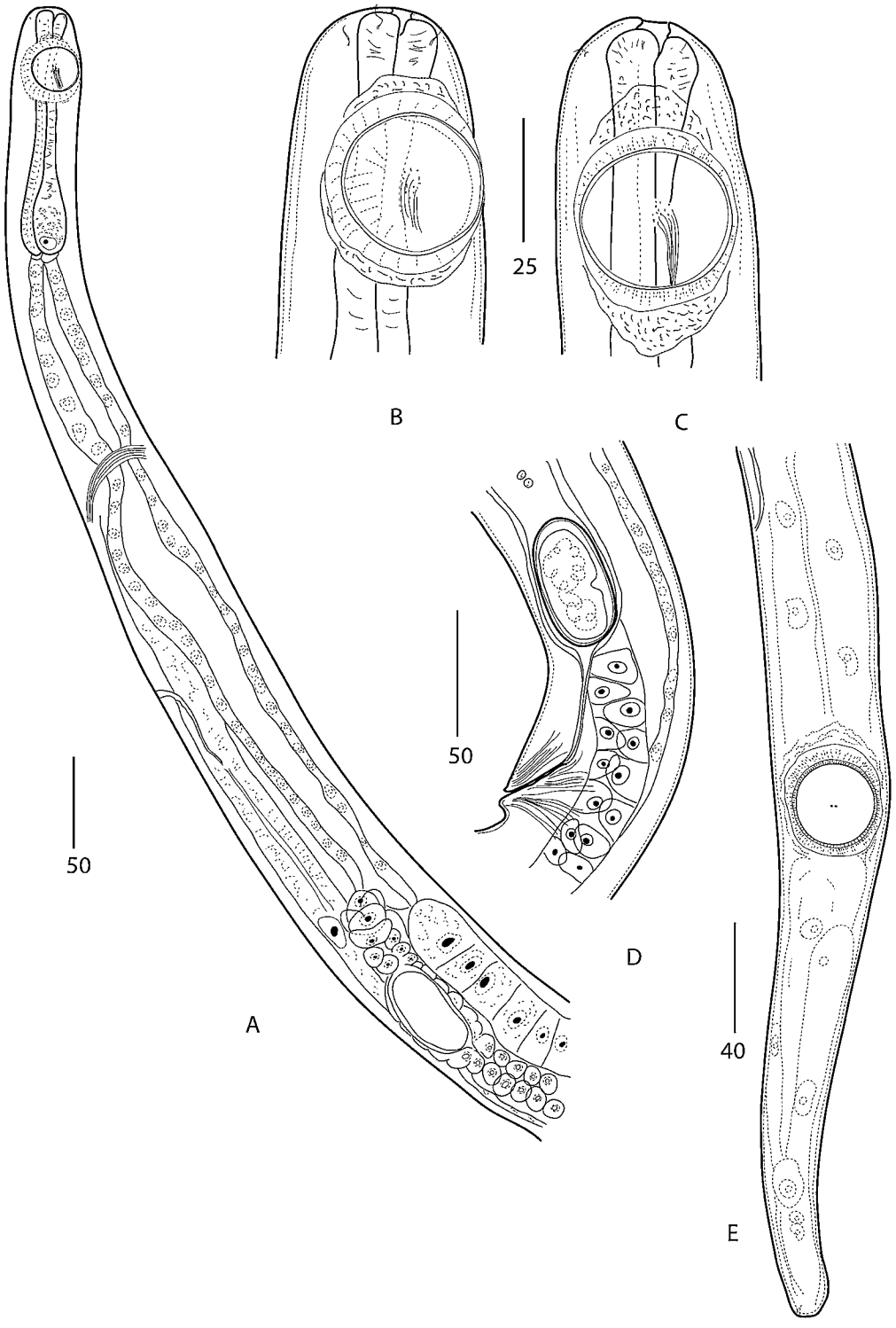

Fig. 8. *Filiponema* sp. Female. A: Anterior region; B: Head end, right side, lateral view; C: Head, left side, lateral view; D: Vulval region, lateral view; E: Tail, lateral view. (Scale bars in μm.)

veloping oocytes initially in one, then three rows of small, spherical cells, then in two rows, and finally in one row of large, square cells. Ovary running anteriad on dorsal region of body then turning posteriad at 584 μm from apex. Spermatheca offset, small, located at flexure. No sperm in spermatheca observed. Oviduct long, composed from large rounded cells. Uterus thin-walled, containing two eggs, eggs smooth, thin shelled, ovoid, 60 $\times$ 33 μm in size. Vulva post-median, V = 63.6. Post-uterine sac absent. Vagina inclined posteriad, *ca* 35 μm long. Vulval lips flat. Rectum cuticularised, rectal glands inconspicuous. Tail 284 μm long, tip bluntly rounded, 13 μm wide. Caudal organs very large, situated symmetrically at anterior third of tail, aperture 37 μm diam., external rim *ca* 5 μm in diam. Two sensilla slightly protruding from aperture.

Remarks

Of all known and described herein members of *Filiponema*, the specimen examined most closely resembles, because of the similar shape of head and tail ends, the pharynx shape, the position of nerve ring and excretory pore, the size and shape of amphids and caudal organs, eggs without ornamentation, *F. sarmathicum*, yet another parasite of the lumbricid earthworms *Eisenia nordenskioldi* (Eisen, 1879) and *Allolobophora caliginosa* (Savigny, 1826). It differs from *F. sarmathicum* by a longer body and tail, much larger amphids (28 *vs* 13-17 μm diam.), and lack of a post-uterine sac and cuticular membrane around the body.

Host and locality

Coelomic cavity of *Lumbricus friendi* Cognetti, 1904, MHNG-INVE-91790 #1131, Behevobie, Basses, Pyrenees, France, 14.7.74, coll. Haymos, det. Zicsi.

Voucher material

Voucher specimen of female (catalogue number MHNG-INVE-91790) deposited in the Muséum National d'Histoire Naturelle, Geneva, Switzerland.

Iponema visayanum* **sp. n.**
(Fig. 9)

Measurements

See Table 4.

Description

Adults

No marked sexual dimorphism except in sexual characters. Body small and slim, cylindroid in mid-course, barely tapering to anterior extremity. Tail in both sexes with long spike-like tip. Cuticle 1 μm thick, lateral fields present, max. diam. 4-5 μm at mid-body. Delicate membrane seen on tail end. Head rounded, no lips, membrane absent. Four bristle-like cephalic papillae located slightly away from tiny mouth aperture. Amphidial aperture medium to large, faint, transversely elliptical, located 9-23 μm from anterior extremity. Bundle of 7-10 long, strong sensilla visible inside amphidial aperture. Faint pouch. No stoma. Pharynx with long, very finely muscled, meandering corpus reaching head end where it slightly expands. Isthmus not pronounced, basal bulb large, elongated, glandular, irregularly shaped. Nerve ring surrounding posterior of corpus. Excretory pore located at level of mid-bulb to bulb-base. Excretory duct 1-2 μm wide, sclerotised, curved, 73-94 μm long. Excretory gland large, containing at least one very large nucleus at its mid-point. Small cardia present. Intestinal lining thickened. Caudal organs slightly asymmetrical, left one located closer to anus than right. Distance between caudal organs *ca* 12 μm. Apertures transversely oval to slit-like, surrounded by slightly raised rim *ca* 2 μm thick, its inner chambers narrow, funnel-shaped.

Female

Amphids located 14 μm from apical region. Amphidial apertures 1-3 μm long and 5-7 μm wide. Corpus 108 $\pm$ 10 (100-120) μm long and 9 μm wide, basal bulb 42 $\pm$ 2 (36-44) μm long and 16 $\pm$ 1 (15-17) μm wide. Monodelphic, prodelphic. Ovary tip located anterior to anus. Ovary wide, running anteriad and twisting around intestine up to vulva level, then going straight until returning midway between vulva and pharynx base, or at 224 $\pm$ 14 (208-234) μm from anterior end. Developing oocytes large, initially in two rows, then one. Spermatheca offset,

* The species name refers to the largest ethnic group in the Philippines.

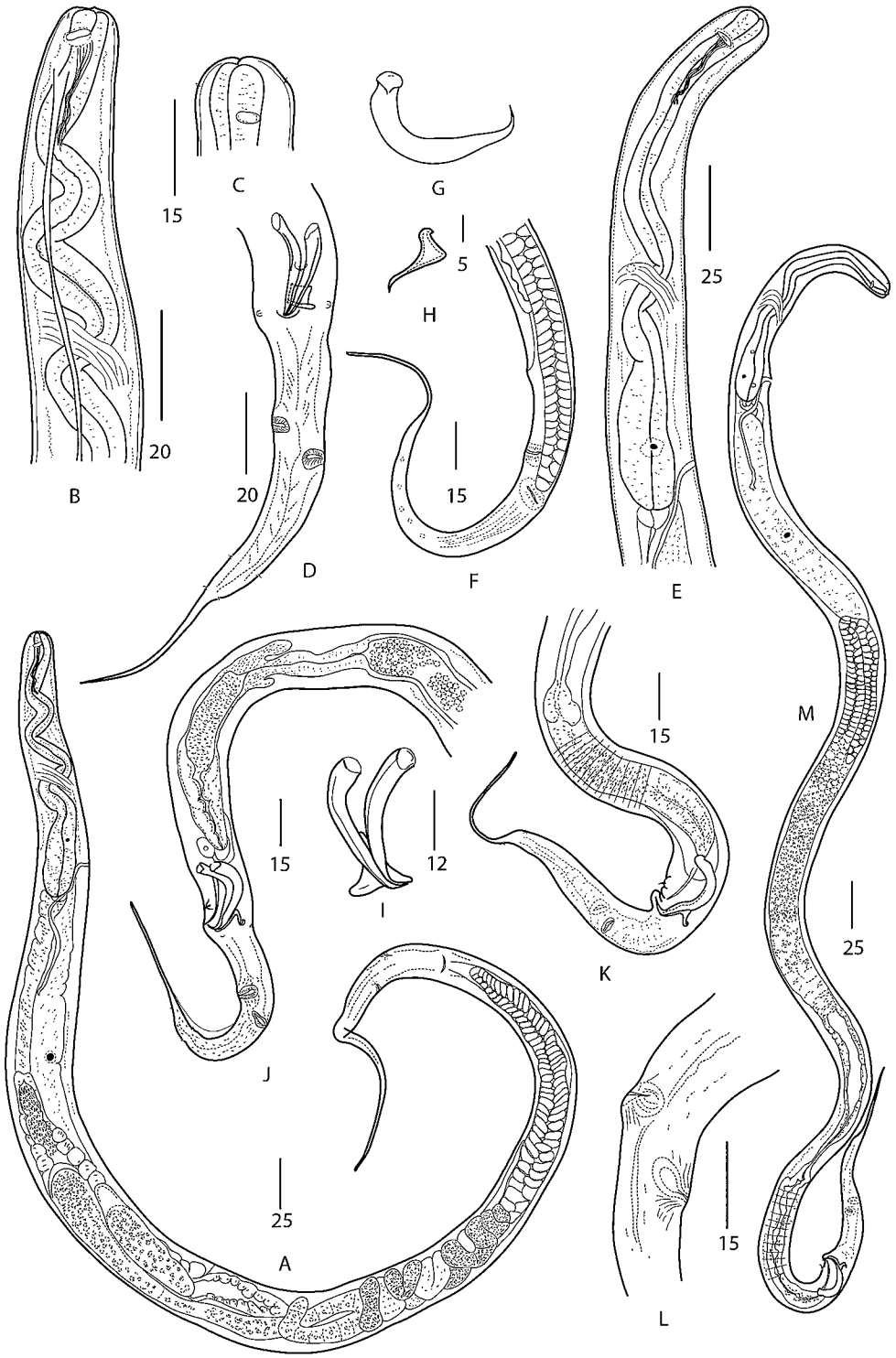

Fig. 9. *Iponema visayanum* sp. n. Female: A, B, F, L. Male: C-E, G-K, M. A, M: Entire worm; B: Anterior end, lateral view; C: Head, lateral view; D: Tail, ventral view; E: Pharynx region, lateral view; F: Tail, lateral view; G: Spicule; H: Gubernaculum; I: Spicules and gubernaculum, subventral view; J: Tail, subventral view; K: Tail, lateral view; L: Caudal organs, ventral view. (Scale bars in μm.)

Table 4. Morphometrics of *Iponema visayanum* sp. n. All measurements are in μm and in the form: mean ± s.d. (range).

Character	Male		Female
	Holotype	Paratypes	Paratypes
n	–	9	3
L	795	821 ± 65 (702-911)	913 ± 83 (861-1009)
a	29.4	30.7 ± 5.8 (24.6-40.4)	30.6 ± 3.1 (27.2-33.1)
b	5.3	5.4 ± 0.4 (4.9-5.9)	14.8 ± 1.7 (13.1-18.7)
c	7.6	7.6 ± 0.8 (6.4-8.9)	6.0 ± 0.1 (5.9-6.1)
V	–	–	42.9 ± 1.2 (42.0-43.7)
Mid-body diam.	27	27 ± 4 (20-31)	30 ± 4 (26-32)
Pharynx	150	151 ± 11 (130-166)	152 ± 16 (115-174)
Head to excretory pore	136	134 ± 12 (113-151)	129 ± 16 (120-147)
Head to nerve ring	90	88 ± 9 (80-110)	87 ± 14 (75-102)
Tail	104	109 ± 5 (98-116)	160 ± 5 (156-165)
Spicule (arc)	37	33 ± 3 (28-39)	–
Spicule (chord)	22	23 ± 2 (20-25)	–
Gubernaculum	12	11 ± 2 (8-13)	–
Egg length	–	–	60 (n = 1)
Egg diam.	–	–	17 (n = 1)

57 ± 34 (27-94) μm long × 15 ± 3 (13-18) μm wide, located on flexure. Single immature egg observed 60 × 17 μm in size. Vulva pre-equatorial, not detected in immature females. Vulval lips flat. Short, thick-walled post-uterine sac present. Tail curved ventrally, conoid, with long pointed terminus 30-40 μm long. Rectum and anus present. Left caudal organ 32 ± 7 (28-40) μm from anus, right one 12 ± 1 (12-13) μm posterior to it.

Male

Anterior end structure similar to that in female. Amphidial aperture 3.5 ± 0.8 (2-5) μm long and 5.3 ± 1.2 (3-6) μm wide, located 13 ± 5 (9-23) μm from apex. Corpus 112 ± 9 (98-127) μm long × 6-8 μm wide, basal bulb 39 ± 5 (32-46) μm long × 15 ± 2 (13-18) μm wide. Testis long, reflexing at 231 ± 32 (185-275) μm from anterior end, flexure 88 ± 12 (73-110) μm long; developing spematocytes large, arranged in two rows. *Vas deferens* and ejaculatory duct distinctive. Spicules paired, equal, strongly curved at mid-point, not distinctly cephalate, tips pointed, bent at right-angle in ventral direction, hook-like, measuring 34 ± 3 (28-39) μm along arc and 23 ± 2 (20-25) μm along chord, appearing very long compared to gubernaculum and anal diam. Gubernaculum small, boat-shaped, with dorsal apophysis *ca* one-third its length. Caudal papillae arranged as a prominent precloacal midventral papilla with bristle, a pair of small lateral adcloacal papillae, and two postcloacal pairs of small subventral papillae with short bristles, one at a short distance from the spike base and another at the spike base. Membrane on tail usually visible, low. Caudal organs as in female. Tail curved ventrally conoid with long pointed spike 39 ± 5 (32-47) μm long.

DIAGNOSIS AND RELATIONSHIPS

Iponema visayanum sp. n. is distinctive because of the similarity in appearance of both sexes, the slim body with thin spike-like tail tip, the presence of narrow lateral fields, the rounded head bearing four bristle-like cephalic sensilla, the large amphids with a faint pouch, a pharynx with a slender muscular corpus and elongated, glandular basal bulb, a nerve ring crossing posterior to the pharyngeal corpus, excretory pore opposite the basal bulb, a long, sclerotised excretory duct, slightly asymmetrically disposed caudal organs, curved spicules longer than the anal body diam. and the male tail with a midventral precloacal papilla, a pair of adcloacal papillae and two pairs of postcloacal papillae.

In its small body size and the slightly asymmetrical position of the caudal organs, *I. visayanum* sp. n. most closely resembles *I. tonkinense* Spiridonov & Ivanova, 1998. It can be distinguished from the latter by its longer, more strongly curved spicules (23 μm on chord *vs* 19 μm), a much longer and wider pharynx, and its larger amphids situated further posterior from the anterior extremity. *Iponema visayanum* sp. n. is close

to *I. major* Timm & Maggenti, 1966 and *I. minor* Timm & Maggenti, 1966 in having a similarly shaped body, pharynx, spicules and caudal organs, but can be distinguished by its much smaller size, the more anterior position of the excretory pore, longer excretory duct, and shorter spicules. *Iponema visayanum* sp. n. is similar to *I. australe* Yeates & Spiridonov, 1996 in having four cephalic papillae and a similarly shaped pharynx but is differentiated from it by the more anterior position of the nerve ring (around pharyngeal corpus *vs* intestine), larger amphids with an almost indistinct pouch, the longer tail spike in both sexes, smaller and differently shaped spicules and gubernaculum, and in the different disposition of the caudal papillae. The new species is also close to *I. pheretimae* Timm, 1971 in having four cephalic papillae and a reduced post-uterine sac and a nerve ring positioned around the pharyngeal corpus, but differs in the smaller body size and much shorter tail in both sexes, in the absence of a cuticular lining of the buccal cavity and an anterior bulb, circular *vs* slit-like amphids, and caudal organs closely positioned to each other *vs* 130-144 μm apart. From *I. laotense* Spiridonov, 1994 it differs in its much smaller size, in having a rounded *vs* truncated head with four *vs* ten cephalic sensilla, less numerous male caudal sensilla, spicate *vs* non-spicate male tail, and asymmetrical *vs* symmetrical caudal organs.

Type host and locality

Coelomic cavity of Megascolecidae gen. sp., collected in Initao National Park, Misamis Oriental Province, the Philippines (08°30′N 124°19′E) in November 2013 by S. Spiridonov.

Type material

Holotype male (catalogue number MHNG-INVE-91836) and a paratype female (catalogue number MHNG-INVE-91837) deposited in the Muséum National d'Histoire Naturelle, Geneva, Switzerland. A paratype male (catalogue number 1269) is deposited in the Museum of the Helminthological collections of the Centre of Parasitology at the Severtsov Institute of Ecology and Evolution, Moscow.

Prevalence/intensity

A single host specimen was infected by 11 males and four females.

Molecular characterisation

The 663 bp long LSU rDNA sequence of *I. visayanum* sp. n. differs by 54 nucleotides from the homologous sequence of *H. tonkinense* (JF323056); by 77 nucleotides from *F. suifunense* sp. n., and by >91 nucleotides from all other nematodes with a known homologous sequence.

Phylogeny

The 28S LSU rDNA sequences of *F. suifunense* sp. n. and *I. visayanum* sp. n. (GenBank accession numbers KT160021 and KT160020, respectively) were compared with available sequences of Drilonematoidea from GenBank (Fig. 10). The dataset also included sequences of several nematodes of the infraorders Panagrolaimomorpha and Cephalobomorpha identified through BLAST search in GenBank as being similar to the newly obtained sequences. The addition of two more sequences has not resolved interrelationships within the Drilonematoidea. The representatives of the genus *Dicelis*, which are also parasitic in earthworms, demonstrate an independent position between panagrolaimid-cephalobid nematodes. The monophyly of the nematodes of the families Drilonematidae, Ungellidae and Homungellidae, all parasitic in earthworms, is well supported. The monophyly of the family Ungellidae is also unequivocally supported. Contrary to the current classification, the sequence of *I. visayanum* sp. n. (Drilonematidae) forms a strongly supported clade with *H. tonkinense*, a species of the family Homungellidae.

Discussion

The subfamily Iponematinae is one of six subfamilies constituting the family Drilonematidae Pierantoni, 1916. The subfamily was revised by Ivanova & Neuhaus (2011) and *Globocephalonema*, previously considered as a synonym of *Iponema* by Spiridonov & Ivanova (2005), was restored. Ivanova & Neuhaus (2011) pointed out the presence of forms with an intermediate position between *Filiponema* and *Iponema* (namely *I. laotense* and *F. baviense*). After discussing the morphology and the ambiguity of key characters in both genera, their diagnoses were emended. Employing DNA barcoding represents a possible solution for the accurate discrimination of these two genera. However, as the genera are rare and of exotic provenance, obtaining suitable material for molecular analysis can be difficult. In the current study we were able to obtain DNA for only one out of four *Filiponema*

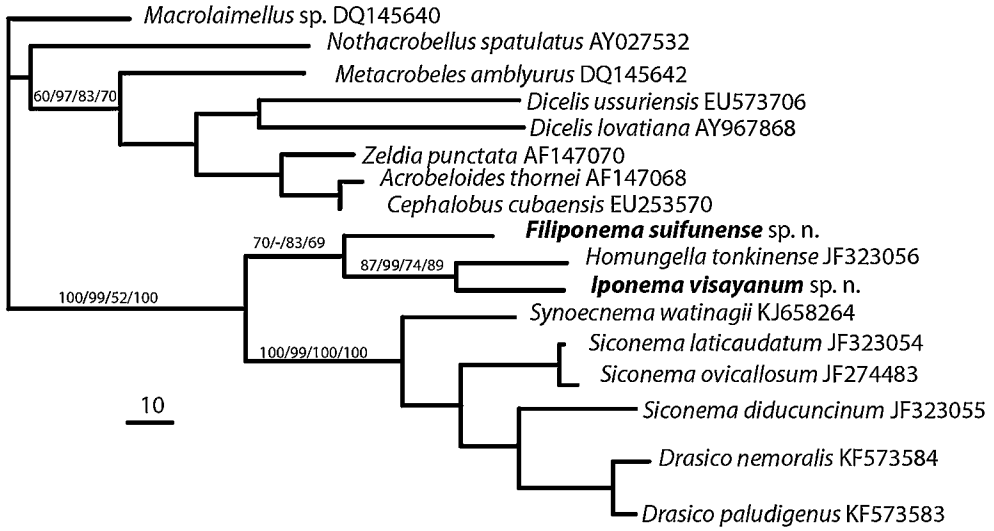

Fig. 10. Phylogenetic relationships of *Iponema visayanum* sp. n. and *Filiponema suifunense* sp. n. as inferred from analysis of the D2-D3 SSU rDNA sequence. The values of bootstrap support/posterior probability are indicated near the nodes in the format MP/NJ/ML/BI. Scale corresponds to the MP analysis and equals to ten nucleotide substitutions. Alignment length 705 bp with 208 parsimony-informative ones.

species described here, namely *F. suifunense* sp. n. One of the key morphological features of *F. suifunense* sp. n., the projecting lateral fields, is exceptional within the genus.

The *Filiponema* sp. described from *L. friendi* is the only species among those described here that fully fits the original diagnosis of the genus by having ten short cephalic setae supporting a raised membrane, circular, conspicuous amphids and symmetrical caudal organs ('suckers' according to Timm), nerve ring posterior to the pharynx and a delicate genital bursa supported by hair-like papillae in the male.

Phylogenetic relationships between all Drilonematoidea for which LSU rDNA sequences were obtained are presented in Figure 10. The molecular data obtained in the current study are the first for Drilonematidae nematodes of non-lumbricid hosts. Previous analyses (Spiridonov & Ivanova, 2005) on 18S SSU rDNA of Drilonematoidea showed that 'lumbricid' drilonematids, such as several species of *Dicelis*, are outside the main Drilonematoidea clade, *i.e.*, Drilonematidae (part.) + Homungellidae + Ungellidae.

The LSU rDNA sequences of *I. visayanum* sp. n. and *F. suifunense* sp. n. can be considered as additional characteristics of the described species, but were also used in the phylogenetic analysis of the wider set of Drilonematoidea. Surprisingly, the sequence of *I. visayanum* sp. n. demonstrates a close affinity to that of *H. tonkinense*. The latter belongs to the Homungellidae Timm, 1966 which is very distinctive in morphology by having a complex cuticular armature at the anterior extremity. The LSU rDNA sequence of *F. suifunense* sp. n. occupies a basal position within Drilonematoidea or clusters with the *Iponema-Homungella* clade with low support values (Fig. 10).

Acknowledgements

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