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Two New Species of Free-Living Marine Nematodes of the Desmodoridae from Mangrove Wetlands of Xiamen Bay, China

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Abstract Mangroves are unique in their biodiversity, but studies on their meiobenthic biodiversity in China are scarce. Despite the importance of mangroves, little work has been done on the classification of nematodes in mangrove wetlands. Fujian Province is the most northern point of China's natural mangrove distribution, and it is also one of the provinces with the earliest constructed mangrove forest. In this paper, two new free-living marine nematode species of Desmodoridae from the Xiamen mangrove wetlands in China are described. *Metachromadora xiamenensis* sp. nov. is characterized by a cylindrical body, and smooth head capsule set off from the rest of the body. The cuticle is finely annulated and thickened at the midbody. Lateral ridges run from the posterior end of the pharynx to the middle of the tail. The amphid foveae is loop shaped and opens at the top with a double contour amphidial, pharynx with bipartite cuticularized internal cavity. There are 18 tubular precloacal supplements and tail with three small protuberances. *Molgolaimus euryformis* sp. nov. is characterized by a relatively short and plump body with finely annulated cuticle, which is particularly obvious in the tail. The head is small and wide with intensive striates. The inner and outer labial sensilla are indistinct with short spicules and ventral apophysis, a gubernaculum with a block-shaped hook, a swollen conical-cylindrical tail and an absence of precloacal supplements.

Key words *Metachromadora xiamenensis* sp. nov.; *Molgolaimus euryformis* sp. nov.; free-living marine nematodes; mangroves

1 Introduction

Mangroves are unique in their biodiversity, but few studies have considered their meiobenthic biodiversity in China (Liu and Huang, 2012). Fujian Province is the northernmost province in China containing natural mangroves and was also one of the earliest provinces to undergo mangrove forest construction in China (Guo *et al*., 2014). The nematode diversity in Fujian Province was previously investigated, and new species were found and described from mangrove wetlands in Xiamen Bay (Li and Guo, 2016; Fu *et al.*, 2018). Only 300 species have been described in detail in China; among them, 90 species are newly found (Shi, 2016). There is still much work to be done on nematode classification, especially in mangrove wetlands.

We discovered two new species of the genera *Metachromadora* and *Molgolaimus* in the mangrove forests of Xiamen Bay. The genus *Metachromadora* includes six subgenera: *Bradulaimus* Stekhoven, 1951; *Chromadoropsis* Filipjev, 1918; *Metachromadora* Filipjev, 1918; *Metachromadoroides* Timm, 1961; *Metonyx* Chitwood, 1936; and *Neonyx* Cobb, 1933. *Metachromadoroides* contains 8 valid species. *Metachromadora* Filipjev, 1918 is characterized by a loop-shaped or round amphidialis, longitudinal lateral ridges (in some individuals), a pharynx bulb internal lining divided into two or three parts, and precloacal supplemental organs (in some individuals) (Gagarin and Tu, 2014). The genus *Molgolaimus* Ditlevsen, 1921, was most recently revised by Fonseca *et al*. (2006) and its latest species was described by Portnova (2009); Shi and Xu (2017) transferred the two reflexed ovaries species of *Microlaimus* to *Molgolaimus*, and *Molgolaimus* now contains 39 valid species.

Two new species, *Metachromadora* (*Metachromadoroides*) *xiamenensis* sp. nov. and *Molgolaimus euryformis* sp. nov., are described in this article.

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2 Materials and Methods

2.1 Study Area

Samples of meiofauna were collected from the mangrove wetland of Tong'an Bay (24.65°–24.70°N, 118.20°–118.30°E) in summer and winter of 2014 in Xiamen Bay, Fujian Province.

2.2 Sampling Method and Sample Processing

Sediment core samples (2.9-cm inner diameter) were obtained, placed in plastic bottles and fixed with 5% formaldehyde. The samples were washed and filtered through 500- and 42-μm mesh sieves, and the material retained on the 42-μm mesh sieve was collected.

The nematodes were extracted using the flotation technique in Ludox-TM with a specific gravity of 1.15–1.18 (De Jonge and Bouwman, 1977). Nematodes were extracted from the samples under a stereoscopic microscope and transferred to a glycerin solution (McIntyre and Warwick, 1984) consisting of 9:1 (V:V) ethanol/glycerol in a cavity block. Then, the ethanol was gradually evaporated, and the nematode specimens were mounted permanently on slides.

Photographs and body measurements were taken with Nikon-50i microscopy equipment. Drawings were made by using a Wacom CTH-690 digital panel. Holotype and paratype specimens were deposited at the College of the Environment and Ecology, Xiamen University, China.

Measurements are in μm. Abbreviations are as follows: a = body length/maximum body diameter, b = body length/esophagus length, c = body length/tail length, abd = anal body diameter, c' = tail length/abd, cbd = corresponding body diameter, V% = distance from vulval opening to the anterior end as percentage of total body length, outer labial setae% = outer labial setae length as a percentage of the cbd, and amphid% = amphid diameter as a percentage of the cbd.

3 Results

3.1 Species Description of *Metachromadora xiamenensis* sp. nov.

Order Desmodorida De Coninck, 1965

Family Desmodoridae Filipjev, 1922

Diagnosis (Modified from Tchesunov, 2014)

Body cylindrical. Cuticle distinctly annulated, without dots, but spines, fringes or longitudinal ornamentations may be present. No specialized ambulatory setae at anterior or posterior body end. Locomotion sinuous, typical for nematodes.

Genus *Metachromadora* Filipjev, 1918

Spiriniinae. Cuticle of head longitudinally striated. Cephalic setae short or papillae. Amphideal fovea strongly cuticularized. Buccal cavity with distinct dorsal tooth. Posterior pharyngeal bulb well-developed with a thick internal cuticular lining partitioned into two or three sections (Tchesunov, 2014).

Subgenus *Metachromadoroides* Timm, 1961

Diagnosis (Modified from Gagarin and Tu, 2014)

Cuticle annulated with longitudinal lateral ridges. Inner labial sensilla papilliform, outer labial and cephalic sensilla elongated papillae or short thick setae. Amphidial foveae on cuticular thickening, circular or loop-shaped with double contours. Posterior pharyngeal bulb well developed, with strongly cuticularized internal lining often divided into two or three parts. Precloacal supplemental

Table 1 Morphometrics of *Metachromadora xiamenensis* sp. nov. (in μm)

Character	Holotype (male)	Paratypes, $n=4$ (males)		Paratypes, $n=3$ (females)	
		Min – max	Mean	Min – max	Mean
Body length	821.9	741.0 – 887.7	812.7	726.3 – 925.8	824.4
a	11.8	11.0 – 14.3	12.8	9.1 – 12.8	10.2
b	4.6	4.4 – 5.2	4.9	4.1 – 5.2	4.7
c	12.0	10.1 – 12.2	11.4	11.3 – 13.8	12.1
c'	1.7	1.5 – 1.7	1.6	1.4 – 1.6	1.5
V%	–	–	–	49.1 – 53.3	51.3
Head diameter	22.7	22.3 – 25.3	23.4	22.1 – 23.1	22.6
Stoma length	30.1	28.7 – 30.8	29.3	24.6 – 28.2	26.5
Amphid width	14.7	14.8 – 15.3	15.0	11.2 – 13.6	12.1
Amphid length	8.2	6.7 – 8.9	7.9	8.5 – 9.9	9.1
Body diameter at amphid level	21.5	22.5 – 25.9	23.9	22.1 – 25.7	23.7
Amphid diameter as percentage of the corresponding body diameter	68%	59% – 66%	63%	48% – 56%	51%
Amphid length divided by amphid width	0.6	0.5 – 0.6	0.5	0.6 – 0.8	0.8
Pharyngeal length	177.5	158.9 – 172.3	167.3	166.6 – 184.0	176.9
Pharyngeal bulb length	57.8	54.1 – 58.5	56.5	57.1 – 62.3	60.2
Pharyngeal bulb length as percentage of pharyngeal length	33%	33% – 34%	34%	32% – 35%	34%
Maximum body diameter	69.8	58.1 – 71.4	63.7	78.6 – 94.2	81.9
Anal body diameter	40.0	43.6 – 46.0	45.2	41.6 – 47.4	44.4
Spicule length as arc	54.0	46.0 – 55.6	51.4	–	–
Gubernaculum length	31.3	22.7 – 29.9	25.2	–	–
Tail length	68.4	67.3 – 73.4	71.3	66.1-77.4	68.5
Number of precloacal supplements	18	18	18	0	0

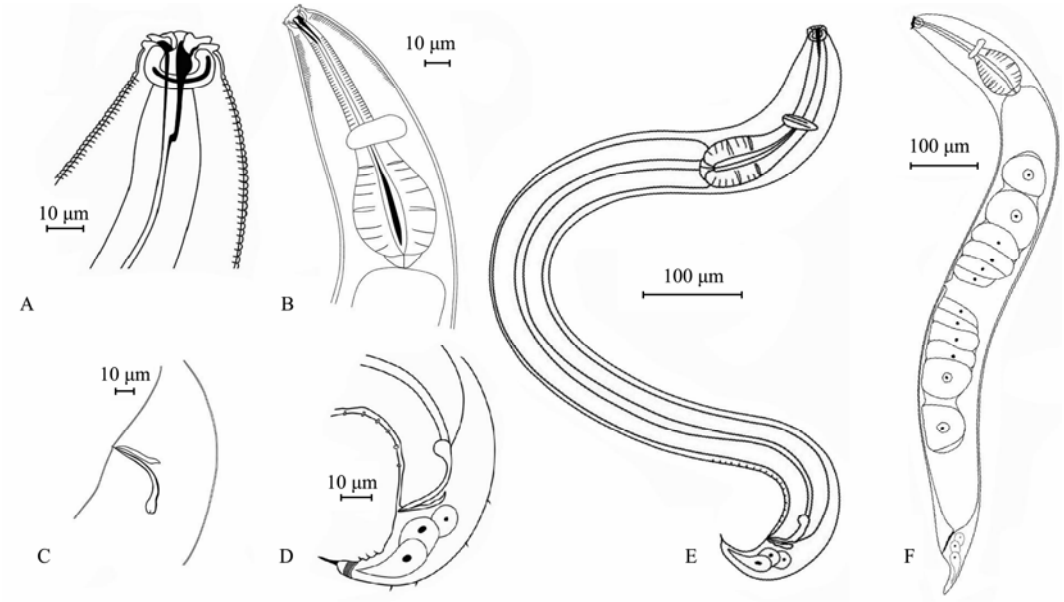

Fig.1 *Metachromadora xiamenensis* sp. nov. A, male head; B, lateral view of female anterior end; C, male spicules and gubernaculum; D, male tail; E, entire view of male body; F, entire view of female body.

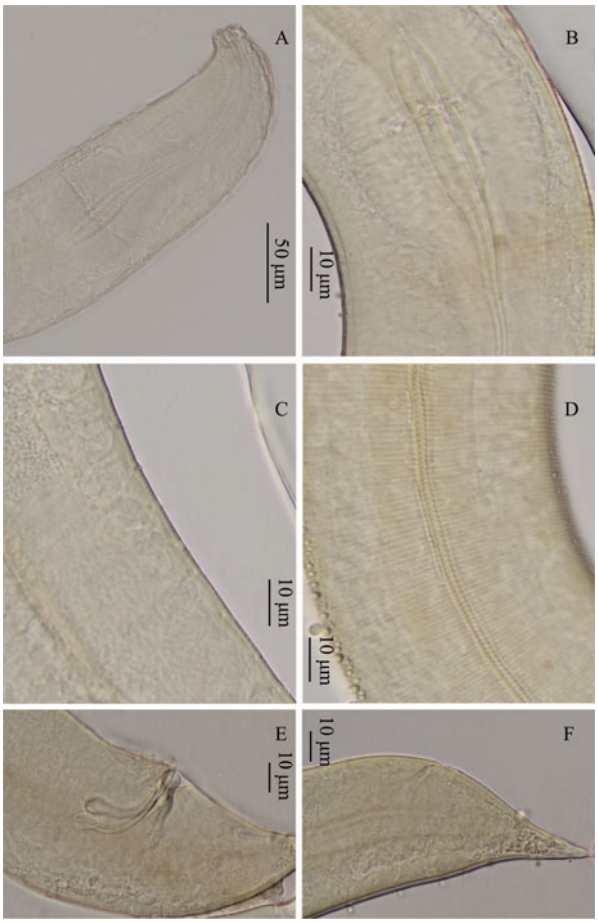

Fig.2 *Metachromadora xiamenensis* sp. nov. A, head of male, showing buccal cavity and labial sense organ; B, finely annulated cuticle of male, with an esophageal bulb; C, eighteen precloacal supplements of male; D, lateral punctuation of female; E, male spicules and gubernaculum; F, tail of female.

organs in the shape of short, faintly cuticularized tubules present or absent.

Type species

Metachromadora (Metachromadoroides) complexa Timm, 1961

Metachromadora xiamenensis sp. nov. (Figs.1, 2, Table 1)

Type material

Male holotype, slide 201402E311. Four male paratypes, slides 201402E311, 201402E312, 201402E211, and 201 402D102. Five female paratypes, slides 201402E306, 201402E308, 201402E312, 201402E314, and 201402E 314. All the specimens were collected from the muddy surface sediment layer (0–10 cm) in an artificial mangrove wetland located in Tong'an Bay in Xiamen. The mangrove species were mainly *Kandelia obovata*, also includes *Aegiceras corniculatum*, *Avicennia marina* and *Sonneratia apetala*.

Etymology

This species is named after the city Xiamen, where it was found.

Measurements

The morphometric characteristics of the holotype and paratypes are given in Table 1.

Description

Male. Body cylindrical, smooth head capsule set off from the rest of the body. Cuticle finely annulated, with a thickness of 1 μm at midbody. Lateral ridges beginning at the posterior end of the pharynx and extending to the middle of the tail. Somatic setae short and sparse, irregularly spaced. Labial sensilla indistinct, with four cephalic sensilla in the shape of large elongated papillae that are 2.5–2.7 μm long (16%–18% of the corresponding lip areawidth). Amphids 14.8–15.3 μm (59%–66% cbd) and loop-shaped, doubly contoured with an open top. Amphids situated in the labial region, extending to the end of the head capsule and surrounded by a widened cuticular ring. Stoma 28.7–30.8 μm deep, with a strong dorsal tooth positioned in the anterior part. Pharynx cylindrical, 158.9–

172.3 μm long, posteriorly enlarged with a well-developed bipartite basal bulb, the internal lining of which is strongly cuticularized. Bulb length 33%−34% of the total pharynx length. Nerve ring indistinct, with small cardia.

Male with one anterior testis, positioned to the right of the intestine. Eighteen tubular precloacal supplements present. Paired curved spicules 46.0−55.6 μm long (1.0−1.4 abd), distinctly cephalated. Gubernaculum with middle piece and double dorsal apophyses. Tail 67.3−73.4 μm long, 1.5−1.7 anal diameter, with a finger-like tip. Three small protuberances with short setae positioned on the ventral side of the tail. Caudal glands present, but poorly visible.

Females. Females similar to males. Reproductive system didelphic, amphidelphic with reflexed ovaries. Anterior ovary positioned to the left of the intestine, posterior ovary to the right of the intestine. Vulva with thick walls, situated at the median body.

Differential diagnosis

M. (*M.*) *xiamenensis* sp. nov. is characterized by loop-shaped amphidial foveae with an open top and double contours, a pharynx with a bipartite cuticularized internal cavity, arcuate spicules, proximal end strongly cephalated, 18 tubular precloacal supplements, and a short, conical tail with three small protuberances.

This new species is morphologically similar to *M.* (*M.*) *orientalis* (Gagarin and Tu, 2014) in the shape of the amphid and the head sensilla pattern, and has a similarly armed tail in males. *Metachromadora xiamenensis* sp.

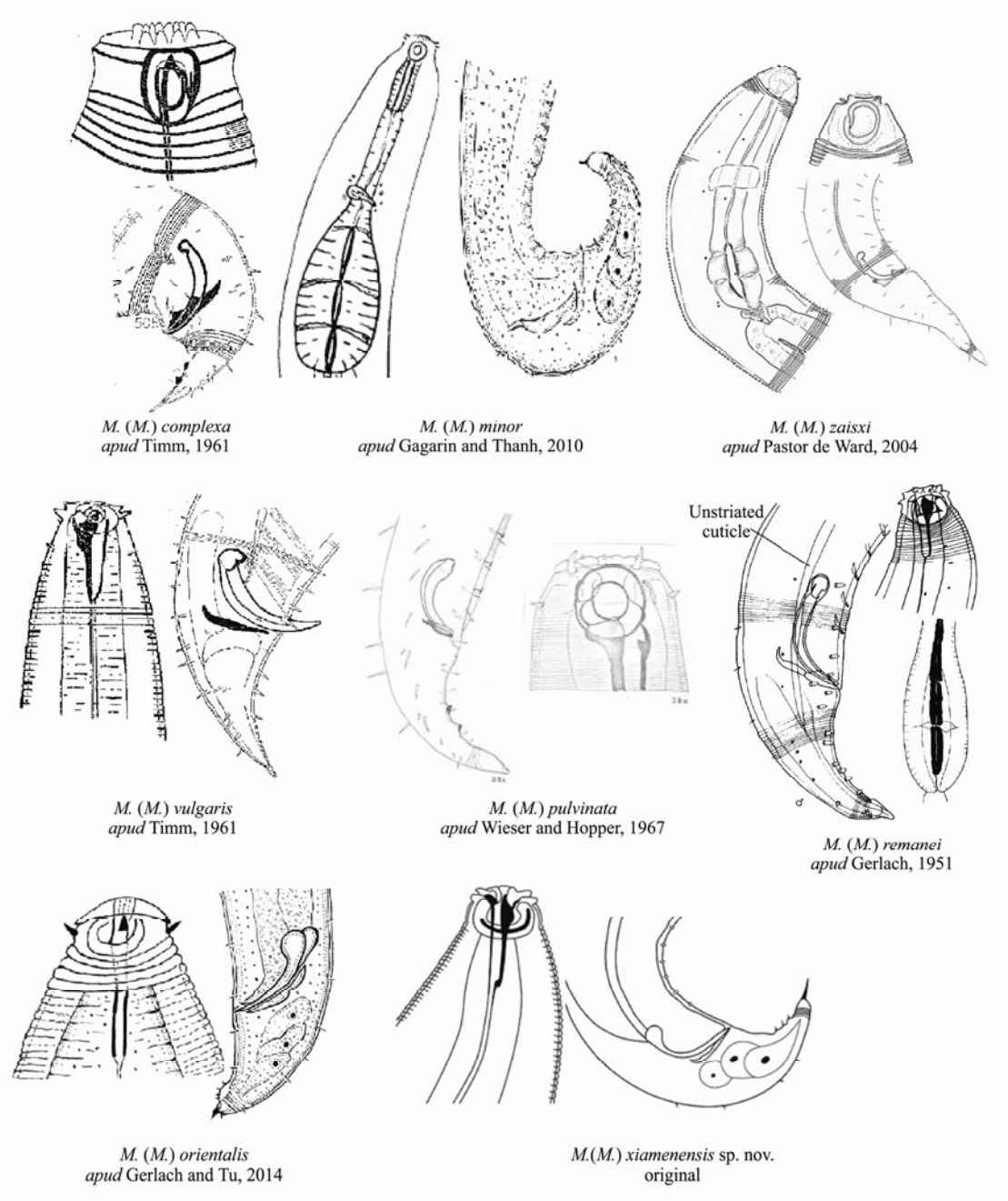

Fig.3 Pictorial key to the species of *Metachromadora* (*Metachromadoroides*).

nov. is also similar to *M. complexa* in the length, a, b and c values, but A% (59–66% in *M.* (*M.*) *xiamenensis* sp. nov. *vs.* 41.6% in *M. complex*), with longer spicules (74 μm in *M.* (*M.*) *xiamenensis* sp. nov. *vs.* 46–56 μm in *M. complexa*), and different numbers of the internal lining of the pharyngeal bulb (2 in *M.* (*M.*) *xiamenensis* sp. nov. *vs*. 3 in *M. complexa*). However, the new species can be distinguished from the other species by the presence of 18 precloacal supplements, a longer and thinner tail (c = 10.1–12.2 and c' =1.5–1.7 in *M.* (*M.*) *xiamenensis* sp. nov. *vs*. c =15.3–19.9 and c' =1.0–1.9 in *M.* (*M.*) *orientalis*) and wider lip area.

The composite differentiating characters for all male *Metachromadora* (*Metachromadoroides*) species are provided to aid in identification (Table 2).

Table 2 Differential characters of males among *Metachromadora* species

Species	L (μm)	a	b	c	c'	A (%)	Spicule length, μm	Internal lining of pharyngeal bulb	Number of supplements	Reference
M. (*M.*) *vulgaris*	1100-1200	14.1-20.0	4.4-4.9	12.1-14.5	1.8-2.8	70	72	bipartite	0	Timm, 1961
M. (*M.*) *zaisxi*	890-1160	10.5-19.3	4.7-7.2	6.8-10.1	1.7-2.8	63	40-52	bipartite	0	Pastor and Ward, 2004
M. (*M.*) *remanei*	1100-1300	17-23	4.9	14	1.9-2.2	70-80	49-55	bipartite	5	Gerlach, 1951
M. (*M.*) *complexa*	758-940	8.4-11.3	3.4-4.4	7.3-12.1	1.3	41.6	74	tripartite	17	Timm, 1961
M. (*M.*) *pulvinata*	1720	–	5.4	15.6	2.0	55	55	bipartite	23	Wieser and Hopper, 1967
M. (*M.*) *orientalis*	859-1133	13-15	4.8-5.3	15.3-19.2	1.0-1.3	62	59-63	bipartite	0	Gagarin and Tu, 2014
M. (*M.*) *minor*	594-641	13-17	4.2-4.7	13.5-14.9	1.4-1.5	90	35-36	bipartite	12, 15	Gagarin and Thanh, 2010
M. (*M.*) *xiamenensis* sp. nov.	741-888	11.0-14.3	4.4-5.2	10.1-12.2	1.5-1.7	59-66	46-56	bipartite	18	this text

Notes: A = amphid diameter as a percentage of the corresponding body diameter. Spicule/cloaca is the ratio of spicule length to body diameter at the cloaca level.

3.2 Species Description of *Molgolaimus euryformis* sp. nov.

Genus *Molgolaimus* Ditlevsen, 1921

Diagnosis (Modified from Fonseca *et al*., 2006)

Cuticle finely annulated. Amphid situated behind the narrowing part of the head. Inner labial and outer labial sensilla small, hard to observe. Cephalic setae close to the cephalic constriction. Buccal cavity small and narrow with small teeth. Esophagus cylindrical with a pronounced posterior spherical bulb, heavily sclerotized at the bulb. Spicules of different lengths and shapes from short and bent to long and straight. Gubernaculum with or without apophysis. Precloacal supplements often present. Tail of varying shape and length, from short and conical to elongate and slender, cylindrical posteriorly.

Type species

Molgolaimus tenuispiculum Ditlevsen, 1921

Table 3 Morphometrics of *Molgolaimus euryformis* sp. nov. (in μm)

Character	Holotype (Male)	Paratypes		
		Males (n = 6)		Female (n = 1)
		Min – Max	Mean	
Body length	509.2	485.6 – 620.7	527.2	387.6
a	11.4	11.3 – 13.7	12.6	13.2
b	4.7	4.3 – 5.3	4.7	4.3
c	6.5	6.1 – 6.9	6.6	5.6
c'	2.4	2.2 – 2.8	2.5	3.5
Head diameter	13.2	12.4 – 14.5	13.5	10.8
Head setae	3.5	3.3 – 4.0	3.5	3.5
Length of buccal cavity	14.3	13.6 – 16.4	14.6	12.3
Amphids from the anterior end	9.5	10.5 – 13.1	11.6	12.9
Amphid diameter	5.1	5.3 – 6.4	6.0	4.8
Body diameter at amphid level	31%	28% – 38%	34%	33%
Pharyngeal length	107.8	104.5 – 121.6	112.2	89.8
Pharyngeal bulb diameter	31.6	23.7 – 35.1	31.1	18.4
Body diameter at pharyngeal bulb level	38.2	29.9 – 42.1	38.3	24.2
Pharyngeal bulb diameter as percentage of the corresponding body diameter	83%	79% – 85%	81%	76%
Maximum body diameter	44.6	36.3 – 45.2	41.6	29.3
Spicule length as arc	22.7	17.8 – 24.3	20.9	–
Anal body diameter	33.3	26.3 – 41.2	32.9	19.9
Spicule length as arc divided by anal body diameter	0.7	0.6 – 0.8	0.6	–
Gubernaculum length	29.3	24.4 – 29.1	27.7	–
Tail length	78.9	72.4 – 92.4	81.0	69.2
V%	–	–	–	52

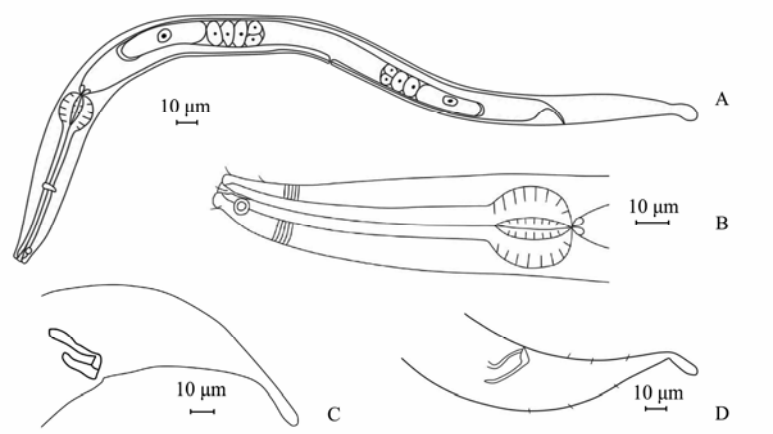

Fig.4 *Molgolaimus euryformis* sp. nov. A, entire view of female body; B, male anterior region; C, structure of spicules and gubernaculum; D, lateral view of male tail.

Molgolaimus euryformis sp. nov.

Type material

Male holotype, slide 201408C203. Five male paratypes, slides 201408C203, 201408C208, 201408B211, 201402 F210, and 201408C207. One female paratype, slide 2014 02C301.

Etymology

This species is named for its lower 'a value' compared to that of other species in the genus.

Measurements

The morphometric characteristics of the holotype and paratypes are given in Table 3.

Description

Male. Body relatively short, cylindrical. Cuticle finely annulated, particularly obvious on the tail. Head small and wide with intensive striation, separated from body by small narrowing before head setae. Inner and outer labial sensilla indistinct. Four cephalic setae approximately 3–4 μm long (30% of head diameter). Amphid fovea circular, 5–6 μm in diameter (28%–38% of cbd), located 11–13 μm from the anterior end. Buccal cavity small with teeth. Esophagus forms a pronounced spherical bulb posteriorly with a 31-μm diameter. Cardia is small and extended, and the glandular body lies posterior to the cardia. Excretory pore not observed. Reproductive system monarchic, outstretched testis situated to the left of the intestine. Spicules short with ventral apophysis, 20.9 μm in length (0.6–0.8 abd). Gubernaculum parallel to spicule, with a block-shaped hook on the terminus. Tail conical along 3/4 of its length, cylindrical posteriorly with a swollen tip, 2.2–2.8 anal diameter.

Female. Similar to male, reproductive system didelphic with reflexed ovaries. The ovaries situated to the left of the intestine. Anterior ovary slightly longer than the posterior ovary.

Differential diagnosis

M. euryformis sp. nov. is characterized by its short and relatively plump body (L = 485.6–620.7 μm, a = 11.3–13.7), relatively short head setae, ventral apophysis (17.8–24.3 μm), gubernaculum with a block-shaped hook, conical-cylindrical shape with a swollen tail tip and absence of precloacal supplements.

Table 4 Comparison of *Molgolaimus euryformis* sp. nov. with allied species (male)

Species	*M. citrus*	*M. lazonus*	*M. turgofrons*	*M. cuanensis*	*M. parallgeni*	*M. drakus*	*M. euryformis* sp. nov.
Body length (μm)	455	707	746	1210	1373	495–575	485.6–620.7
a	27	28.2	29	46.5	45.6	35.5–42.7	11.3–13.7
b	5.6	7.2	7.3	11.5	13	6.1–7.0	4.3–5.3
c	7	9.1	8.5	14.2	14.6	7.0–7.9	6.1–6.9
c'	4.5	3.5	2.9	3.5–5.0	3.4	5.3–6.2	2.2–2.8
Spicule length as arc (μm)	15	21	30	25	28	19–20	17.8–24.3
Anal body diameter (μm)	14.5	22.1	30	25	28	–	36.3–45.2
Spicule length as arc divided by anal body diameter	1.03	0.95	1.0	1.0	1.0	1.4–1.6	0.6–0.8
Number of precloacal supplements	2	0	0	2	2	2	0
Length of head setae (μm)	4	2	2.5	4.5–5	5.7	2	3.3–4.0
Head setae length as percentage of head diameter	–	30	–	50	60	–	30
Body length divided by spicule length as arc	30.3	33.7	24.9	48.4	49.0	24.8	21.9–27.7
Reference	Gerlach, 1959	Vitiello and Boucher, 1970	Lorenzen, 1972	Platt, 1973	Vitiello and Boucher, 1973	Fonseca *et al.*, 2006	this text

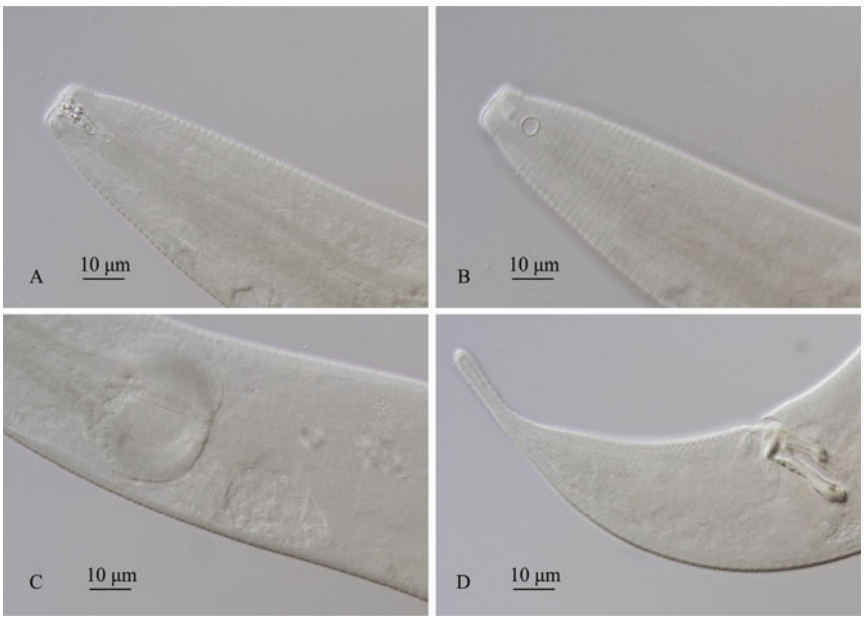

Fig.5 *Molgolaimus euryformis* sp. nov. A, anterior body of male, showing the buccal cavity and cephalic setae; B, anterior body of male with amphids; C, spherical esophageal bulb of male; D, spicules, gubernaculum and tail with swollen tip.

Based on the morphological parameters, the new species belongs to group 1a in the identification key (Fonseca *et al.*, 2006), members of which are characterized by short spicules (< 35 μm) and a spicule to abd ratio less than 1.

Among the species in the group (*M. citrus* (Gerlach, 1959), *M. lazonus* (Vitiello, 1971), *M. turgofrons* (Lorenzen, 1972), *M. cuanensis* (Platt, 1973) and *M. parallgeni* (Vitiello, 1973)), *M. euryformis* sp. nov. has the smallest value and spicule/abd ratio. This species is most closely related to *M. lazonus* and *M. turgofrons* but differs from them by its conical-cylindrical body with a swollen tail tip and short spicules with ventral apophysis.

The new species resembles *M. drakus* (Fonseca *et al.*, 2006) in the ratio of body length to spicule length. However, *M. drakus* differs from *M. euryformis* sp. nov. by its slim body, its spicule form (short and without ventral apophysis) and the presence of only one precloacal supplement.

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