

## Two new species of Enoplolaiminae (Enoplida: Thoracostomopsidae) from Río Negro and Chubut, Argentina

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### Abstract

Two new species of free-living marine nematodes of the subfamily Enoplolaiminae are described from Río Negro and Chubut Provinces, Argentina. *Enoplolaimus variispiculum* sp. n. is characterized by having asymmetric spicules, different in size and distal tips. It is the only species with this characteristic in the whole genus. *Mesacanthoides flagellatum* sp. n. is characterized by mandibles texture, weakly cuticularized and mottled and mandibular plates with pronounced “v” shaped incision posterior to the anterior bar. Cephalic capsule wider than long with setae on posterior end, males with long spicules, complex gubernaculum and flagellate tail.

**Key words:** *Enoplolaimus variispiculum* sp. n., *Mesacanthoides flagellatum* sp. n., systematics

### Introduction

In the period 2006–2012, two survey projects (PNUD/GEF 02/018; PID/CONICET N°11220090100440) were carried out to examine biodiversity of meiofauna in several beaches along the Argentinean Atlantic coast. During preliminary examination of the material from this survey, nematodes belonging to the genus *Enoplolaimus* de Man, 1893 and another interesting nematode were noted in winter and summer samples. On closer investigations it appeared to belong to the genus *Mesacanthoides* Wieser, 1953.

The genera *Enoplolaimus* and *Mesacanthoides* are a predatory group of free-living marine nematode taxa belonging to the subfamily Enoplolaiminae, which includes seventeen (17) valid genera (Smol & Coomans, 2006; Smol *et al.*, 2014).

The genus *Enoplolaimus* has been reviewed by Fadeeva & Zograf (2010). They had given an identification and a pictorial key for 35 valid species within the genus. Identifying five groups of species, using as diagnostic characters presence or absence of precloacal supplement, presence of apophysis in gubernaculum and tail length. They did not include those species which were described based on juveniles (*E. conicaudatus* Allgén, 1929, *E. macrocharius* Allgén, 1929 and *E. similis* Allgén, 1929), and based on females (*E. niger* Allgén, 1959, *E. opacus* Allgén, 1959, *E. vilosus* Gerlach, 1953). We agree with this. But they did not include other four species considered as valid in Nemys (*E. asymmetricus* Keppner, 1987, *E. halophilus* Ditlevsen, 1928, *E. longigubernaculum* Shimada *et al.*, 2009 and *E. strandi* Allgén, 1940). Also *E. longigubernaculum* and *E. asymmetricus* were considered valid species in Smol *et al.* (2014). Here, we discuss their validity and add comments and include some of them in Fadeeva & Zograf groups.

*E. asymmetricus*, where one male and one female were described, has three lips with striations at the base, asymmetric mandibles, straight spicules, no precloacal supplement, no gubernaculum, and a tail 3.7 anal diameters long. So, following Fadeeva & Zograf (2010), it belongs to group 1.

*E. halophilus*, where two males and one female were described, has precloacal organ, arched spicules, gubernaculum without apophysis and tail less than 4 anal body diameters in length. So, following Fadeeva & Zograf (2010), it belongs to group 6. The description of males seem to be correct, considering the date of the paper, but the description of female gonads was likely based on another genus or there was a drawing mistake, because it looks different to what is known in *Enoplolaimus*.

*E. longigubernaculum* was contemporary to Fadeeva & Zograf (2010) (it was published one month before). It has precloacal supplement, arched spicules, a long gubernaculum without apophysis, and a tail shorter than 4 anal body diameters in length, so it belongs to the group 6 (Fadeeva & Zograf, 2010).

To study *E. strandi*, *E. filiformis* (Allgén, 1935) Wieser, 1953 and *E. filiformis* (Allgén, 1935) Allgén, 1959, we had access to original specimens, through Swedish Museum of Natural History (Stockholm, Sweden) loan facility. We obtained very good, detailed photos from old specimens, sent to us by senior curator Dr. O. Holovachov.

*E. strandi* original male specimen (Slide number SMNH Typ-3587) presents three triangular mandibles solid anteriorly; no teeth observed; labial and cephalic setae seem to be anterior to cephalic capsule; cylindrical straight spicules, with acute cloacal end; without gubernaculum, with one or two (?) plate-shaped precloacal supplements. *E. strandi* was considered as doubtful species by Wieser (1953). Following Smol & Coomans (2006) key to the genera of Enoplolaiminae, morphology of this particular species does not match the diagnosis of *Enoplolaimus*, neither of Enoplolaiminae. So probably it is a new genus. We consider *E. strandi* as **genus** and **species inquerendae**.

*E. filiformis* (Allgén, 1935) Wieser, 1953 was described based on one female (Allgén, 1935) from Oresund, North Hemisphere. After Wieser (1953) established it as doubtful, Allgén (1959) redescribed it from one male, one female and one juvenile from specimens from South Georgia, Southern Hemisphere. It was included in a group 6 by Fadeeva & Zograf (2010). *E. filiformis* (Allgén, 1935) Wieser, 1953 original female specimen (Slide number SMNH Typ-8815) presents striated lips, solid mandibles in anterior part, with two lateral claws, with very conspicuous rounded bases on each side of mandible, labial and cephalic setae on posterior end of cephalic capsule and short tail. Following the key to Enoplolaiminae by Smol & Coomans (2006), it is not a member of the genus *Enoplolaimus*. It could be a species from the genus *Enoploides* or a new genus entirely. So, we consider it as **genus** and **species inquerendae**.

*E. filiformis* male specimen described by Allgén (1959) from South Georgia is a different genus and species than the female holotype of *E. filiformis* Allgén, 1935. It presents labial and cephalic setae on the anterior end of cephalic capsule, long mandibles (1 head diameter) with a posterior part hollow and an anterior part solid, three teeth observed. The spicules are arched, cephalated, with a tubular gubernaculum. The pre-cloacal organs seem to be plate-shaped. Following Allgén's drawings the first one from cloaca has two setae. These setae are not visible in the photographs, being obscured by the diffraction on the dehydrated specimen border. Following the key to Enoplolaiminae by Smol & Coomans (2006), this species does not belong to *Enoplolaimus*, neither to *Paramesacanthion* or *Mesacanthion* because they all have tubular pre-cloacal organs, so it could be a new genus. So, we consider *E. filiformis* Allgén, 1959 as **genus** and **species inquerendae**.

With the inclusion of *E. asymmetricus*, *E. halophilus*, *E. longigubernaculum* and the new species the genus *Enoplolaimus* presents now 38 valid species:

- E. abnormis* Kreis, 1928
- E. acanthospiculum* Allgén, 1959
- E. arcospiculum* Allgén, 1959
- E. asymmetricus* Keppner, 1987
- E. attenuatus* Brunetti, 1949
- E. balgensis* Skwarra, 1921
- E. conicollis* Gerlach, 1952
- E. connexus* Wieser, 1953
- E. denticulatus* Warwick, 1970
- E. derjugini* Filipjev, 1929
- E. distortus* Gerlach, 1957
- E. enatus* Hopper, 1962
- E. falklandiae* Allgén, 1959
- E. glabrus* Brunetti, 1949
- E. halophilus* Ditlevsen, 1928
- E. karachiensis* Maqbool, Nasira & Turpeenniemi, 1999
- E. kuri* Gerlach, 1959
- E. lenunculus* Wieser, 1959
- E. litoralis* Schulz, 1936

*E. longicaudatus* (Southern, 1914) Filipjev, 1921  
*E. longigubernaculum* Shimada, Kajihara & Mawatari, 2009  
*E. medius* Pavlijuk, 1984  
*E. mus* Inglis, 1964  
*E. notopropinquus* Allgén, 1959  
*E. paralitoralis* Wieser, 1959  
*E. parapropinquus* Allgén, 1951  
*E. pararegius* Keppner, 1987  
*E. patrokli* Fadeeva & Zograf, 2010  
*E. pectinatus* Fadeeva & Zograf, 2010  
*E. propinquus* de Man, 1922  
*E. psammae* Gerlach, 1952  
*E. punctatus* Hopper, 1961  
*E. regius* Hopper, 1962  
*E. robustus* Gerlach, 1954  
*E. subterraneus* Gerlach, 1953  
*E. variispiculum* sp. n.  
*E. vulgaris* de Man, 1893  
*E. zosterae* Schulz, 1932

We add a table with the differential characters of species belonging to group 6 following Fadeeva & Zograf (2010), which includes the new species described in the present paper.

The genus *Mesacanthoides* was reviewed by Wieser (1953), Wieser & Hopper (1967) and Wieser (1959). We add a table with comparison of the 8 valid species (Nemys, Bezerra *et al.*, 2021) plus the new species described herein.

Following the suggestions of Wieser & Hopper (1967) and Jeong *et al.* (2020), we describe the two new species on the base of male characters. Also, we include detailed description of head.

## Materials and methods

**Description of study sites.** Samples were collected from two littoral coastal sediments located in Río Negro and Chubut Provinces, Argentine.

**Site A.** *M. flagellatum* sp. n.: Samples were collected from San Matías Gulf, Argentine (Figure 1A), which is the northernmost of the Patagonian gulfs. The gulf has a width of about 100 km at the entrance and 150 km at the end. San Antonio Bay river estuary is located in the northwest part of San Matías Gulf. The specimens were collected in front of the city of San Antonio Oeste “Balneario” site (40°43' 66" S; 64°56'52" W), at low tide, Río Negro Province, Argentine.

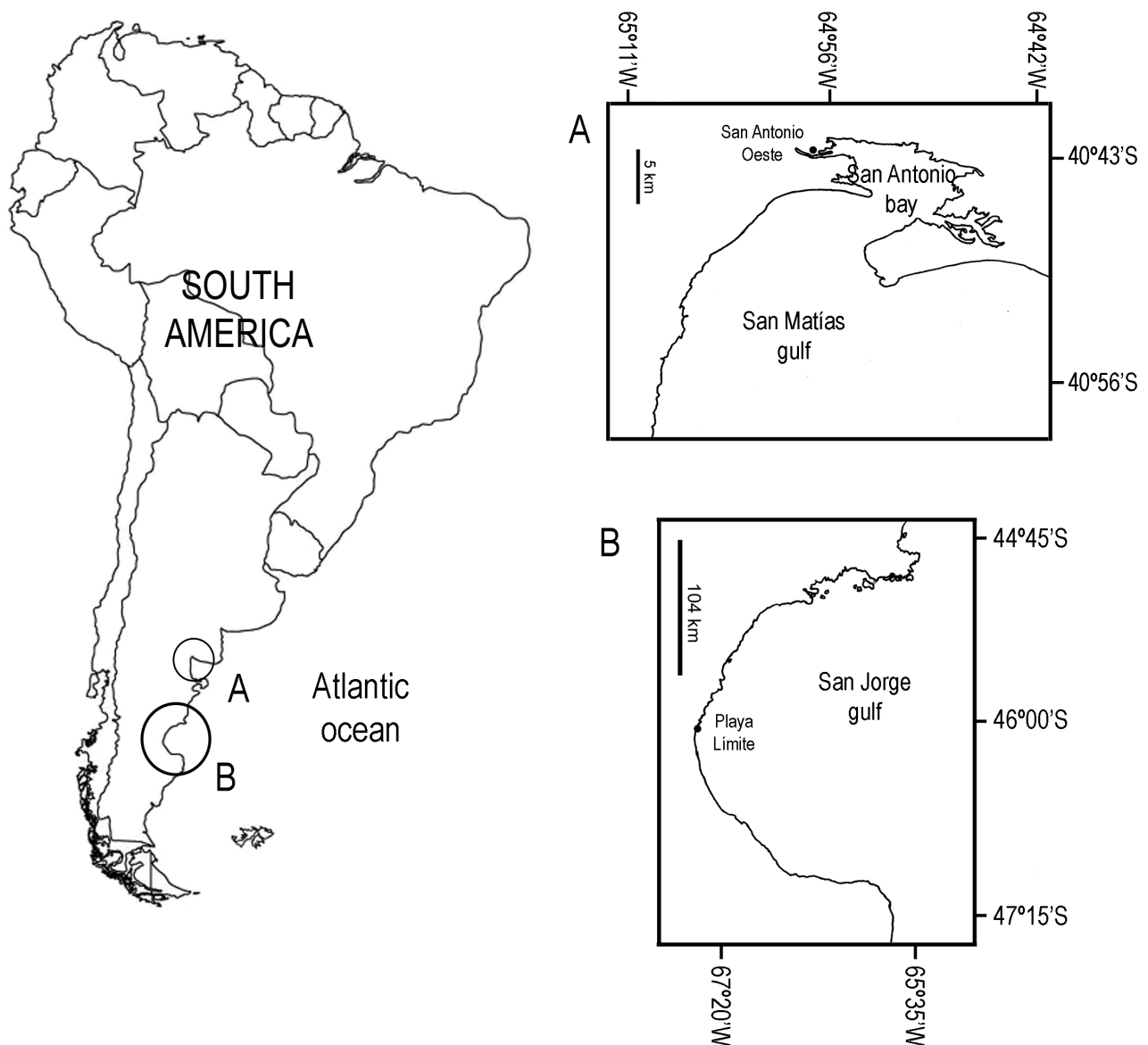
**Site B.** *E. variispiculum* sp. n.: Samples were collected from “El Límite” beach, San Jorge Gulf (45°59'38"S and 67°35'31"W), Figure 1B. The “El Límite” beach is a wide coastal recreational area 2.5 km long. It is a dissipative beach that lies on the border between the provinces of Chubut and Santa Cruz. The specimens described in this paper came from the north of the beach in the province of Chubut, Argentine. They were collected in mean tidal littoral sand.

**Sample collection and treatment.** The samples were collected with a cylindrical Plexiglas corer, 10 cm high and 2.9 cm in diameter. They were preserved in 5% formaldehyde in filtered seawater, sieved through both 500 µm and 50 µm mesh sieves. The nematodes present on the 50 µm mesh sieve were separated by the LUDOX-TM method (de Jonge & Bouwman, 1977). Preserved specimens were transferred to pure glycerine through a solution of ethanol-water-glycerine in 2:2:1 proportion respectively, and left for at least one week in a desiccator. After that, permanent mounts were prepared on glass slides and sealed with CADANAX resin (Somerfield *et al.*, 2005).

**Specimen analysis.** Morphometric data and photos were obtained by a digital camera NIKON D80/D90 with adapted software or from drawings made with a Camera Lucida attached to a light microscope (Zeiss fotomicro-

scope or Olympus BX51) equipped with DIC optics. All curved structures were measured along the arc or median line. Sediment analyses were carried out by dry sieving and classified according to the Wentworth scale. The de Man's ratio a, b and c used in this paper were calculated as standard. Literature was obtained from Nemys (Bezerra *et al.*, 2021). Type specimens are deposited in the Centro Nacional Patagónico, Chubut, Argentina, belonging to the National Biological Information System.

**Abbreviations used in the text:** cbd = cloacal body diameter; H/W=height/width; vs=versus; Q2 (50%) = corresponds to the 50% point of the cumulative scale ( $\approx$  50); FF (%) = percentage of fine fraction; SO index = sorting coefficient; SK index = skewness coefficient; VCS = very coarse sand.



**FIGURE 1.** Map showing the study sites. Map of sampling areas: A. “San Antonio Oeste river estuary” beach, Río Negro Province; B. “El Límite” beach, Chubut Province.

## Systematics

### Family Thoracostomopsidae Filipjev, 1927

### Subfamily Enoplolaiminae De Coninck, 1965

## Genus *Enoplolaimus* de Man, 1893

**Diagnosis.** From Fadeeva & Zograf, 2010. Enoplolaiminae. Cuticle usually smooth or finely striated. Cephalic region blunt, anteriorly rounded, with distinct external groove at base of cephalic capsule. Inner labial setae long, inserted at base of lip lobes; outer labial and cephalic setae situated at posterior edge of cephalic capsule. Three thin triangular lip-lobes with or without semi-lunar striation. Three broad, arched, mandibular plates, not solid medially as seen in *Enoploides* Saweljev, 1912 or *Epacanthion* Wieser, 1953. Anterior curved rim with short pointed tips projecting into stoma. One dorsal and two ventrosublateral equal or unequal onchia. Cephalic capsule well developed, broad, posteriorly divided into six lobes. Single ventral pre-cloacal sensory supplementary organ present or absent. Spicules not very elongated, straight or arcuate, with or without apophyses.

### *Enoplolaimus variispiculum* sp. n.

(Figure 1B, 2, 3; Table 1, 2)

**Measurements.** See Table 1.

**Description. Males:** Body cylindrical, slender, anteriorly narrow, weakly tapering posteriorly. Cuticle finely striated. Head rectangular and set off, with three lips (one dorsal and two ventrosublateral), without striations and separated by clefts, 8  $\mu$ m high. Each lip bearing two inner labial sensilla, 0.2 corresponding body diameters in length. Cephalic glandular organ was observed on ventrosublateral lips. It is straight in males and arched in females. Six outer labial sensilla, 2.1 corresponding body diameters in length, inserted at level of posterior margin of cephalic capsule. A second crown of four cephalic setae 1.8 corresponding body diameters in length. A third crown of six subcephalic setae 1.4 corresponding body diameters in length. Three crowns of cervical setae in dorsosublateral, lateral and ventrosublateral position. First crown with 6 setae, 0.6 corresponding body diameters in length, second crown with 6 setae, 0.5 corresponding body diameters in length, third crown with 4 setae, 0.5 corresponding body diameters in length. Body with dispersed somatic setae, 8–10  $\mu$ m long.

Cephalic capsule well developed. Posterior margin of cephalic capsule with deep incisions accommodating the first crown of outer labial setae, cephalic setae and subcephalic setae. Three broad, arched, mandibular plates with pointed tips projecting into stoma. Mandibular plates slightly curved and thin and with two small projections located laterally, at level of mandibular ring, 1  $\mu$ m long. Three onchia, one dorsal and two ventrosublateral, equal in size, at the base of each mandible. Amphids and excretory pore not observed. Metanemes not observed. Pharynx cylindrical, slightly englobed anteriorly and with outer wall crenated posteriorly.

Reproductive system diorchic, anterior testis outstretched, posterior one reflexed. Spicules paired, asymmetrical, longest spicule (left) arcuate, cephalated (1.7 cloacal body diameters in length), with pointed distal end. The other spicule (right) short and straight, slightly cephalated with sharp distal end, 0.8 cloacal body diameters in length. Gubernaculum present, plate-like, triangular, without any structures, 0.5 cloacal body diameters in length. One preanal supplement present. One setae observed before cloaca. Tail 4.2 cloacal body diameter long. Three caudal glands and spinneret were observed.

**Females:** Similar to male in general body shape, but usually larger. It differs in head shape and outer labial and cervical setae number. Six labial sensilla 0.2 corresponding body diameters in length, six long outer labial sensilla 2.1 corresponding body diameters in length and four short cephalic setae 1 corresponding body diameter in length, inserted at posterior margin of cephalic capsule. One crown of four cervical setae 0.4 corresponding body diameters long. Lateral mandibles and onchia slightly smaller than in males. Three arched, mandibular plates with pointed tips projecting into stoma. Mandibular plates slightly curved and thin. Three onchia, one dorsal and two ventrosublateral, equal size, at the base of each mandible. Reproductive system didelphic, amphidelphic, ovaries reflexed. Vagina with thick walls and well developed sphincter muscles. Tail 4 anal body diameters in length, without setae.

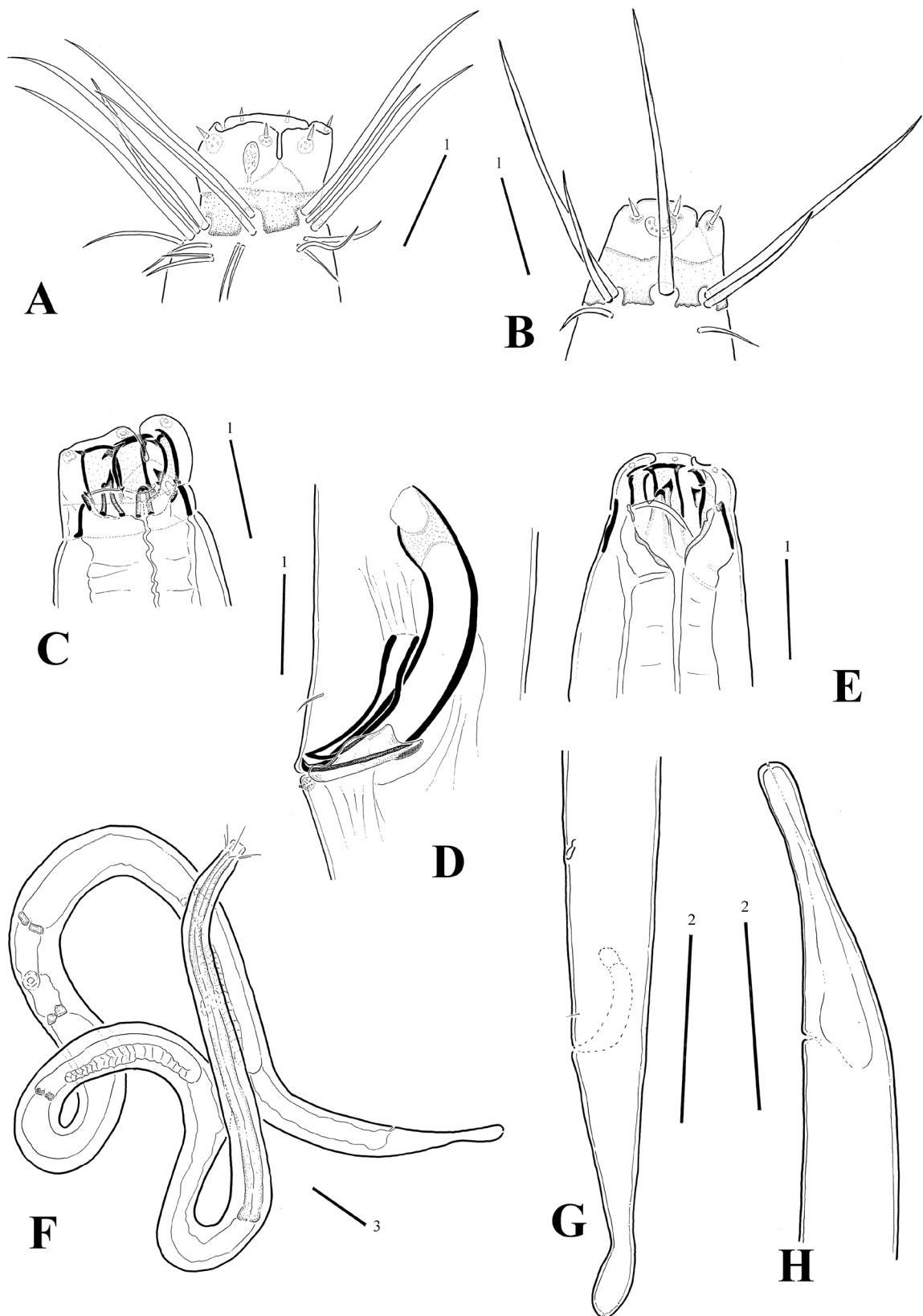
**Material examined.** Holotype Male, CNP-NEM N° 27798, collected on 1/4/2006 at “El Límite” beach (lat. 45°59'38"S, long. 67°35'31"W), during mid tide. Collector: Catalina Pastor. Paratypes two males and one female, same data as holotype, mounted on slide numbers CNP-NEM N° 27849, 27888, 27601.

**Type locality and habitat.** “El Límite” beach, 1/4/2006, mid tide 30 cm deep in sediment; salinity = 38 psu; temp. = 13.2–14.5 °C. Characteristics of surface sediment layer: mean particle size Q2 (50%) = 0.2; FF (%) = 0.8–1.2; SO index = 0.7–0.8; SK index = 1.0–1.2. Organic matter content/100g sediment = 1.2–1.5 mg.

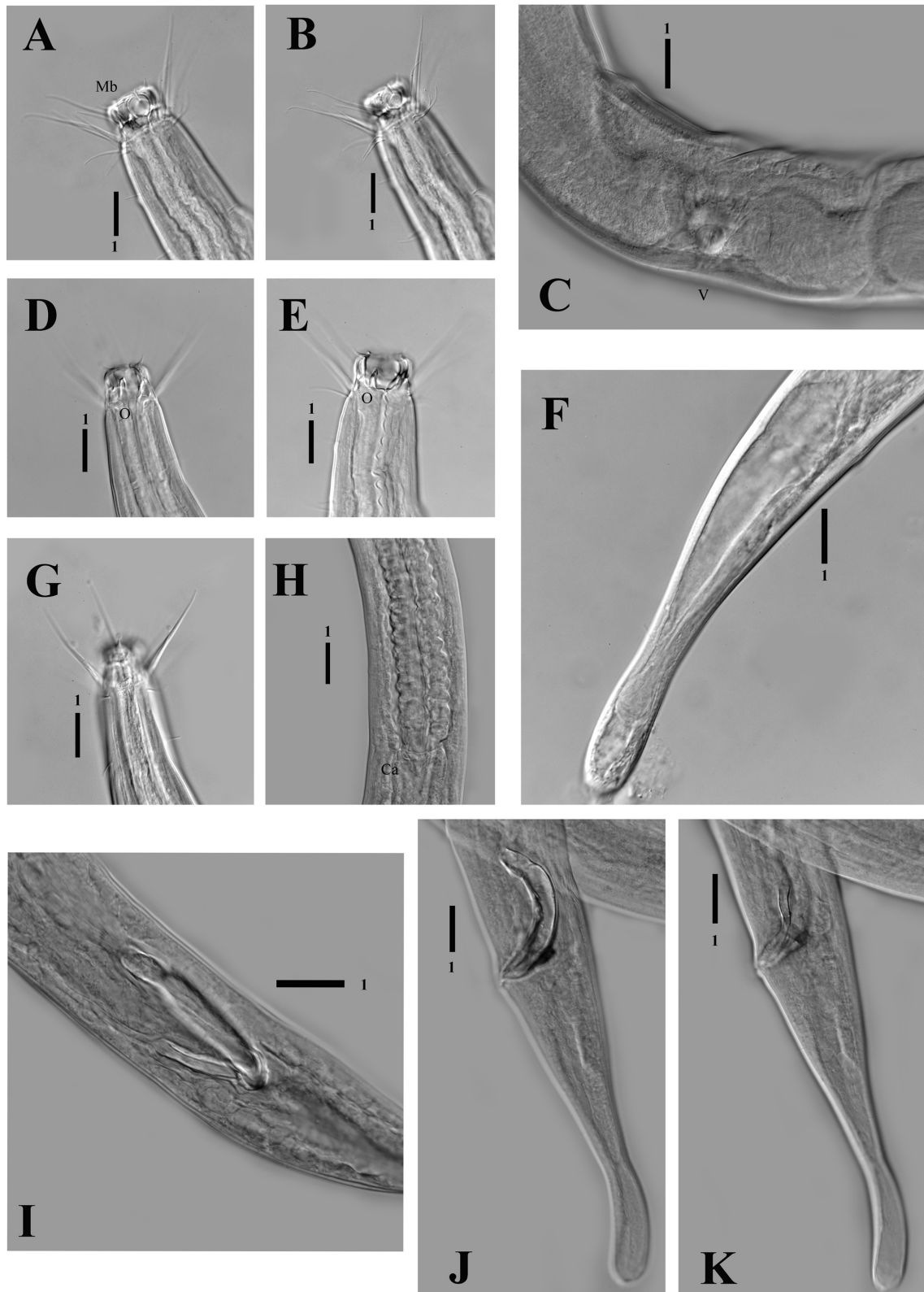


**TABLE 1.** Morphometrics of *Enoplolaimus variispiculum* **sp. n.** from Chubut Province, Argentina, and *Mesacanthoides flagellatum* **sp. n.** from Río Negro Province, Argentina. All measurements are in  $\mu\text{m}$  and in the form: min-max  $\pm$  s.d. (range).

Character	<i>Enoplolaimus variispiculum</i> <b>sp. n.</b>			<i>Mesacanthoides flagellatum</i> <b>sp.n.</b>		
	Male	Male	Female	Male	Male	Female
	Holotype	Paratypes	Paratype	Holotype	Paratype	Paratype
n		2	1		1	1
L	2500	2550–2570 $\pm$ 14	2720	5750	5900	7850
a	44.6	46.4–55.9 $\pm$ 6.7	38.8	71.9	75.6	78.5
b	4.7	4.5–4.9 $\pm$ 0.2	2.8	6.4	6.1	7.5
c	14.7	15.6–19.6 $\pm$ 2.8	17.0	6.8	7.2	7.5
c'	4.2	3.9–4.2 $\pm$ 0.3	4.0	10.6	11.7	16.1
V%	–	–	50.5	–	–	49.7
Maximum body diameter	56.0	46.0–55.0 $\pm$ 6.3	70.0	80.0	78.0	100.0
Anal or cloacal Diameter	40.0	38.0–42.0 $\pm$ 2.8	40.0	78.0	70.0	65.0
Body diameter at nerve ring	45.0	38.0	45.0	60.0	70.0	59.0
Body diameter at cardia	55.0	40.0–48.0 $\pm$ 5.6	70.0	90.0	75.0	90.0
Anterior end to cephalic setae	24.0	22.0–23.0 $\pm$ 0.7	20.0	34.0	35.0	38
Anterior end to nerve ring	150	150–160 $\pm$ 7.1	270	280	300	390
Anterior end to cardia	535	525–570 $\pm$ 31.8	590	900	960	1050
Anterior end to ano/cloaca	2340	2405–2420 $\pm$ 11	2530	5100	5170	6950
Anterior end to vulva	–	–	1620	–	–	10.6
Inner labial sencilla (6)	5.0	5.0	5.0	14.0	16.0	11.0
Outer labial sencilla (6)	54.0	48.0–50.0 $\pm$ 1.4	56.0	73.0	80.0	69.0
Cephalic setae (4)	37.0	34.0–36.0 $\pm$ 1.4	26.0	30.0	32.0	29.0
Subcephalic setae (6)	47.0	38.0–40.0 $\pm$ 1.4	–	–	–	–
Cervical setae (6)	16.0	15.0–16.0 $\pm$ 0.7	–	–	–	–
Cervical setae (6)	12.0	11.0–12.0 $\pm$ 0.7	–	–	–	–
Cervical setae (4)	14.0	12.0–13.0 $\pm$ 0.7	12.0	40.0	45.0	20.0
Anterior organ length (in cord)	7.0	5.0–6.0 $\pm$ 0.7	9.0	–	–	–
Cephalic capsule diameter	26.0	23.0–24.0 $\pm$ 0.7	27.0	65.0	70.0	74.0
Cephalic capsule height	8.0	7.0–8.0 $\pm$ 0.7	7.0	26.0	33.0	28.0
Lip-lobe height	20	20.0–23.0 $\pm$ 2.1	18	15.0	16.0	20.0
Mandible height	10.0	9.0–10.0 $\pm$ 0.5	13.0	30.0	32.0	34.0
Mandible width	8.0	10.0–11.0 $\pm$ 0.5	8.5	15.0	16.0	16.0
Onchium	9.0	9.0	9.0	23.0	24.0	25.0
Spicule (right, arc) / both	70.0	65.0–42.0 $\pm$ 4.9	–	325	310	–
Spicule (left, arc)	33.0	32.0–34.0 $\pm$ 1.4	–	–	–	–
Gubernaculum	22.0	20.0–21.0 $\pm$ 0.7	–	35.0	34.0	–
Pre-anal supplement length	8.0	8.0–10.0 $\pm$ 1.4	–	13.0	12.0	–
Cloacal opening to supplement	100	120–130 $\pm$ 7.1	–	100	110	–
Cloacal opening to preanal setae	15.0	13.0–15.0 $\pm$ 1.4	–	–	–	–
Tail	170	130–165 $\pm$ 24.7	160	830	820	1050



**FIGURE 2.** *E. variispiculum* n. sp. Line drawings. A. Cephalic sense organs on anterior end of male holotype; B. Cephalic sense organs on female paratype; C. Buccal cavity and mandibles of male holotype; D. Copulatory apparatus, spicules and gubernaculum of male holotype; E. Buccal cavity and mandibles of female paratype; F. Entire female paratype showing vulva and gonadal apparatus; G. Posterior end of male holotype; H. Posterior end of female paratype. Scale bar: 1 = 20 µm; 2 and 3 = 100 µm.



**FIGURE 3.** *E. variispiculum* **n. sp.** Micrographs of slide mounted specimens. A. Buccal cavity and mandibles of male holotype; B. Cephalic sense organs on anterior end of male holotype; C. Vulva opening in subventral view of female paratype; D. Onchia in female paratype; E. Onchia in male paratype; F. Posterior end of female paratype; G. Cephalic sense organs on female paratype; H. Cardia and muscular esophagus in male holotype; I. Subventral view of copulatory apparatus, spicule and gubernaculum of male paratype; J. Lateral view of left spicule, gubernaculum and tail of male holotype; K. Lateral view of right spicule, gubernaculum and tail of male holotype. Abbrev: Ca = Cardia; Mb = Mandible; O = Onchia; V = Vulva. Scale bar: 1 = 20  $\mu$ m.



**TABLE 2.** Comparison of diagnostic morphological characters of male holotypes of *Enoplolaimus* species without an apophysis on the gubernaculum and with short tail. \*Measurements taken from original holotype drawings. \*\*Not seen. \*\*\* No holotype head drawings. \*\*\*\* Mean values.

Species	Body length (µm)	a	b	c	Length inner labial sensilla in µm	Length outer labial sensilla in µm	Ce-phalic capsule height/width	Man-dible height/width	On-chia length in µm	Spicule length in c.b.d	Gubernaculum length in c.b.d.	Tail length in c.b.d	Distribution
<i>E. abnormis</i> Kreis, 1928	3248	54.3	9.7	15.6	16.6	45.0****	**	**	4.0	1.2*	0.5*	4.6	Spilberguen, Artic
<i>E. balgensis</i> Skwarra 1921, Gerlach, 1953	1800	24.0	3.5	10.0	15.0****	45.0****	**	**	15.5	1.2	0.5*	3.5*	North Sea
<i>E. enatus</i> Hopper, 1962	1650	55.0	4.4	10.9	10.5****	45.5****	2.4	1.2	8.0	1.3	0.5	5.5****	Rhode island, USA
<i>E. halophilus</i> Ditlevsen, 1928	5200	62.0	5.4	23.8	17.8*	67.4*	0.3*	1.3*	7.1*	1.9*	1.2*	4.1*	Greenland
<i>E. lenunculus</i> Wieser, 1959	2500	46.4	5.4	25.5	13.5****	47.5****	***	1.2	***	1.3	0.3	3.4****	Puget sound, USA
<i>E. longigubernaculum</i> Shimada <i>et al.</i> , 2009	5150	60.6	5.3	22.9	19	52	0.2*	0.7*	10*	1.3	1.4	3.5	Northern Japan
<i>E. mus</i> Inglis, 1964	4700	31.3	4.5	16.2	17.0	105.0	1.3	1.2	27.0	1.0	0.5*	3.6	South Africa
<i>E. paraltoralis</i> Wieser, 1959	1670	42.0	4.5	17.0	11.5****	64.0****	0.7	5.8	***	1.2	0.4	4.3****	Puget Sound, USA
<i>E. varispiculum</i> sp. n.	2540	49.0	4.7	16.6	5.0	50.7	0.3	2.5	8.0	1.7	0.6	3.9	Chubut, Argentina

**Etymology.** “*variispiculum*” from Latin word “variis” = various and “spicules” referring to the different size and shape of spicules.

**Differential diagnosis.** *E. variispiculum* **sp. n.** is characterized by having asymmetric spicules, different in size, shape and distal tips. It is the only species with this characteristic in the whole genus. It also has the shortest inner labial sensilla than all the species without an apophysis on the gubernaculum and with short tail (group 6, Fadeeva & Zograf, 2010) plus the species added in this paper, nine species in total. In Table 2 we show the comparison of the diagnostic morphological characters of those nine species.

*E. variispiculum* **sp. n.** is related to *E. enatus* Hopper, 1962, *E. lenunculus* Wieser, 1959, *E. mus* Inglis, 1964 and *E. paralitoralis* Wieser, 1959.

*E. enatus* has similar outer labial sensilla length, onchia and gubernaculum length in cloacal body diameter, and de Man’s ratio “b”. But it differs in length of inner labial sensilla (5 µm vs 10–11 µm), cephalic setae length (35.7 µm vs 19–20 µm), cephalic capsule height/width (0.3 vs 2.4), mandible height/width (2.5 vs 1.2), spicule length in cloacal body diameter (two different 1.7 and 0.8 vs 1.3) and tail length in cloacal body diameter (3.9 vs 5.0–6.0).

*E. lenunculus* has similar general size, outer labial sensilla, de Man’s ratio “a, b, c” and tail length. But it differs in length of inner labial sensilla (5 µm vs. 12–15 µm), length of cephalic sensilla (35.7 µm vs 22.0 µm), mandible height/width (2.5 vs 1.2), and spicule length in cloacal body diameter (two different 1.7 and 0.8 vs 1.3).

*E. mus* has similar “b” and “c” de Man’s ratios, gubernaculum and tail length in cloacal body diameter. But it differs in total body length (4700 vs 2540), inner labial, outer labial and cephalic sensilla length (17 vs 5 µm; 105 vs 50 µm; 55 vs 35.7 µm), mandible H/W (1.2 vs 2.5), and onchia length (27 vs 8 µm).

*E. paralitoralis* has similar “b” and “c” de Man’s ratios, cephalic sensilla and tail length. But it differs in total length (1670 vs 2540 µm), in inner labial and outer labial sensilla length (10–12 vs 5 µm; 62–66 vs 50.7), in cephalic capsule H/W (0.7 vs 0.3), and in mandible H/W (5.8 vs 2.5).

## Family Thoracostomopsidae Filipjev, 1927

### Subfamily Enoplolaiminae De Coninck, 1965

#### Genus *Mesacanthoides* Wieser, 1953

**Emended diagnosis.** From Wieser, 1953 (Smol *et al.* 2014). Enoplolaiminae. Transition between *Mesacanthion* and *Enoploides*. Lips striated or not striated. Mandibles solid or dense mottled with claws. Teeth shorter than mandibles. Marine.

#### *Mesacanthoides flagellatum* **sp. n.**

(Figure 1A, 4, 5; Table 1, 3)

**Measurements.** See Table 1.

**Description. Males:** Body cylindrical, tapering posteriorly and ending in a flagellate tail. Cuticle finely striated. Head rectangular in shape, not set off, with three lips (one dorsal and two ventrosublateral) with striations and separated by deep clefts, 18 µm high. The striations are delicate and can only be seen along the lips borders. Each lip bearing a small glandular area on the tips of the lips, and two inner labial sensilla, 0.3 corresponding body diameters in length. Six outer labial sensilla, 1.4 corresponding body diameters in length, inserted at the level of posterior margin of cephalic capsule. A second crown of four cephalic setae 0.6 corresponding body diameters in length. Two crowns of cervical setae in dorsosublateral and ventrosublateral position. First crown of 4 setae, 0.7 corresponding body diameters in length, second crown of 4 setae, 0.4 corresponding body diameters in length. Amphids not seen. Body with somatic setae scattered along anterior pharyngeal area.

Cephalic capsule well developed, shorter than wide. Anterior margin at the base of the lips. Posterior margin with incisions accommodating the first crown of outer labial setae and cephalic setae. Cuticularized arch surrounding the setae insertion. Three arched, mandibular plates with anterior curved bar, to the extremes claws-like, with no conspicuous pointed tips, projecting into stoma. Mandibular plates with “v” shaped incision posterior to the

anterior bar. Mandible surface is densely mottled, but sufficiently transparent to see the onchia. Three onchia, one dorsal and two ventrosublateral, equal in size, at the base of each mandible. Mandibular ring with three cuticularized semicircular structures between the mandibles, at level of half of the onchia length. Amphids, excretory pore and metanemes not observed. Anterior part of the pharynx around buccal cavity globose, then cylindrical.

Reproductive system diorchic, anterior testis outstretched, posterior one reflexed. Spicules with diagonal reinforcement (4.2 cloacal body diameters in length), with pointed distal end. Gubernaculum complex, “s” shaped with a dorsal small apophysis, a dorsoventral knob and a distal cannula part ending in two small teeth. One precloacal supplement present. No setae or papillae were observed in pre and post cloacal area. Tail flagellate, 10.9 cloacal body diameter long.

**Females:** Similar to male in general body shape, but much larger. No differences in shape and number of labial and cephalic setae. Six labial sensilla 4.3 corresponding body diameters in length, six long outer labial sensilla 1.2 corresponding body diameters in length and four short cephalic setae 2.5 corresponding body diameters in length, inserted at posterior margin of cephalic capsule. One crown of four cervical setae 3.7 corresponding body diameters in length. Three arched, mandibular plates with pointed tips projecting into stoma. Mandibular plates same in shape as in males and with mottled surface. Three onchia, one dorsal and two ventrosublateral, equal in size. Reproductive system didelphic, amphidelphic, ovaries reflexed. Vagina with thick walls and well developed sphincter muscles. Tail flagellate, 16.1 anal body diameters in length.

**Material examined.** Holotype Male, CNP-NEM N° 1282, collected on 17/6/2006 at “Balneario San Antonio Oeste” (lat. 40° 43' 55"S, long. 64° 56' 52"W), during low tide. Collector: Catalina Pastor. Paratypes one male and one female, same data as holotype mounted on slide numbers CNP—NEM N° 1283–1284.

**Type locality and habitat.** “Balneario San Antonio Oeste”, winter, temperature = 9.6 °C, salinity = 33.5 psu. They were found 26 cm depth in sediment, at low tide. Characteristics of sediment: mean particle size Q2 (50%) = 0.4; VCS = 96.8%; SO index = 0.54; Organic matter / 100 g sediment = 4.1 mg.

**Etymology.** “*flagellatum*” from latin word “flagellum”, in relation to the tail shape.

**Differential diagnosis.** *M. flagellatum* sp. n. is characterized by high striated lips, with a striation only visible along the borders of the lips. The mandible texture is weakly cuticularized and mottled. Mandibular plates with pronounced “v” shaped incision posterior to the anterior bar. Cephalic capsule shorter than wide, with setae in posterior position. Males with long spicules, with “s” shaped gubernaculum, precloacal supplement and flagellate tail.

*M. flagellatum* sp. n. is related to *M. fibulatus* Wieser & Hopper, 1967, *M. psittacus* Wieser & Hopper, 1967 and *M. magna* Coles, 1977. For more information see Table 3.

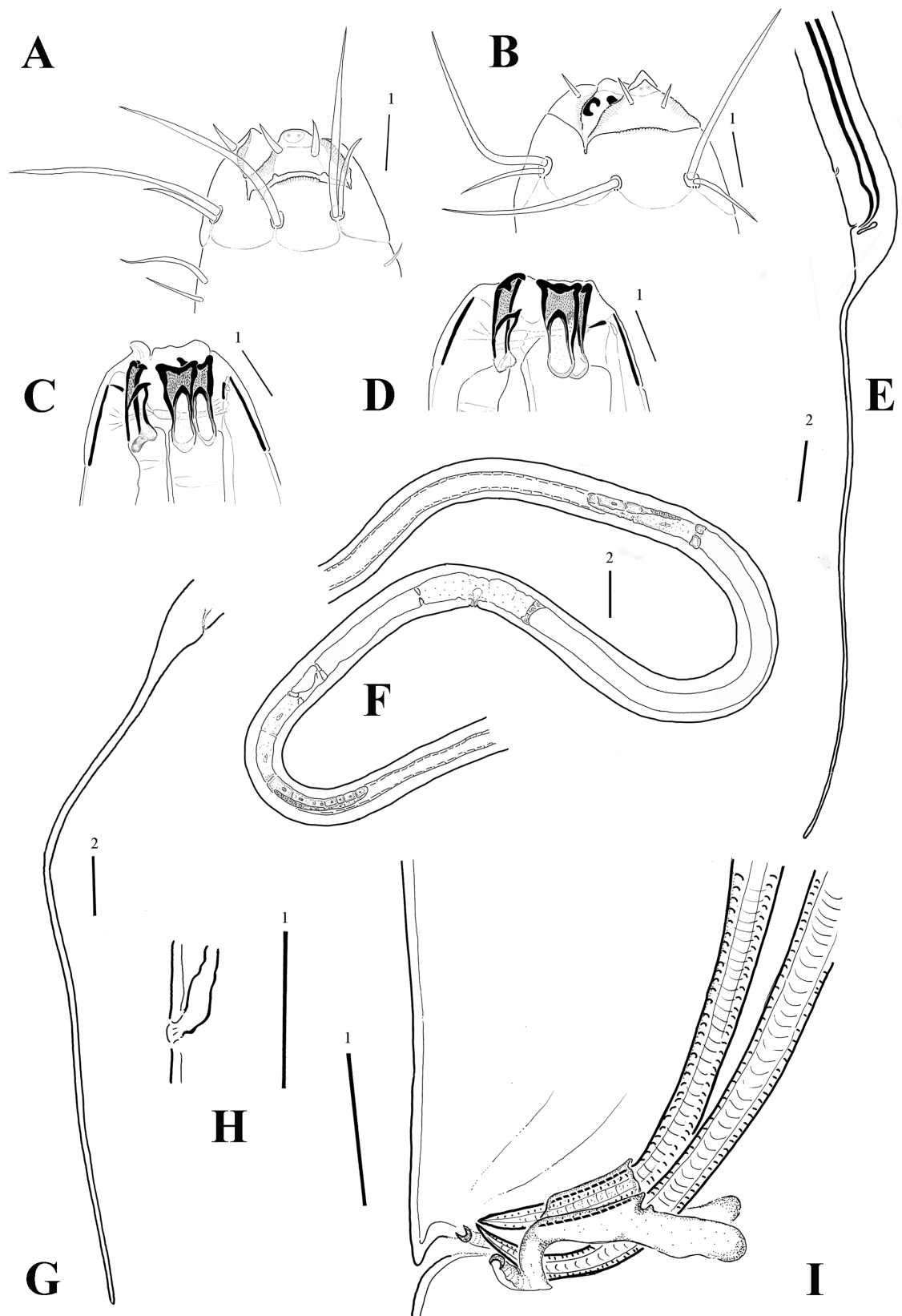
*M. fibulatus* has similar body size, cephalic capsule proportions (height/width), spicules length in anal diameters. But it differs in having unstriated lips, different length of inner labial sensilla (14–16 µm vs 7 µm), cephalic setae length (73–80 µm vs 21 µm), mandible length (30.0–32.0 µm vs 18 µm), tail length (10.6–11.7 c.b.d. vs 20 c.b.d.) and de Man's ratio “a” (71.9–75.6 vs 57.3), “b” (6.1–6.4 vs 8.5) and “c” (6.8–7.2 vs 12.3).

*M. psittacus* has similar cephalic capsule height/width (0.5 vs 0.4–0.5), mandible height/width (2.0), inner labial sensilla length (15.0 µm vs 14.0–16.0 µm), and cephalic setae length (30–32 µm vs 32.7 µm). But it differs in having unstriated lips, different de Man's ratios “a, b, c”, spicule length in c.b.d (4.2–4.4 vs 1.0), gubernaculum length in c.b.d (0.4–0.5 vs 1.9) and tail length in c.b.d. (10.6–11.7 vs 5.0).

*M. magna* has similar length of inner labial sensilla (10–15 µm vs 10.0–15.0 µm), outer labial sensilla (73–80 µm vs 50.0–100.0 µm), cephalic setae length (30–32 µm vs 40 µm), mandible length (30.0–32.0 µm vs 45.0–50.0 µm), onchia length (23.0–24.0 µm vs 25.0–27.0 µm) and De Man's ratios “a” (71.9–75.6 vs 61.0–70.4) and “b” (6.1–6.4 vs 2.1–6.2). But differs in the small size (5750.0–5900.0 µm vs 12200.0–15500.0 µm), in having unstriated lips, in spicule length in c.b.d. (4.2–4.4 vs 7.6–9.0), tail length in c.b.d (10.6–11.7 vs 2.0) and De Man's ratio “c” (6.8–7.2 vs 69.7–83.8).

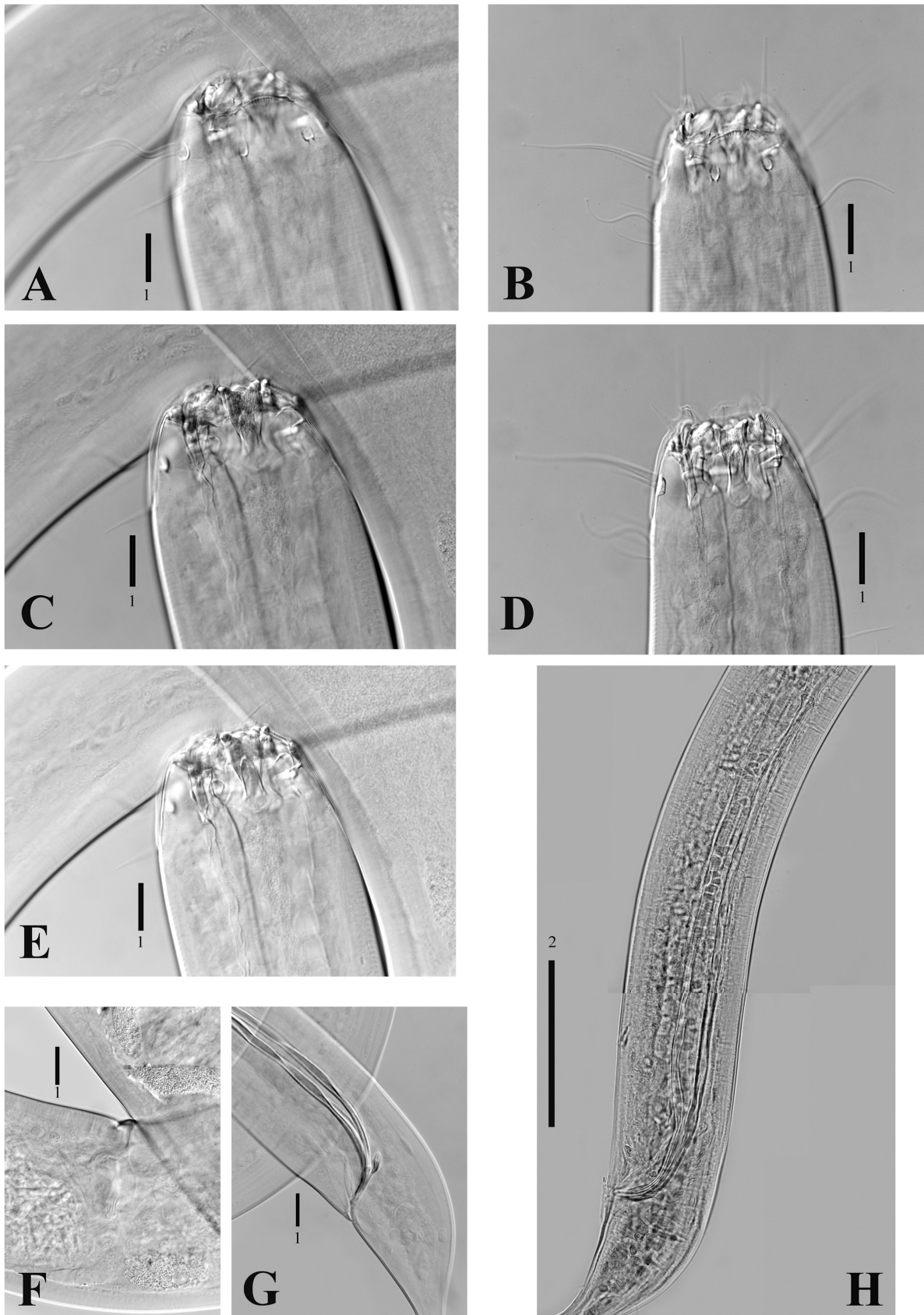
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**FIGURE 4.** *M. flagellatum* **sp. n.** Line drawings. A. Cephalic sense organs on anterior end of male holotype; B. Cephalic sense organ on female paratype; C. Buccal cavity and mandibles of male holotype; D. Buccal cavity and mandibles of female paratype; E. Posterior end of male holotype; F. Vulva and gonadal apparatus of female paratype; G. Posterior end of female paratype; H. Detail of precloacal organ in male holotype; I. Copulatory apparatus, spicules and gubernaculum of male holotype. Scale bar: 1= 20 µm; 2= 100 µm.





**FIGURE 5.** *M. flagellatum* sp. n. Micrographs of slide mounted specimens. A. Cephalic sense organs on female paratype; B. Cephalic sense organs of male holotype; C. Buccal cavity and mandibles of female paratype; D. Mandibles and onchia on male paratype; E. Mandibles and onchia in female paratype; F. Vulva opening in female paratype; G. Detail of gubernaculum of male paratype; H. Lateral view of precloacal organ, gubernaculum and spicules of male holotype. Scale bar: 1= 20  $\mu$ m.



**TABLE 3.** Comparison of diagnostic morphological characters of male holotypes of *Mesacanthoides* species. In *M. magna* Coles, 1977 the male holotype measurements are not specified. \*Measurements taken from original holotype drawings. \*\*No measurements are given by the author. \*\*\*Mean values.

Species	Body length (µm)	a	b	c	Length inner labial sensilla in µm	Length outer labial sensilla in µm	Length cephalic setae in µm	Cephalic capsule height/width	Man-dible length in µm	Man-dible height/width	Onchia length in µm	Spicule length in c.b.d	Gubernaculum length in c.b.d	Tail length in c.b.d	Distribution
<i>M. brevicaudatus</i> Keppner 1987	1454	52.1	3.18	22.9	14	31	13	0.6*	26	2.6	15.0*	2.1	not seen	2.6	Florida,EEUU
<i>M. caputmedusae</i> (Ditlevsen, 1918) Wieser, 1959	6100	69	5.1	18.9	**	**	**	**	**	**	**	**	**	**	Denmark
<i>M. fibulatus</i> Wieser & Hopper, 1967	4300	57.3	8.5	12.3	7.0*	42.0	21.0	0.5*	18.0	1.8	9.0	4.0	1.0	20.0	Florida,EEUU
<i>M. flagellatum</i> sp.n.	5750	71.9	6.4	6.8	14.0	73.0	30.0	0.4	30.0	2.0	23.0	4.2	0.4	10.6	Río Negro, Argentina
<i>M. latignathus</i> (Ditlevsen, 1918) Wieser, 1953	4500	70	4.4	14.0	**	**	**	**	**	**	**	**	**	**	Denmark
<i>M. magna</i> Coles, 1977	12200–15500	61.0–70.4	2.1–6.2	69.7–83.8	10.0–15.0	50.0–100.0	40.0	0.6*	45–50	1.5–1.6	25.0–27.0	7.6–9.0	0.9–1.1	2.0	Cape province, South Africa
<i>M. psittacus</i> Wieser & Hopper, 1967	2320	46.4	4.3	10.4	15.0	62.0	27.0	0.5*	19.0	2.0*	9.5*	1.0*	1.9	5.0	Florida,EEUU
<i>M. sculptilis</i> Wieser, 1953	1870	37.6	4.2	16.0	13.0	33.0	16.5	0.3*	19.5	1.5	10.0	1.7	not seen	2.6	Chile
<i>M. sinuosus</i> Wieser, 1959	3400	77.4	5.5	32.3	10.5***	41.0	18.0	0.5	19.0	1.3	13.0	1.2	0.5	3.2***	Florida,EEUU

## References

- Allgén, C.A. (1929) Freilebende marine Nematoden aus den Umgebungen der Staatlichen Zoologischen Station Kristineberg an der Westküste Schwedens. *Capita Zoologica*, 2, 1–52.
- Allgén, C.A. (1940) Über einige norwegische marine Tiefen-Nematoden. *Folia Zoologica et Hydrobiologica*, 10, 258–281.
- Allgén, C.A. (1959) Freelifving marine nematodes. In: Odhner, N.H. (Ed.), *Further zoological results of the Swedish Antarctic expedition, 1901–1903 under the direction of Dr. Otto Nordenskjöld*. Kungl Boktryckeriet P.A. Norstedt & Söner, Stockholm, pp. 293.
- Bezerra, T.N., Eisendle-Flöckner, U., Hodda, M., Holovachov, O., Leduc, D., Mokievsky, V., Peña Santiago, R., Sharma, J., Smol, N., Tchesunov, A., Venekey, V., Zhao, Z. & Vanreusel, A. (2021) Nemys: World Database of Nematodes. Available from: <http://nemys.ugent.be> (accessed 16 July 2021)  
<https://doi.org/10.14284/366>
- De Coninck, L. (1965) Systématique des nématodes. In: Grassé, P.P. (Ed.) *Traité de Zoologie: anatomie, systématique, biologie*. 4 (2). *Nemathelminthes*. Masson, Paris, pp. 586–681.
- de Jonge, V.N. & Bouwman, L.A. (1977) A simple density separation technique for quantitative isolation of meiobenthos using the colloidal silica Ludox-TM. *Marine Biology*, 42, 143–148.  
<https://doi.org/10.1007/BF00391564>
- de Man, J.G. (1893) Cinquième Note sur les Nématodes libres de la mer du Nord et de la Manche. *Mémoires de la Société Zoologique de France*, 6, 81–125.
- Ditlevsen, H. (1928) Free-living Nematodes from Greenland, Land and Freshwater. *Meddelelser om Gronland*, 23, 157–250.
- Fadeeva, N.P. & Zograf, J.K. (2010) New and known species of *Enoplolaimus* (Enoplida: Thoracostomopsidae) from the Sea of Japan. *Nematology*, 12, 731–749.  
<https://doi.org/10.1163/138855409X12607871174535>
- Filipjev, I.N. (1927) Les nématodes libres des mers septentrionales appartenant à la famille des Enoplidae. *Archiv für Naturgeschichte*, 91 A, 1–216.
- Gerlach, S.A. (1953) Die Nematodenbesiedlung des Sandstrandes und des Küstengrundwassers an der italienischen Küste. I. Systematischer Teil. *Archiv Zoologie Italian*, 37, 517–640.
- Hopper, B.E. (1962) Free-living marine nematodes of Rhode Island waters. *Canadian Journal of Zoology*, 40, 41–52.  
<https://doi.org/10.1139/z62-007>
- Inglis, W.G. (1964) The marine Enoplida (Nematoda): A comparative study of the head. *Bulletin of the British Museum (Natural History) Zoology*, 11, 265–376.  
<https://doi.org/10.5962/bhl.part.4719>
- Jeong, R., Tchesunov, A.V. & Lee, W. (2020) Two species of Thoracostomopsidae (Nematoda: Enoplida) from Jeju Island, South Korea. *PeerJ*, 8, e9037.  
<https://doi.org/10.7717/peerj.9037>
- Keppner, E.J. (1987) Five New Species of Free-living Marine Nematodes (Nematoda: Enoplida) from a Northwest Florida, U.S.A. Estuary. *Transactions of the American Microscopical Society*, 106, 333–347.  
<https://doi.org/10.2307/3226224>
- Shimada, D., Kajihara, H. & Mawatari, S.F. (2009) Three new species of free-living marine nematodes (Nematoda: Enoplida) from Northern Japan. *Species Diversity*, 14, 137–150.  
<https://doi.org/10.12782/specdiv.14.137>
- Somerfield, P.J., Warwick, R.M. & Moens, T. (2005) Meiofauna Techniques. In: Eleftheriou, A. & McIntyre, A. (Eds.), *Methods for the Study of Marine Benthos*. Blackwell Science, Oxford, pp. 229–272.  
<https://doi.org/10.1002/9780470995129.ch6>
- Smol, N. & Coomans, A. (2006) Order Enoplida. In: Abebe, E., Andrassy, I. & Traunspurger, W. (Eds.), *Freshwater nematodes: ecology and taxonomy*. CABI Publishing, Wallingford, pp. 225–292.  
<https://doi.org/10.1079/9780851990095.0225>
- Smol, N., Muthumbi, A. & Sharma, J. (2014) Order Enoplida. In: Schmidt-Rhaesa, A. (Ed.), *Gastrotricha, Cycloneuralia and Gnathifera. Vol. 2. Nematoda*. De Gruyter, Berlin/Boston, pp. 193–249.
- Ssaweljev, S. (1912) Zur Kenntnis der freilebenden Nematoden des Kolafjords und des Relictensee Mogilnoje. *Travaux de la Société impériale des naturalistes de St. Pétersburg*, 43, 108–126.
- Wieser, W. (1953) Free-living marine nematodes I. Enoploidea. *Acta Universitatis Lundensis*, Neue Folge, 49, 1–155.
- Wieser, W. (1959) *Free Living Nematodes and Other Small Invertebrates of Puget Sound Beaches*. University of Washington Press, Seattle, Washington, ix + 179 pp.
- Wieser, W. & Hopper, B. (1967) Marine nematodes of the east coast of North America. I. Florida. *Bulletin of the Museum of Comparative Zoology at Harvard College*, 135, 239–344.