Description of *Marylynnia puncticaudata* n. sp. (Nematoda, Cyatholaimidae) from Bizerte Lagoon, Tunisia

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Abstract

Description of Marylynnia puncticaudata *n. sp. (Nematoda, Cyatholaimidae) from Bizerte Lagoon, Tunisia.*— A new free–living marine nematode species of Cyatholaimidae, *Marylynnia puncticaudata* n. sp. from Bizerte Lagoon (Tunisia) is morphologically described. Males are characterized by a slightly larger body than females, a cephalic ring followed by ten subcephalic setae, modified cuticular punctuation, caudal lateral differentiation of large dots, and strongly cuticularized gubernaculum with a unique shape and bidenticulated distal half. The cuticle ornamentation of females is similar to the males. However, their caudal lateral differentiation is composed of smaller and more spaced dots. An updated morphological key to species of *Marylynnia* is given.

Key words: Free–living marine nematodes, Bizerte Lagoon, *Marylynnia puncticaudata* n. sp.

Resumen

Descripción de Marylynnia puncticaudata *sp. n. (Nematoda, Cyatholaimidae) del lago de Bizerta, Túnez*.— Se describe morfológicamente una nueva especie de nematodo marino de vida libre de la familia Cyatholaimidae: *Marylynnia puncticaudata* sp. n. del lago de Bizerta (Túnez). Los machos se caracterizan por tener un cuerpo ligeramente más grande que las hembras, un anillo cefálico seguido de diez sedas subcefálicas, puntuación distinta de la cutícula, diferenciación laterocaudal de grandes puntos y gubernáculo muy cuticularizado con una forma única y la mitad distal bidenticulada. La ornamentación de la cutícula de las hembras es parecida a la de los machos. Sin embargo, su diferenciación laterocaudal se compone de puntos más pequeños y espaciados. Se ofrece una clave morfológica actualizada de la especie *Marylynnia*.

Palabras clave: Nematodos marinos de vida libre, lago de Bizerta, *Marylynnia puncticaudata* sp. n.

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Introduction

To date, 6,900 species of free–living marine nematodes have been morphologically described (Appletans et al., 2012), most of which are from West Europe and North America. Very few have been described from the African continent (Semprucci & Balsamo, 2012). In the case of Tunisia, 249 species of free–living marine nematodes have been collected since 1977 from several lagoons, beaches and mudflats (Boufahja et al., 2014). Of these, 20 species are being considered as new to science (Boufahja et al., 2014). So far, morphological descriptions have been published, by Aïssa & Vitiello (1977), for three validated species collected from the Northern Lake of Tunis (Tunisia): *Chromadorina metulata*, *Metalinhomoeus numidicus* and *Synonchiella edax*. However, 17 species have not yet been described (Boufahja et al., 2014).

Our specimens are recognized as *Marylynnia,* which belong to the second richest nematode family in Tunisia, the Cyatholaimidae (25 reported species) (Boufahja et al., 2014). The aim of the present paper is to provide new insights into the composition of nematode in the Tunisian area, to describe a new species of this genus, and to propose an updated identification key to the species of the genus.

The family Cyatholaimidae, included in the superfamily Chromadoroidea Filipjev 1917, was first described by Filipjev (1918) and comprises two subfamilies, 23 genera and 215 species (Hodda, 2011). The genus *Marilynia* was established by Hopper (1972) with type species *Marilynia annae* (synonym: *Longicyatholaimus annae* Wieser & Hopper, 1967), and thereafter modified to *Marylynnia* by Hopper (1977). According to Platt and Warwick (1988), *Marylynnia* species are characterized by cuticle with transverse rows of dots with a lateral differentiation of larger and more widely spaced dots. Two types of pores are present on the cuticle: simple rounded and longitudinally oval. The genus *Marylynnia* is closely related to *Longicyatholaimus* but differs by the presence of a larger buccal cavity, prominent dorsal tooth and paired ventrosublateral teeth, a less complex form of the gubernaculums, and lateral modified punctuations on the conoid portion of the tail.

Material and methods

Sediment collection and processing

Undisturbed sediment samples were taken on 1 VII 13 in the channel zone of Bizerte Lagoon (37° 15.906' N, 09° 52.052' E) (fig. 1). At low tides, the sediment was removed following parallel tracks that were 5cm deep. The uppermost 2 cm of the intermediate zones were then collected using a large spatula (Boufahja & Semprucci, in press). Immediately, the sediment was fixed with neutralized 4% formalin for morphology details, and a few drops of Rose Bengal (0.2 g/l) were added to stain the specimens (Higgins & Thiel, 1988).

In the laboratory, all sediment samples were rinsed with a gentle jet of freshwater over a 1 mm and 40 µm sieves and nematode separation followed the classic decantation–floatation method (Mahmoudi et al., 2007). Nematodes were taken from every sampled core under a 50x stereoscopic microscope (Model Wild Heerbrugg M5A) and fixed in neutralized 4% formalin.

Mounting and identification

Nematodes were transferred to a 9:1 (V:V) solution of 50% ethanol:glycerol in block cavity to slowly evaporate ethanol. They were then mounted in a drop of anhydrous glycerol on permanent slides (Seinhorst, 1959).

Type specimens were deposited in the collections of free–living marine nematodes at the Faculty of Sciences of Bizerte (collection code: FB–BL–NV1) (Boufahja et al., 2014). Drawings were done directly from the slide using (1) a Nikon DS–Fi2 camera coupled to a Nikon microscope (Image Software NIS Elements Analysis Version 4.0 Nikon 4.00.07 (build 787) 64 bit) and (2) a Olympus XC50 camera coupled to a Olympus BX53 microscope (Image Software CellSens Standard Version 1.6). All measurements (not ratios) are given in micrometers (µm) and all curved structures are measured along the arc (table 1). The abbreviations used in the text are as follows: a. Body length divided by maximum body diameter; b. Body length divided by pharyngeal length; c. Body length divided by tail length; c'. Tail length divided by anal body diameter; abd. Anal body diameter; cbd. Corresponding body diameter; hd. Head diameter at the level of the cephalic setae; L. Body length; M. Maximum body diameter; Spic. Spicule length along arc; V%. Position of vulva from anterior end expressed as a percentage of total body length; LMPs. Lateral modified ponctuations, that is, ring–shaped connections between the punctuations.

Results

Systematic account

Order Chromadorida Chitwood, 1933
Suborder Chromadorina Filipjev, 1929
Superfamily Chromadoroidea Filipjev, 1917
Family Cyatholaimidae Filipjev, 1918
Genus *Marylynnia* Hopper, 1977

Marylynnia puncticaudata n. sp. (figs. 2–5)

Type material
Ten males and ten females were observed. Holotype: 1 ♂ (collection code: FB–BL–NV1–1); paratypes: 2–10 ♂(collection codes: FB–BL–NV1–2–10) and 1–10 ♀ (collection codes: FB–BL–NV1–11–20). The type materials are held at the Laboratory of Biomonitoring of the Environment located in the Faculty of Sciences of Bizerte (Carthage University, Tunisia).

Sampling date, type locality and habitat
The specimens were collected on 1 VII 13 in a poorly sorted sediment with a mixture of coarse sand and mud from the channel zone of Bizerte Lagoon (37° 5.906' N, 09° 52.052' E) (fig. 1).

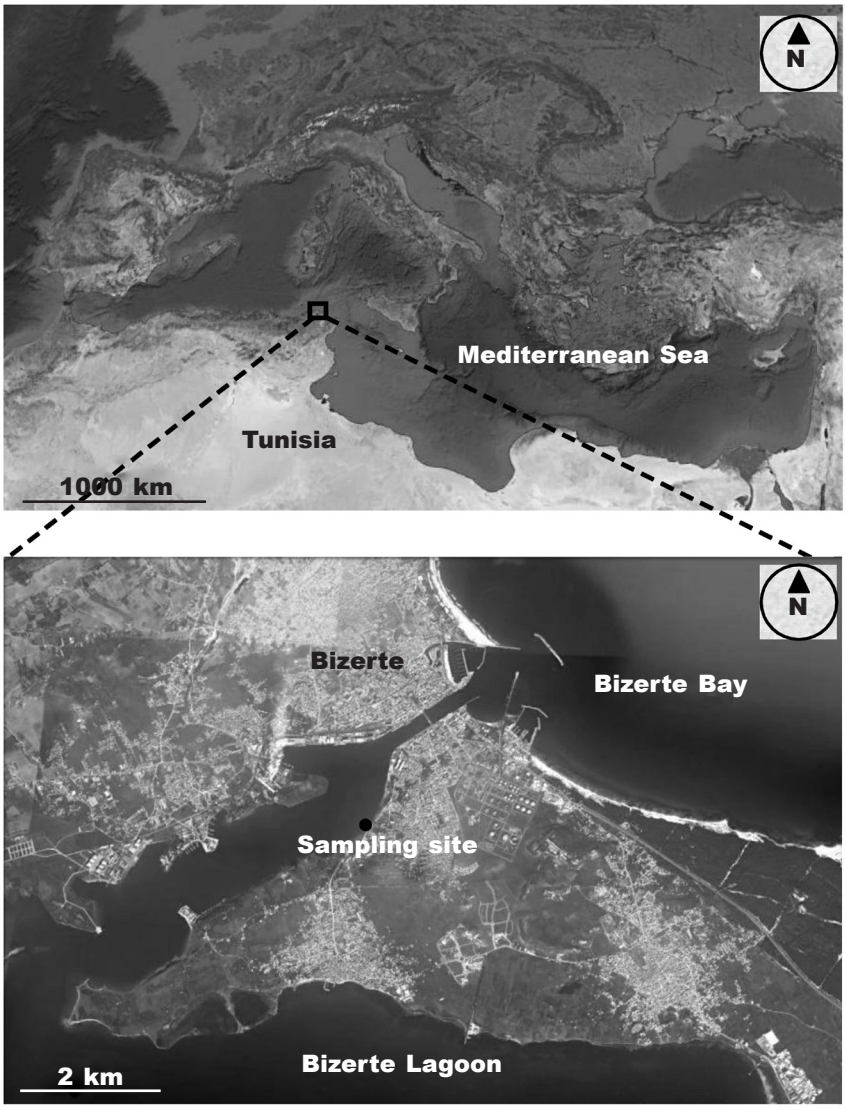

Fig. 1. Study area and location of sampling site.

Fig. 1. Zona de estudio y ubicación del sitio de muestreo.

Etymology
This species is named after the characteristic larger dots in the lateral differentiation on the lower 75% conical region of the tail which is visible even at 10x magnification, especially for males.

Measurements
See table 1.

Description
Males. Organism relatively large. Body cylindrical, narrowing towards the cervical region (75–85 µm from the anterior end) where a front portion slightly set off by constriction is noted (30–40%). Buccal cavity cylindrical (width 14–18 µm, height 15–20 µm) with prominent large dorsal tooth and two ventrosublateral teeth. Thick cylindrical pharynx, slightly wider at base. No distinct cardia. Nerve ring at 40–45% of pharyngeal length from anterior end. Lower half of the head region characterized by a cephalic ring (figs. 2A, 4B) with 10–12 µm wide and 6–9 µm height. The arrangement of sensorial organs is: six inner labial sensilla 1–2 µm, followed by six outer labial sensilla 7–8 µm and four shorter cephalic sensilla 3–5 µm; the latter two arranged in a circle 5–6 µm from the anterior end. Ten subcephalic setae 16–18 µm (six submedian and four lateral) in a circle at 12–16 µm from the anterior end. Amphids, 12–15 µm far from the anterior end, multispiral, with ~4.5 turns in ventral direction, and 7–13 µm wide (~40% of the cbd) by 7–8 µm long (fig. 2).

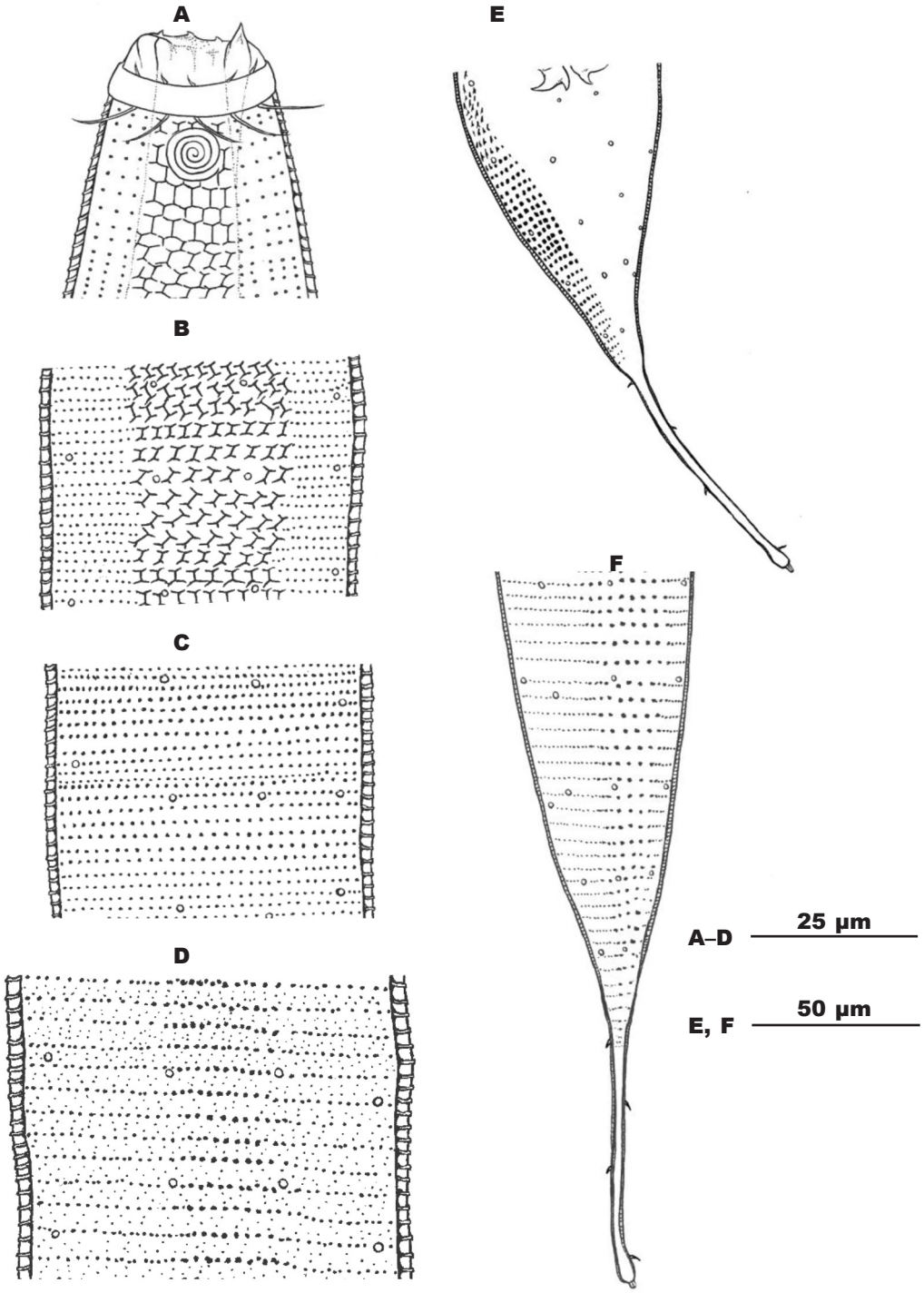

Fig. 2. *Marylynnia puncticaudata* n. sp.: A. Male head, showing the lateral differentiation in the cervical region; B–C. Variable cuticle punctuation in pharyngeal region; D. Modified punctuation and lateral differentiation in the middle of the body; E. Male tail and lateral differentiation; F. Female tail and lateral differentiation.

Fig. 2. Marylynnia puncticaudata *sp. n.: A. Cabeza del macho donde se aprecia la diferenciación lateral en la región cervical; B–C. Puntuación variable de la cutícula en la región faríngea; D. Puntuación modificada y diferenciación lateral en la zona central del cuerpo; E. Cola del macho y diferenciación lateral; F. Cola de la hembra y diferenciación lateral.*

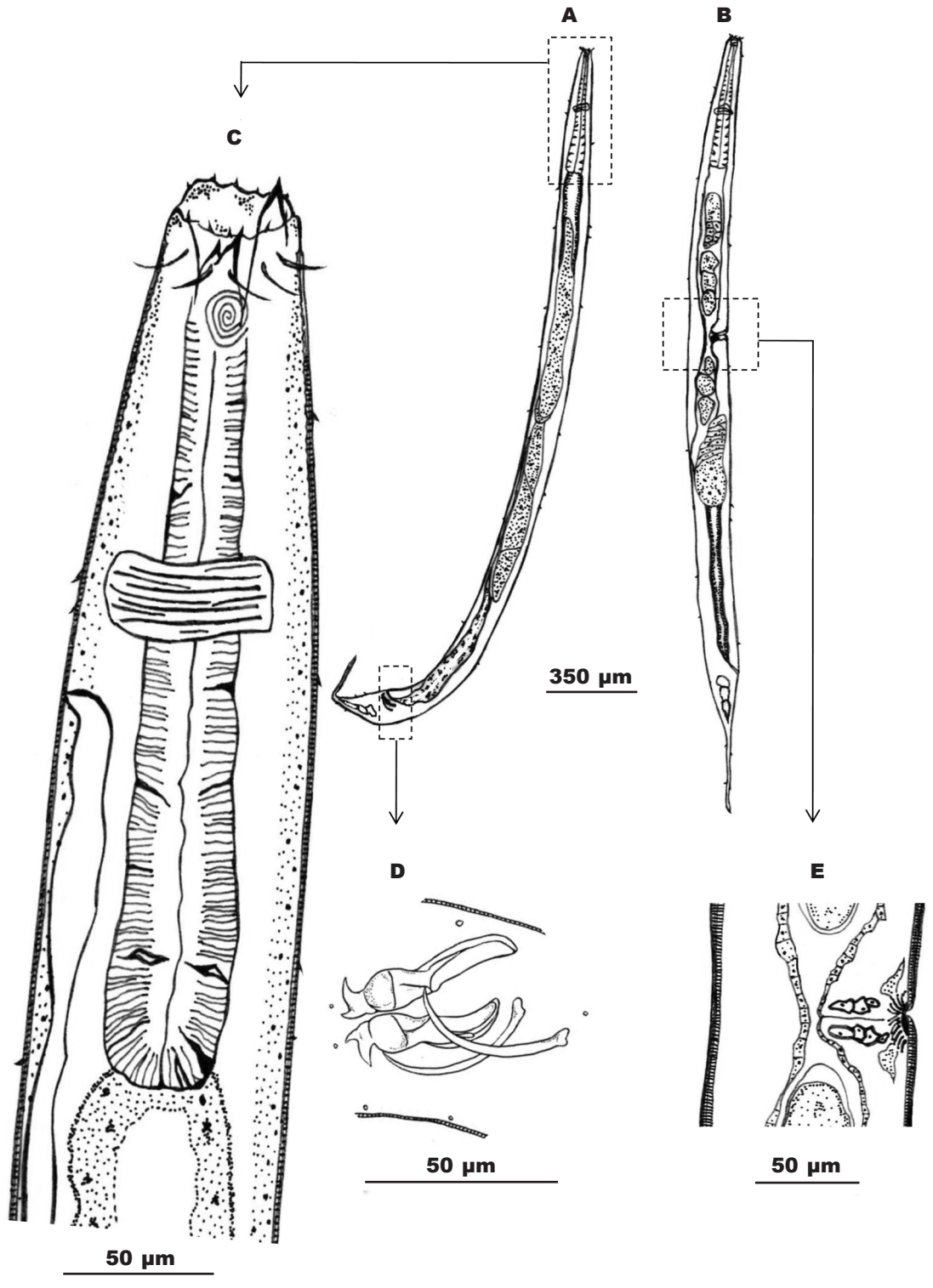

Fig. 3. *Marylynnia puncticaudata* n. sp.: A. Male whole body; B. Female whole body; C. Male head end; C. Male spicular apparatus; D. Female vulvar region.

Fig. 3. Marylynnia puncticaudata *sp. n.: A. Cuerpo entero del macho; B. Cuerpo entero de la hembra; C. Extremo cefálico del macho; D. Aparato espicular del macho; E. Región vulvar de la hembra.*

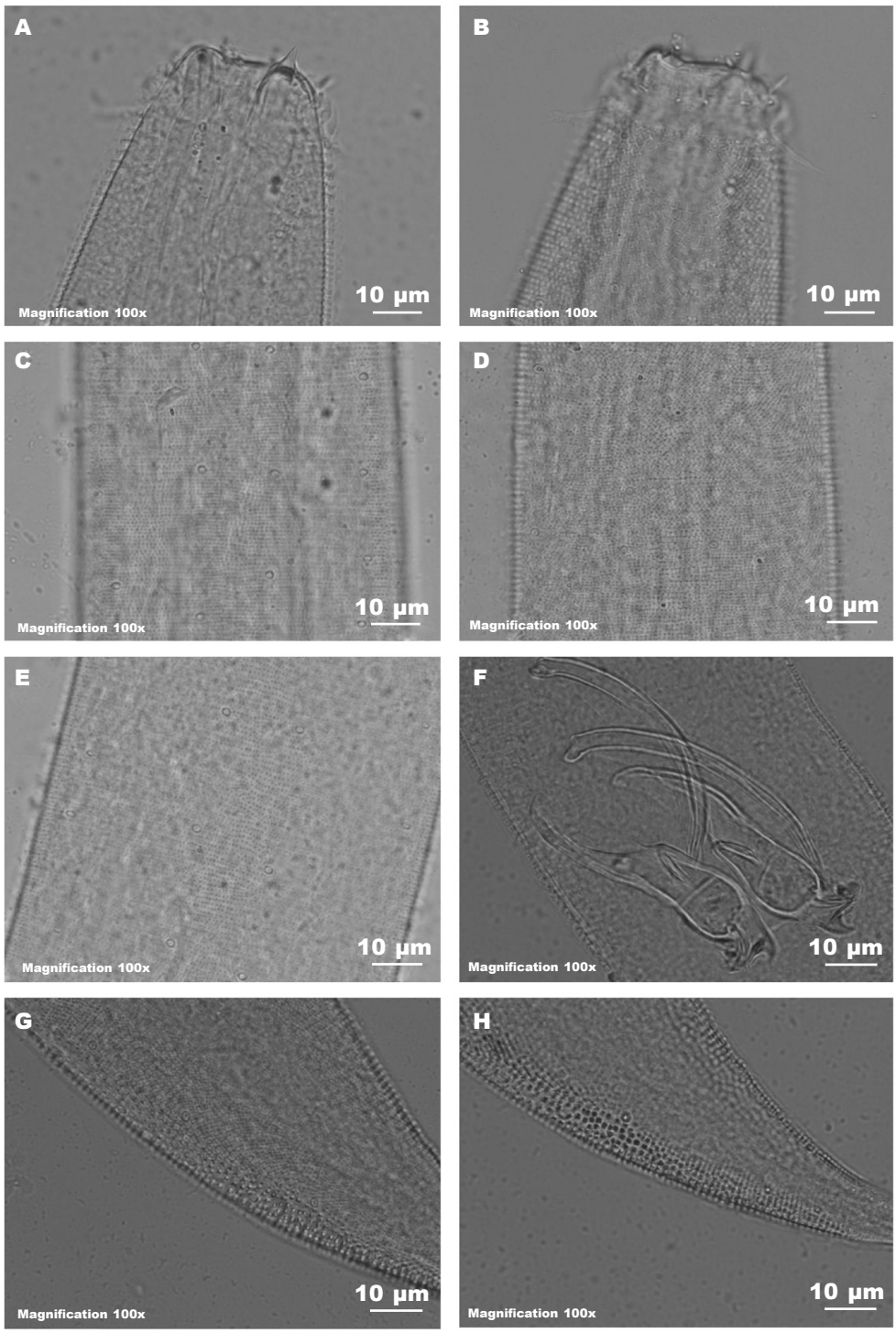

Fig. 4. *Marylynnia puncticaudata* n. sp. (holotype male): A. Head region, showing buccal cavity; B. Head region, showing cephalic ring and lateral differenciation of doublecrosses in cervical region; C–D. Variable lateral differentiation in pharyngeal region (fine dots to doublecrosses); E. Modified punctuation and lateral differentiation in the middle of the body; F. Copulatory armature (spicules and gubernacula); G–H. Caudal lateral differentiation.

Fig. 4. Marylynnia puncticaudata *sp. n. (holotipo macho): A. Región cefálica en la que se aprecia la cavidad bucal; B. Región cefálica en la que se aprecia el anillo cefálico y la diferenciación lateral en dobles cruces en la región cervical; C–D. Diferenciación lateral variable en la región faríngea (de puntos finos a cruces dobles); E. Puntuación modificada y diferenciación lateral en la zona central del cuerpo; F. Órgano copulador (espículas y gubernáculos); G–H. Diferenciación laterocaudal.*

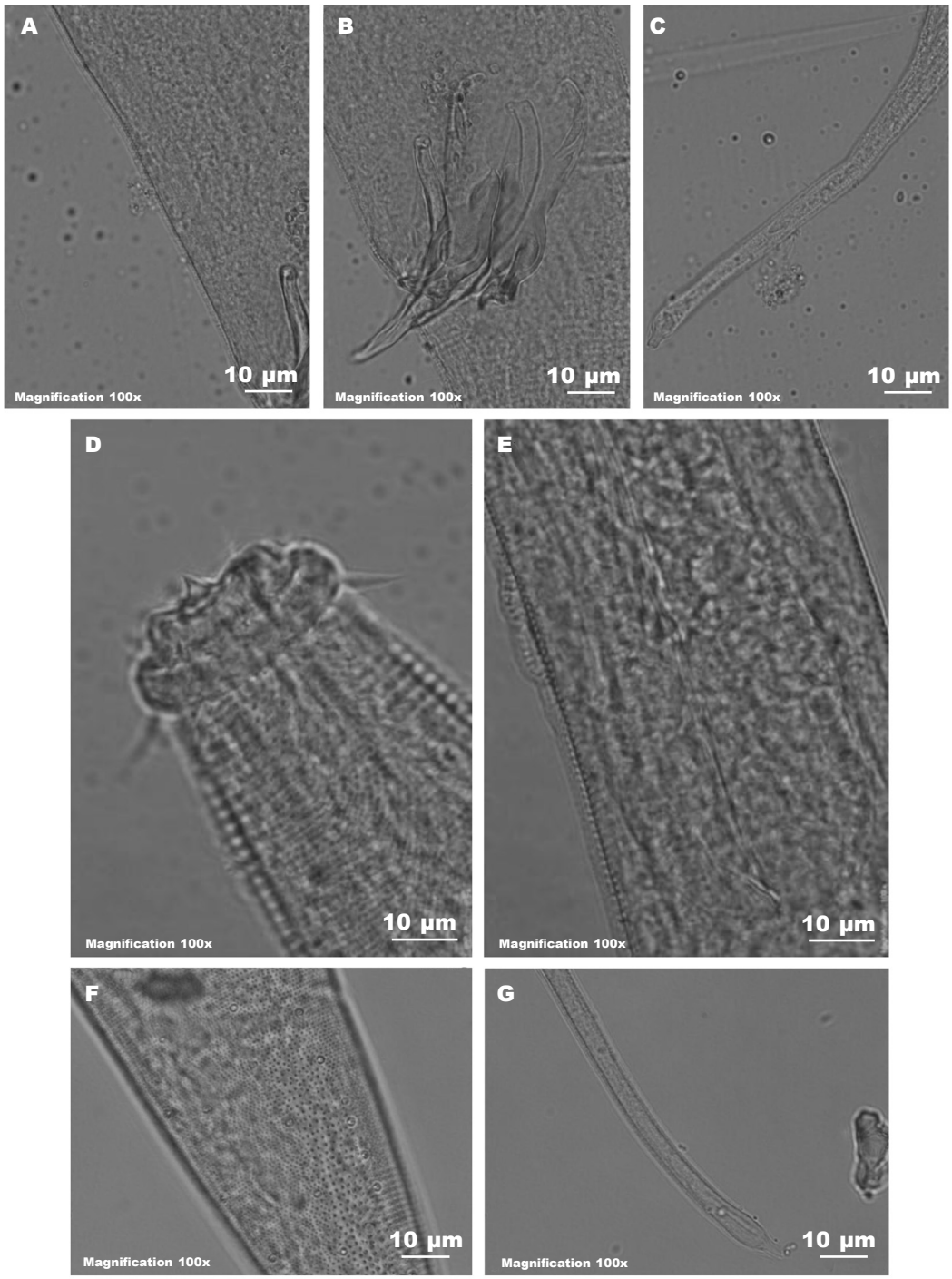

Fig. 5. *Marylynnia puncticaudata* n. sp. (paratypes). Male: A. Precloacal zone, showing the absence of supplements; B. Copulatory armature (spicules and gubernacula); C. Tail end. Female: D. Head region, showing the cephalic ring and the lateral differenciation in cervical region; E. Pharyngeal end; F. Caudal lateral differentiation; G. Tail end.

Fig. 5. Marylynnia puncticaudata sp. n. (paratipos). Macho: A. Zona precloacal en la que se aprecia la ausencia de suplementos; B. Órgano copulatorio (espículas y gubernáculos); C. Extremo caudal. Hembra: D. Región cefálica en la que se aprecia el anillo cefálico y la diferenciación lateral en la región cervical; E. Extremo faríngeo; F. Diferenciación laterocaudal; G. Extremo caudal.

Table 1. Measurements of *Marylynnia puncticaudata* n. sp. (μm except De Man ratios (a, b, c, c' and V%). The cbd of the pharynx was measured at its base. (For abbreviations see the text.)

Tabla 1. Mediciones de Marylynnia puncticaudata *sp. n. (μm excepto los índices De Man (a, b, c, c' y V%). El cbd de la faringe se midió en su base. (Para las abreviaturas ver el texto.)*

Measurements	Holotype ♂	Paratypes ♂ (*n* = 9)	Paratypes ♀ (*n* = 10)
Total body length	2,554	2,611 (2,503–2,657)	2,715 (2,568–2,851)
Maximum body diameter	81	72 (69–85)	70 (62–83)
Head diameter	35	30 (22–37)	31 (20–39)
Length of cephalic setae	4	4 (3–5)	3 (2–5)
Diameter of amphids	11	10 (7–13)	10 (7–12)
Nerve ring from the anterior end	149	151 (116–181)	149 (124–172)
Nerve ring cbd	51	52 (44–67)	45 (38–50)
Pharynx length	355	369 (325–419)	324 (279–363)
Pharynx cbd	74	65 (55–72)	63 (55–71)
Spicule length as arc	51	56 (48–61)	–
Length of gubernacula	65	70 (63–77)	–
abd	63	62 (59–65)	45 (37–53)
Tail length	217	262 (222–305)	250 (194–305)
Tail cylindrical portion (%)	38.24	41.26 (36.9–47.05)	47.08 (42.11–52.26)
c'	3.44	4.39 (4.04–4.70)	5.55 (4.95–6.74)
Vulva from anterior	–	–	1,693 (1,616–1,772)
Vulva cbd	–	–	70 (62–80)
V%	–	–	62.11 (54.02–70.65)
a	31.53	35.85 (29.82–40.03)	38.75 (31.01–46.73)
b	7.19	7.17 (5.10–9.24)	8.33 (5.62–10.97)
c	11.76	10.29 (7.47–12.98)	10.82 (8.37–13.06)

Immediately below the cephalic ring there begin four rows of cuticular hypodermal pores and a lateral differentiation with an average width of 27 μm (~50% cbd). These rows divide the entire body into lateral fields and extend from behind the amphideal fovea to the end of the conical portion of tail. The lateral differentiation of 45–50 μm width is only present along 70–85 μm (*i.e.* until the middle of the distance from the anterior end of the nerve ring). It consists of larger dots which change to a honeycomb–like structure when the fine focus is used. Honeycomb–like structures are made with separated double–crosses; this variable lateral differentiation seems to be the result of the presence of thickened crossings. The cervical lateral differentiation is replaced with fine punctuation (and double–crosses) following by horizontal and parallel lines (0.8–1 μm annules) right through the middle region of the body. Approximately in the middle of the body, the punctuation becomes different and horizontal rows of fine dots border irregular fields of smaller dots. This type of punctuation overlaps from time to time with the lateral differentiation made with bigger dots. The fat dots (and double–crosses) characteristic of the cervical region reappear just up to the cloaca and become very clear in the lower 75% portion of the conical part of the tail. LMPs are not observed.

Reproductive system diorchics. Spicules 0.8–0.95 abd along arc, ventrally curved, with two unequal cephalate proximal tips. Gubernaculum larger than spicule, strongly cuticularized and highly complex. It consists of two blades proximally and distally separated but ventrally juxtaposed at their median parts. The proximal parts curved and expanded at base by conical thickenings. Bidenticulated distally: one large and lateral (11–15 μm) and the other one curved inward (6–8 μm). Absent precloacal supplements. Cylindrical portion of tail generally folded over the conical portion. In the caudal position, setae located immediately posterior to cloacal aperture and three caudal glands. Spinneret present. Three small setae (2.5–3.5 μm) alternately present on cylindrical part of tail and separated by ~15 μm. One slightly

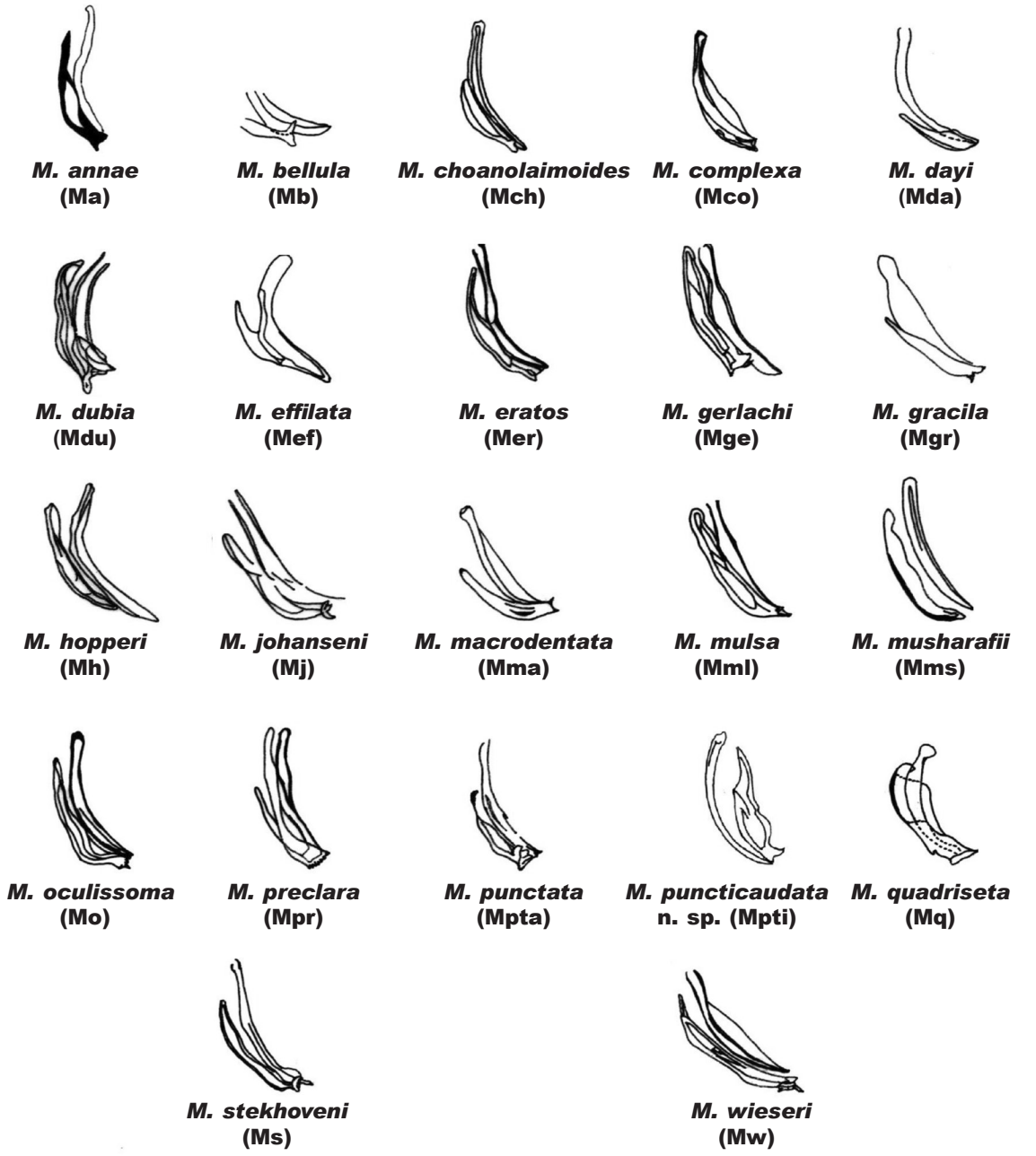

Fig. 6. Male genital armatures of the 21 reported *Marylynnia* species in comparison with that of *Marylynnia puncticaudata* n. sp. For the known species, the drawings have been modeled on those provided by the original descriptions.

Fig. 6. Órganos genitales masculinos de las 21 especies registradas de Marylynnia *en comparación con los de* Marylynnia puncticaudata *sp. n. Para las especies conocidas, las ilustraciones se han adaptado a partir de las proporcionadas por las descripciones originales.*

larger terminal setae (4–5 µm) present at the level of the terminal swollen tip and far from the other setae by ~25 µm.

Females. In most respects, the cuticle ornamentation is similar to the males. Lesser body maximum width with longer body and tail. Lower anal width, making the tail shape filiform. Caudal lateral differentiation with smaller and more spaced dots. No setae on the conical portion of the tail. Reproductive system didelphic, ovaries reflexed; anterior ovary situated subventrally to the right of the intestine, posterior ovary subventrally to the left of the intestine. Vagina well sclerotized.

Table 2. Comparative table of biometric and morphological data for holotype (or syntype) males of known species and *Marylynnia puncticaudata* n. sp.: L. Total body length; a, b, c. De Man's ratios; Sp. Spicule; abd. Anal body diameter; Gub. Gubernaculum; CHP. Row number of circular hypodermal pores; AT. Amphideal turns; CLD. Caudal lateral differentiation; PS. Number of precloacal supplements; CS. Number of cephalic setae; ? Not specified by authors. (For abbreviations of species see figure 6.)

Tabla 2. Tabla comparativa de los datos biométricos y morfológicos de los holotipos macho (o sintipo) de especies conocidas y de Marylynnia puncticaudata *sp. n.: L. Longitud total del cuerpo; a, b, c. Índices De Man; Sp. Espícula; abd. Diámetro corporal anal; Gub. Gubernáculo; CHP. Hilera inferior de poros hipodérmicos; AT. Vueltas de los anfidios; CLD. Diferenciación laterocaudal; PS. Número de suplementos precloacales; CS. Número de setas cefálicas; ? No especificado por los autores. (Para las abreviaturas de las especies, ver figura 6.)*

Species	L(µm)	a	b	c	Sp/abd	Gub/Sp(%)	CHP	AT	CLD	PS	CS
Ma	2,250	29.2	7.5	10.3	1.35	88.75	12	4.5	No	6	10
Mb	1,390	30.2	5.8	10	0.90–0.97	48.64–53.33	2	5.5–6.3	Yes	4	10
Mch	1,120	23.2	5.6	7	1.38	70	?	4.5	No	No	10
Mco	1,500–1,700	27.6–29	6.4–6.7	6.3–6.7	1.6–2.0	~50	2	6.5–7.5	Yes	6	10
Mda	3,100	31.0	4.8	12.4	0.79	58.64	?	3.75	No	No	10
Mdu	2,050	29	6	7.6	0.6	~90	?	4	No	5	?
Mef	1,452	24.2	6.6	8.8	1.8	~50	?	4.5	No	No	24
Mer	3,220	37.8	8.3	13.8	1.29	95.77	12	5	No	6	10
Mge	2,000	28	5.7	8.3	1.20	90.47	?	5	No	6	?
Mgr	1,386	44.7	6.7	8.2	1.36	76.47	8	5	No	6	8
Mh	2,140	31.5	7.3	10.2	2.01	73.86	8	~5	No	5	4
Mj	2,870	38	7.4	5.9	1.33	~80	8	4–5	No	5	10
Mma	1,620	40.6	6.5	10.8	1.41–1.6	87.5	?	4.3	No	7	8
Mml	1,770	23.9	7.0	8.11	1.30	104.68	10	4.25	No	6	10
Mmsi	1,400	59.1	7.4	9.8	1.41	88.23	4	9–10	No	5	4
Mo	1,980–2,240	35–37.3	9.1–9.2	7.98–8	1.29–1.31	96.22–101.69	8	4.5	No	6	10
Mpr	2,010–2,420	20.1–22.4	6.5–6.7	9.1–9.6	1.40–1.47	71.42–72.34	12	4	No	6	10
Mpta	1,653	29	6.2	8.4	1.53	66.66	4	~5.25	No	6	8
Mpti	2,554	31.5	7.2	11.7	0.8–0.95	116.66–120	4	4.5	Yes	No	4
Mq	1,210	31.8	5.8	6.6	1.3	90	?	5	No	7	?
Ms	2,464	47.2	15.3	8.55	1.5	89.33	?	4	Yes	3	10
Mw	3,200	34.8	7.0	10.3	1.14	96.87	?	5.25	No	10	10

Discussion

To date, 21 valid species of *Marylynnia* are known (Huang & Xu, 2013). The most distinctive characters of *M. puncticaudata* n. sp. are: (1) the cephalic ring followed by a circle of ten subcephalic setae, (2) the variable lateral differentiation from double crosses to dots when fine focus is used, (3) the modified cuticle punctuation along the body, (4) the complex and cuticularized gubernaculum with a unique shape in comparison with the remaining 21 known *Marylynnia* species (fig. 6), and (5) the characteristic larger dots in the lateral differentiation on the lower 75% conical region of the tail, especially for males. It is true that *Marylynnia puncticaudata* n. sp. resembles *M. stekhoveni*, *M. complexa* and *M. bellula* in having such lateral differentiation on conical portion of tail (table 2). Basically, *Marylynnia complexa* and *M. bellula* differ from *M. puncticaudata* n. sp., for their shorter body length (1,390–1,700 µm *vs.* 2,500–2,850 µm). When the new species is compared with *M. stekhoveni*, males of *M. puncticaudata*

Key to species of the genus *Marylynnia* Hopper, 1977.

Clave para las especies del género Marylynnia*, Hopper, 1977.*

1	Body length > 2,450 µm	2
	Body length < 2,450 µm	7
2	Male with 3–10 precloacal supplements	3
	Male without precloacal supplements	6
3	Tail 3.5–4.2 abd	4
	Tail ≥ 6.5 abd	5
4	Tail 3.5–4 abd, 10 precloacal supplements, amphid 5.25 turns	*M. wieseri* (Inglis) Hopper, 1972
	Tail 4.2 abd, 6 precloacal supplements, amphid 5 turns	*M. eratos* Hopper, 1972
5	Tail flagelliform 10.8 abd, 5 precloacal supplements, amphid 4–5 turns	*M. johanseni* Jensen, 1985
	Tail 6.5 abd, 3 precloacal supplements, amphid 4 turns	*M. stekhoveni* (Wieser) Hopper, 1972
6	Gubernaculum simple 50–60% spicule length, without caudal lateral differentiation	*M. dayi* (Inglis) Hopper, 1972
	Gubernaculum complex 110–120% spicule length, with caudal lateral differentiation	*M. puncticaudata* n. sp.
7	Tail with more than 60% of distal cylindrical portion (filiform)	*M. oculissoma* Hopper, 1972
	Tail with less than 50% of distal cylindrical portion	8
8	Male without precloacal supplements	9
	Male with 4–7 precloacal supplements	10
9	Gubernaculum < 60% spicule length, amphid diameter 35% cbd	*M. effilata* (Stekhoven) Hopper, 1972
	Gubernaculum 70% spicule length, amphid diameter > 50% cbd	*M. choanolaimoides* (Stekhoven) Hopper, 1972
10	Male with 4–5 precloacal supplements	11
	Male with 6–7 precloacal supplements	14
11	Male with 4 precloacal supplements, amphid 5.5 turns	*M. bellula* (Vitiello) Hopper, 1972
	Male with 5 precloacal supplements	12
12	Amphid diameter > 95% cbd	*M. musharafii* Nasira, Kamran & Shahina, 2007
	Amphid diameter < 55% cbd	13
13	Spicule 2.01 abd, gubernaculum distally pointed	*M. hopperi* Sharma & Vincx, 1982
	Spicule 0.6 abd, gubernaculum distally enlarged	*M. dubia* (Filipjev) Hopper, 1972
14	Male with 7 precloacal supplements	15
	Male with 6 precloacal supplements	16
15	Amphid 4.3 turns, amphid diameter 40% cbd, spicule 1.41–1.6 abd	*M. macrodentata* (Wieser) Hopper, 1972
	Amphid 5 turns, amphid diameter 50% cbd, spicule 1.3 abd	*M. quadriseta* (Wieser) Hopper, 1972
16	Body length ≥ 2,000 µm	17
	Body length < 2,000 µm	19
17	Gubernaculum with one dorsal–lateral tooth and two sub–ventral teeth	18
	Gubernaculum with distal extremity enlarged and heavily denticulated	*M. preclara* Hopper, 1972
18	Amphid diameter 55% cbd, equitably arranged preanal supplements, spicule 1.2 abd	*M. gerlachi* (Wieser *sensu* Gerlach) Hopper, 1972
	Amphid diameter 37.5% cbd, 2+4 arranged preanal supplements, spicule 1.35 abd	*M. annae* (Wieser & Hopper) Hopper, 1972
19	Spicule with alae (wing–like extension)	20
	Spicule without alae	21
20	Inner labial sensillae papillose, 1 µm long	*M. complexa* (Warwick) Hopper, 1972
	Inner labial sensillae setose, 3 µm long	*M. punctata* Jensen, 1985
21	Gubernaculum longer than spicules, distal end with 3 teeth	*M. mulsa* Hopper, 1972
	Gubernaculum shorter than spicules, distally forked	*M. gracila* Huang & Xu, 2013

n. sp. are distinguished by numerous biometric and morphological characteristics: (1) very clear caudal lateral differentiation, (2) lower De Man's ratios, (3) modified punctuations along the body, (4) higher amphideal turns (4.5 *vs*. 4), (5) absence of precloacal supplements, (6) lower number of cephalic setae (4 *vs.* 10), (7) different shapes of gubernacula and spicules, (8) larger gubernaculum compared with spicule, (9) absence of post–cloacal setae, and (10) presence of 4 regular rows of hypodermal pores.

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