

# SEM Observations on the Marine Nematode *Dracognomus simplex* (Gerlach, 1954) Allen & Noffsinger, 1978 (Draconematidae: Prochaetosomatinae)

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**Abstract:** The free-living marine nematode *Dracognomus simplex* (Gerlach, 1954) Allen & Noffsinger, 1978 was studied by scanning electron microscopy (SEM). The morphology of males and females is described and illustrated in detail. In addition to the typical and modified adhesion tubes, a new type of posterior adhesion tube was discovered. A neotype is proposed for *Dracognomus simplex*, and *D. simplex sensu* Decraemer & Goubault, 1986 is renamed as *Dracognomus americanum* n. sp. Additionally, a key toward the *Dracognomus* species is proposed.

**Key words:** *Dracognomus americanum* n. sp., *Dracognomus simplex*, Draconematidae, identification, key, marine nematode, morphology, new species. Prochaetosomatinae, SEM observation, taxonomy.

The marine nematode family Draconematidae Filipjev, 1918 is a relatively small family, subdivided into two subfamilies and 14 genera, and includes at present 70 nominal species. Members of the family Draconematidae are, despite their small body size, relatively easy to recognize at a low magnification because of their typical draconematid habitus. They are distributed worldwide and occur from the intertidal exposed sands to deep-sea hydrothermal vents. In general, they are common in algal habitats (Allen and Noffsinger, 1978; Decraemer et al., 1997).

*Dracognomus simplex* was described by Gerlach (1954) from sandy beach interstitial water near Banyuls-sur-Mer (one female) and Cannes (two juveniles) in France as *Drepanonema simplex*. Unfortunately, no type material was deposited. In addition, Allen and Noffsinger (1978) redescribed 10 *D. simplex* females and one fourth-stage juvenile from subterranean water along the Mediterranean shore (France and Italy) and transferred *D. simplex* to a new genus: *Dracognomus*. No type material was reinstated or any slides deposited. Decraemer and Goubault (1986) collected and described in detail males, females, and all four juvenile stages of *D. simplex* from sandy beach interstitial water from Guadeloupe and Martinique.

At present, the genus *Dracognomus* includes six species. Only *D. annae* Verschelde and Vincx, 1993 has been described by scanning electron microscope (SEM). In 1999, we collected bottom debris and sand from the Mediterranean Sea and detected high numbers of *Epsilonema pustulatum* (Gerlach, 1952) Lorenzen, 1973 and *Dracognomus simplex* (Karssen et al., 2000). These samples were used for emended observations on adult *D. simplex* by SEM. We compared this and

other available *D. simplex* material by light microscope with deposited type material of all described *Dracognomus* species and prepared an identification key.

## MATERIALS AND METHODS

Sediment and bottom debris samples were collected in June 1999 near the water-line from the sand beach of Malaga (Spain), and nematodes were isolated as described by Lorenzen (1992). Thirty male and female specimens were fixed in 4% formaldehyde at 70 °C and after the rapid glycerine-ethanol method (Seinhorst, 1959) mounted in glycerine on Cobb slides. These slides were used for light microscopical observations and deposited afterward.

For SEM observations, 50 male and female nematodes were fixed in 3% glutaraldehyde, buffered with 0.05 M phosphate buffer (pH 6.8), and post-fixed with 2% osmium tetroxide for 2 hours at 20 °C. The specimens were dehydrated in a seven-graded series of ethanol and critical-point dried with carbon dioxide (Wergin, 1981), sputter coated with 10 nm platinum in a dedicated preparation chamber (CT 1500 HT, Oxford Instruments, High Wycombe, UK), and examined with a field emission electron microscope (Jeol 6300 F) at 3.5 kV.

Additionally, type material of the following *Dracognomus* species was studied: *D. annae* Verschelde and Vincx, 1993, *D. dermatoglyphus* Verschelde and Vincx, 1993, *D. marioni* (Allen and Noffsinger, 1978) Decraemer and Goubault, 1986, *D. notohalensis* Allen and Noffsinger, 1978, *D. tinae* Jensen, 1981, and voucher specimens of *D. simplex* were used for the preparation of a dichotomous key.

## SYSTEMATICS

Family Draconematidae Filipjev, 1918

*Dracognomus simplex* (Gerlach, 1954) Allen & Noffsinger, 1978

Syn. *Drepanonema simplex* Gerlach, 1954  
(Figs. 1–6)

### Description (emended)

*Females* ( $N = 25$ ): Length = 310–365 µm; diameter of body: at level of rostrum base = 23–25 µm; at level of

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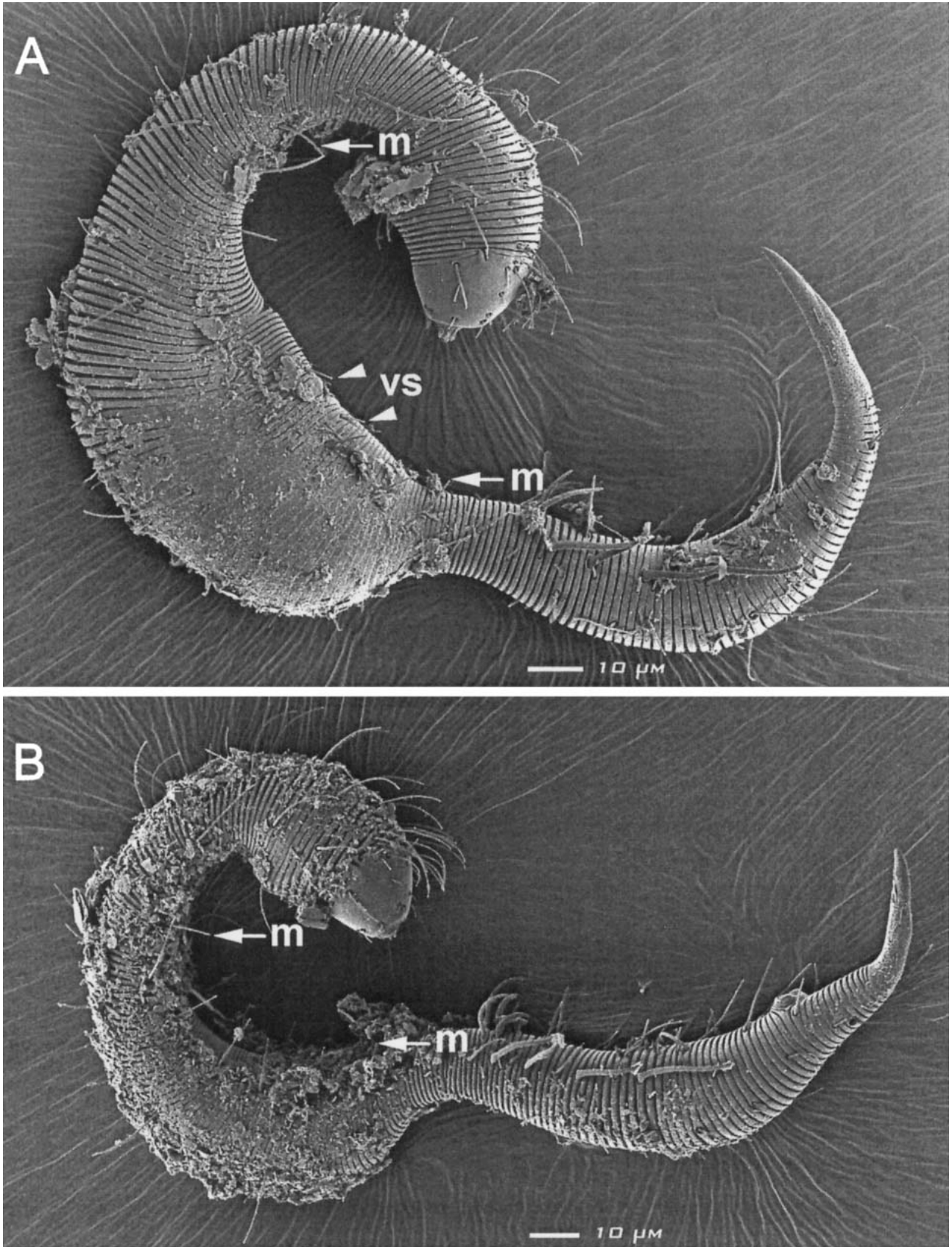


FIG. 1. *Dracognomus simplex*. A) Female habitus. B) Male habitus. m = first and last modified adhesion tube, vs = vulval setae.

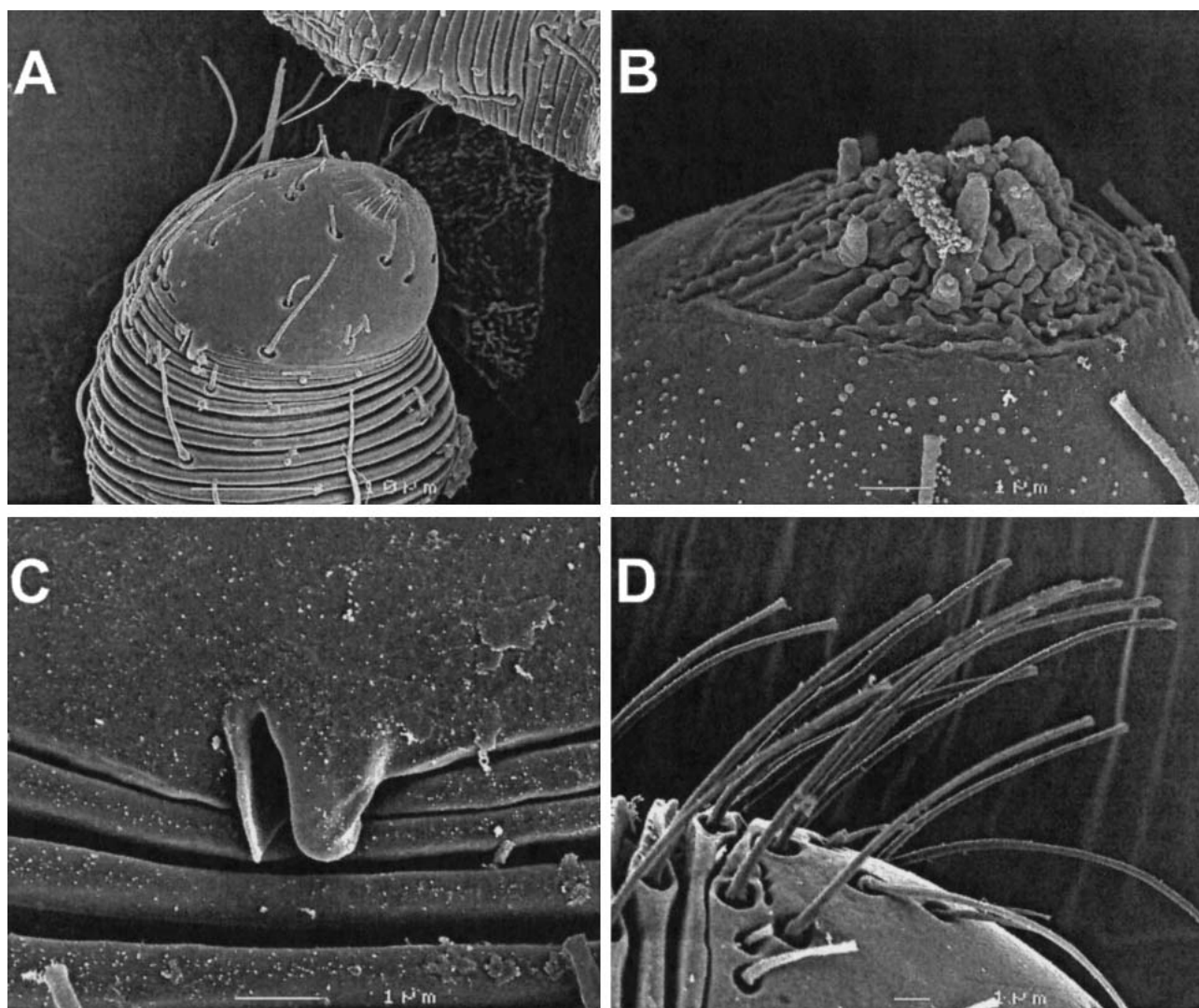


FIG. 2. *Dracognomus simplex* female. A) Head (lateral). B) Lip region. C) Amphid. D) Cephalic adhesion tubes.

nerve ring = 29–31  $\mu\text{m}$ ; at level of posterior end of pharynx = 17–21  $\mu\text{m}$ ; at level of vulva = 38–44  $\mu\text{m}$ ; at level of posterior end of reproductive system (prs) = 14–16  $\mu\text{m}$ ; at level between prs and anal opening = 20–33  $\mu\text{m}$ ; and at level of anal opening = 11–15  $\mu\text{m}$ ; tail length = 46–53  $\mu\text{m}$ ; non-annulated tail part length = 28–32  $\mu\text{m}$ . Body nearly epsilon-shaped, pharyngeal-, vulval-, and pre-anal region swollen. Except for head and tail end, body clearly annulated; annules smooth, 180–190 in number and about 1  $\mu\text{m}$  wide and high (Figs. 1,3).

Helmet smooth, asymmetric, ventral side shorter (Fig. 2A). Lip region protruding or non-protruding in fixed specimen (Figs. 2B,5A). Oral aperture rounded and small (<1  $\mu\text{m}$  in diam.), embraced by six small fused lips, surrounded by a small circular rim. Labial sensilla papilliform, arranged in two crowns of 6 inner and 6 outer labial sensilla, respectively, 1 and 0.5  $\mu\text{m}$  long (Figs. 2B,5B). Four cephalic setae present on helmet, 3.5  $\mu\text{m}$  long, enclosed by a circle of 8 subcephalic setae, 2.5  $\mu\text{m}$  long; 4 subcephalic setae more posterior

at dorsal and lateral helmet side, 2.5–3  $\mu\text{m}$  in length; one long seta, 9  $\mu\text{m}$ , at lateral helmet-base (Fig. 2A). Amphids small, 3  $\mu\text{m}$  long and 2  $\mu\text{m}$  wide, lateral to dorso-lateral at helmet-base and extending into first two body annules; fovea a longitudinal slit (amphid inverted 'U'-shaped with the light microscope) nearly 2  $\mu\text{m}$  long (Fig. 2C).

Twelve fine modified cephalic adhesion tubes (CAT), paired, partly dorsal and subdorsal on helmet edge extending till first four annules; tubes without swollen base of insertion and with slightly swollen open tip, 11–14  $\mu\text{m}$  long (Fig. 2D). Two sublateral rows of 9 fine modified adhesion tubes (Mod-AT), posterior to pharyngeal level extending posteriorly near sublateral adhesion tubes (Fig. 1A); first three pairs 10–14  $\mu\text{m}$  long, other pairs 6–8  $\mu\text{m}$  long; tubes with slightly swollen base and narrowing open tip.

Somatic setae arranged in the pharyngeal region with 2 subdorsal (12–17  $\mu\text{m}$  in length), 2 long (9–21  $\mu\text{m}$ ), and 2 small (<2.5  $\mu\text{m}$ ) sublateral and 2 subventral rows (first pair about 13  $\mu\text{m}$ , others <2.5  $\mu\text{m}$  long); mid-body

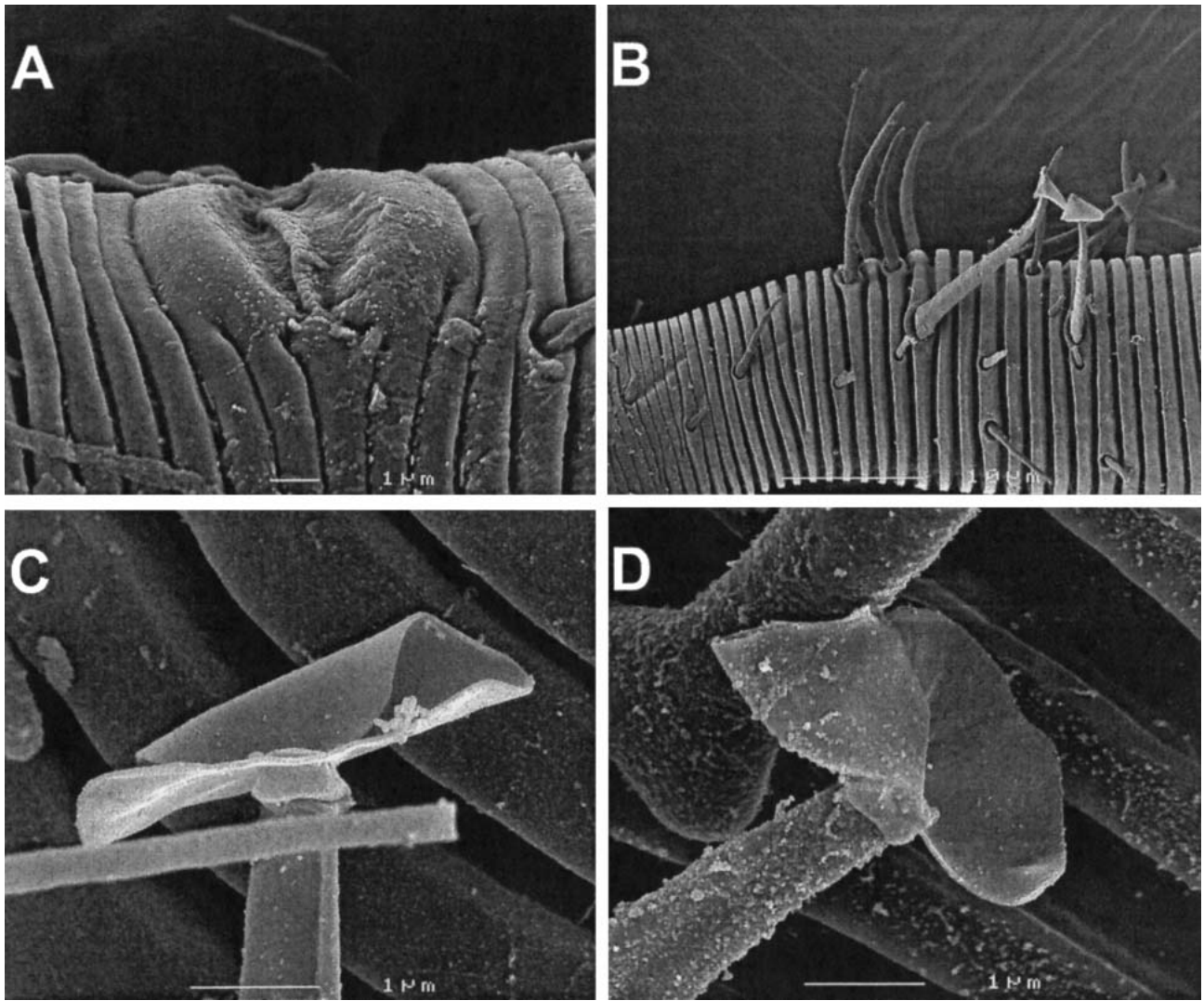


FIG. 3. *Dracognomus simplex* female. A) Vulva. B) Anterior part: posterior adhesion tubes. C–D) Disc-shaped sublateral PAT tips.

region with 2 subdorsal (4–8  $\mu\text{m}$  in length), 2 sublateral (4 pairs 16–21  $\mu\text{m}$ , others <2.5  $\mu\text{m}$  long), and 2 subventral rows (<2.5  $\mu\text{m}$  long); posterior to body constriction with 2 subdorsal (<2.5  $\mu\text{m}$  in length), 2 sublateral (4 pairs increasing in length toward punctured tail part, 9–29  $\mu\text{m}$ , rest <2.5  $\mu\text{m}$ ), and 2 subventral rows (<2.5  $\mu\text{m}$  long); a single ventral setae (3–4  $\mu\text{m}$  in length) present anterior near anal tube (Fig. 4A); punctured tail part with 2 sublateral pairs (2–4  $\mu\text{m}$  long). All somatic setae straight and open, without swollen base and narrowing tip (i.e., tips are slightly widening).

Small vulva, about 4- $\mu\text{m}$  width, located at mid-body (45–49%) with smooth vulval lips and flap; distal end of vagina vera with two large sclerotizations; 2 subventral pairs of vulval setae (4–5  $\mu\text{m}$  long) present (Fig. 3A). Posterior adhesion tubes (PAT), between body constriction and anus, arranged in 2 subventral and sublateral rows (Fig. 1). Anteriorly with one group of 4 well-developed subventral PAT, slightly bending posteriorly and gradually tapering toward a narrowing open tip;

first pair 14–15  $\mu\text{m}$  long, second pair 10–11  $\mu\text{m}$  long (Fig. 3B). Additionally, 5 pairs of modified subventral PAT present, 4 to 5 annules posterior (anterior) subventral PAT group and anterior of anal tube; pairs posteriorly increasing in length (10 to 16  $\mu\text{m}$ ). Tubes straight with narrowing open tip and slightly swollen base (Fig. 4A). Posterior near level of anterior subventral PAT, 2 sublateral PAT rows each with robust, straight, and long tubes, posteriorly increasing in length (17 to 22  $\mu\text{m}$ ). Tubes gradually tapering toward an open tip; tip provided with unusual flexible, thin, asymmetrical disc (palette-shaped), often flexed backward or forward, forming a disc-like structure; maximum diam. 3.5  $\mu\text{m}$  (Figs. 3B–D, 5C, D, and 6A).

Anus provided with non-annulated tube, 2–3  $\mu\text{m}$  in length (Fig. 4A). Tail bending ventrally, non-annulated tail part equally punctured, posterior tail part smooth, 14–18% of total tail length (Fig. 4C). Tail tip provided with one caudal-gland opening, approx. 200 nm in diam. (Fig. 4D).

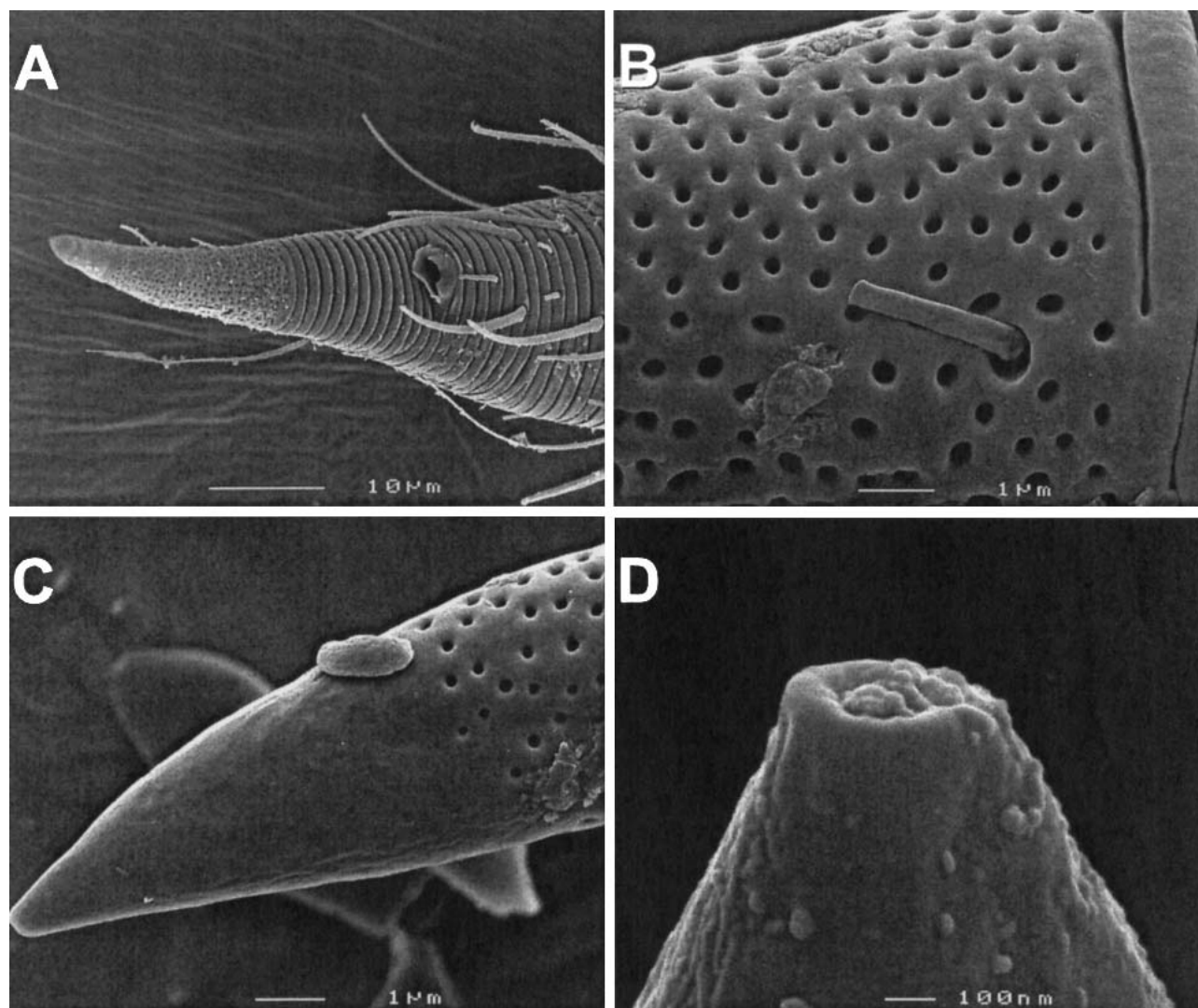


FIG. 4. *Dracognomus simplex* female. A) Tail region, ventral view. B) Tail punctures. C) Smooth tail part. D) Tail tip.

*Males* ( $N = 25$ ): Length = 330–380  $\mu\text{m}$ ; mid-body region diam. = 32–34  $\mu\text{m}$ ; posterior body region diam. = 23–27  $\mu\text{m}$ ; spicules length = 29–32  $\mu\text{m}$ ; gubernaculum length = 12–14  $\mu\text{m}$ ; tail length = 52–60  $\mu\text{m}$ ; non-annulated tail part length = 29–31  $\mu\text{m}$ . General characteristics similar to females (Fig. 1B) but differs from them in a less swollen mid-body region and a more swollen posterior body between sublateral PAT and anus. Two sublateral rows of 7 Mod-AT, first three pairs 10–15  $\mu\text{m}$  long, other pairs becoming shorter posteriorly (last pair 8–10  $\mu\text{m}$ ). Posterior adhesion tubes, anteriorly with four well-developed subventral tubes; first pair strongly curving posteriorly, about 14  $\mu\text{m}$  long; second pair more robust, less curved, and about 9  $\mu\text{m}$  long (Fig. 5C).

One copulatory thorn present ventrally, immediately behind first two pairs of PAT; additionally provided with a group of 10 copulatory thorns (Fig. 6A), between first pairs of PAT and cloaca; thorns about 2.5  $\mu\text{m}$  in length, cuticularized, thin, and twisted (Fig. 6B).

Four pairs of modified subventral PAT, first pair between anterior single thorn and thorn-group, second pair near thorn-group level, last pairs situated between thorn-group and cloaca; first two pairs 10–12  $\mu\text{m}$  long, last two pairs 15–17  $\mu\text{m}$  long. A single pair of subventral cloacal setae behind cloaca, about 8  $\mu\text{m}$  in length (Fig. 6C,D). Sublateral PAT with 4 long robust tubes as in females and provided with the same disc-like structure, posteriorly increasing in length (15–23  $\mu\text{m}$ ); all sublateral PAT alternate with somatic setae, first pair (i.e., anterior to first sublateral PAT pair) 10–11  $\mu\text{m}$  long; setae posteriorly increasing in length (till about 15  $\mu\text{m}$ ) toward fourth setae pair (i.e., anterior to fourth sublateral PAT pair); a dubbel somatic setae pair present, 9–10  $\mu\text{m}$  long, posterior to last sublateral PAT pair (Figs. 1B,5C,D).

Spicules arcuate, proximal rounded cephalizations with small ventral projection, ventral alae well developed. Gubernaculum straight and parallel to distal part of spicule. Cloaca opening provided with two trans-

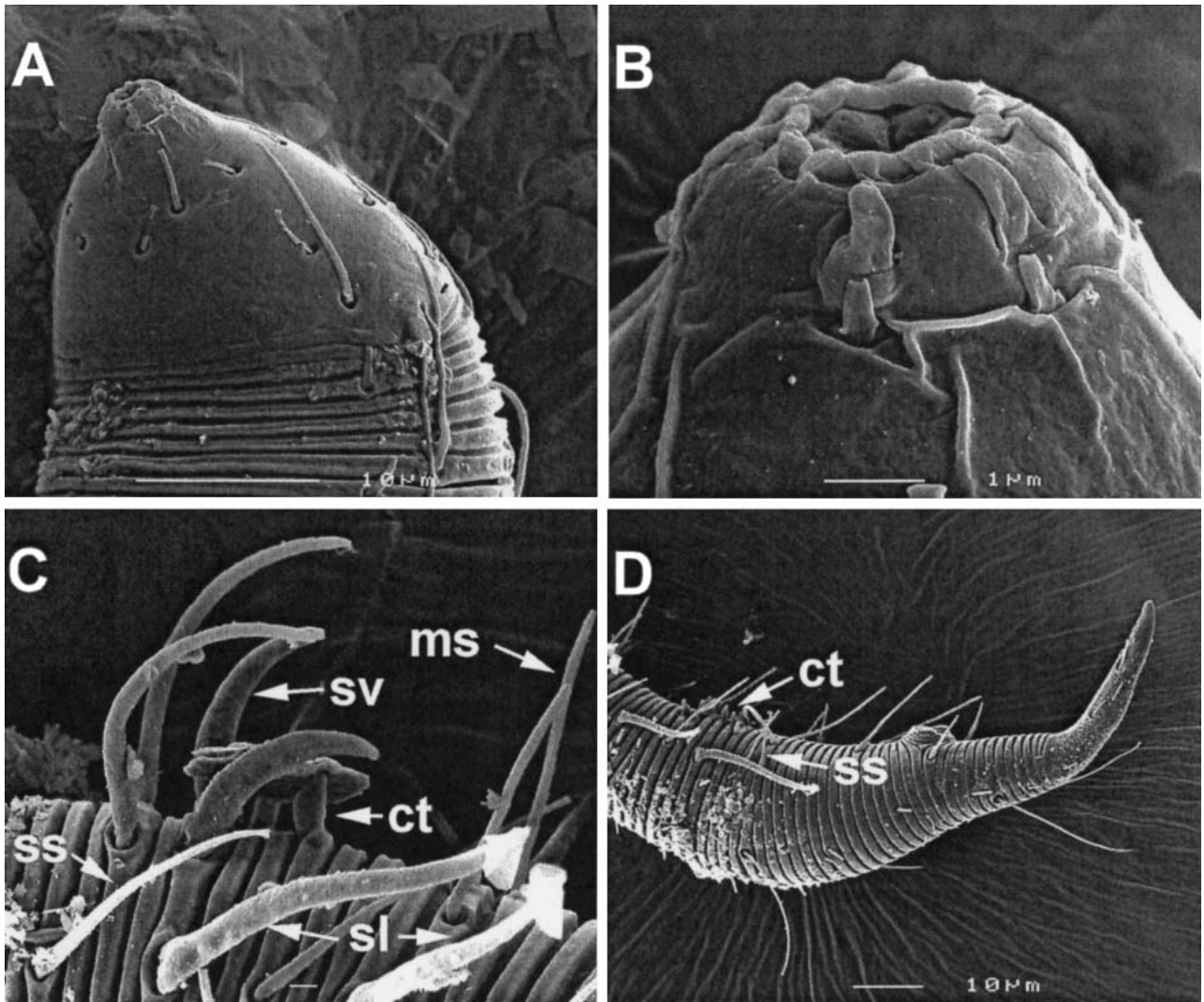


FIG. 5. *Dracognomus simplex* male. A) Head. B) Lips region. C) Anterior part: posterior adhesion tubes. D) Posterior body, lateral view. sv = subventral posterior adhesion tube, sl = sublateral PAT, ms = modified subventral PAY, ss = somatic setae, ct = copulatory thorn.

versely annulated (3–4 annules) flaps (appears as a protuberance in lateral position), 2–4  $\mu\text{m}$  high (Fig. 6C,D). Non-annulated tail part bending ventrally, equally punctured, except for ventral side and posterior tail part; 3 subventral pairs of somatic setae present on non-annulated tail part, 3–5  $\mu\text{m}$  in length (Fig. 6C). Non-annulated tail part devoid of cuticularized protuberances.

*Distribution and habitat*

Mediterranean Sea: Banyuls-sur-Mer and Cannes, France (Gerlach, 1954), and Malaga, Spain (present study). Ligurian Sea: Tirrenia and San Rossore, Italy (Allen and Noffsinger, 1978; Raski, 1962 unpub.; Gheskiere, pers. comm.). Celtic Sea: Isles of Scilly, UK (Platt and Warwick, 1988). All specimens collected from sand or bottom debris from the intertidal zone at coarse sand beaches.

*Specimens examined*

Two females, collected from the Ligurian Sea near Tirrenia, Italy (July, 1962, coll. D. J. Raski). Slide nos. GC 91 3c and 3e, deposited in the Department of Nematology, University of California, Davis. Three males and two females, collected from the Ligurian Sea near San Rossore, Italy (July, 2001, coll. T. Gheskiere). Slides nos. BP IV meioB 6 and BP IV A 9, deposited in the Biology Department, University of Gent, Gent, Belgium. Nineteen males, 17 females, and 5 juveniles, collected from the Costa del Sol near Malaga, Spain (June, 1999, coll. G. de Deyn & M. van Keelen). Slides deposited in the Department of Nematology, University of California, Davis; Department of Invertebrates, Koninklijk Belgisch Instituut voor Natuurwetenschappen, Brussels, Belgium, and Department of Nematology, Wageningen University, Wageningen, The Netherlands.

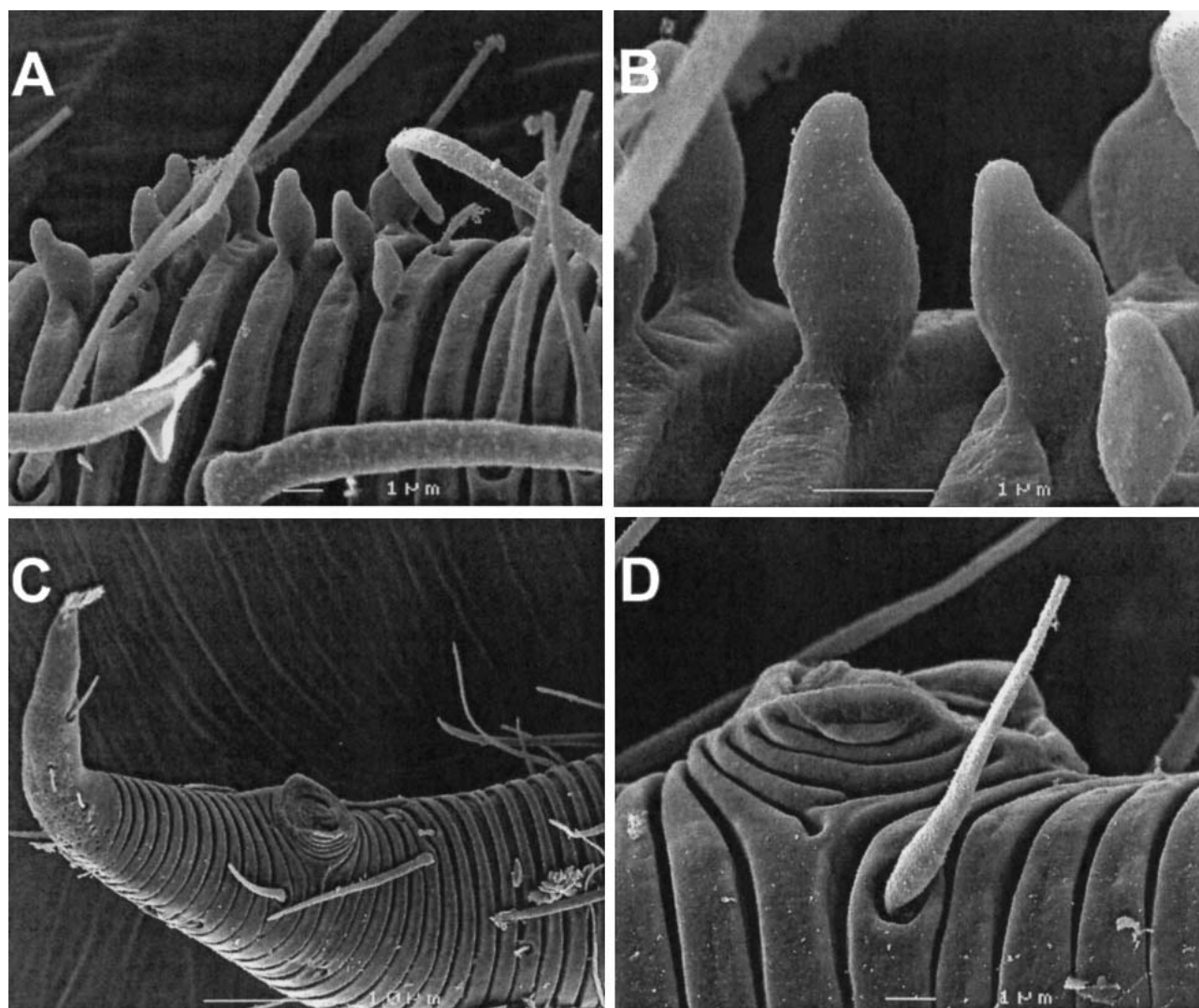


FIG. 6. *Dracognomus simplex* male. A) Pre-cloacal copulatory thorns. B) Idem, detail. C) Tail region, lateroventral view. D) Cloacal flaps.

### Diagnosis

Twelve fine modified cephalic adhesion tubes present, most tubes just posterior to helmet. Amphids small, 3 µm long, extending into first two body annules. Two sublateral rows of 7 to 9 modified adhesion tubes between pharyngeal region and posterior adhesion tubes. No posterior adhesion tubes (PAT) present posterior to anus/cloaca. All subventral PAT modified, first two pairs robust and bending posteriorly in males. Four pairs of robust sublateral PAT with thin disc-like structure on tip, in males intermingled with somatic setae. Male with one copulatory thorn, immediately behind first two pairs of subventral PAT, additionally a group of 8 to 10 thorns present between first single thorn and anus. Cloaca provided with prominent transversely annulated (3–4 annules) flaps. Non-annulated tail part in males without cuticularized protuberances. Ventral spicules alae well developed.

### Remarks

The studied *D. simplex* population from Malaga (Spain) is in agreement with the original *D. simplex* description (Gerlach, 1954) and redescription by Allen & Noffsinger (1978) and also with other available material collected from the Mediterranean Sea (i.e., Tirrenia and San Rossore, Italy). The reported CAT number by Gerlach (1954) is 6, while Allen & Noffsinger (1978) and Decreamer et al. (1997) reported 8 CAT for the genus *Dracognomus*. We observed 12 CAT with the SEM and noticed that the exact number is rather difficult to count with the light microscope, due to the very fine structure of these tubes and the curved lateral body position in *D. simplex*. A higher CAT number is not impossible for the genus *Dracognomus*, as it was also described (10–12 very thin CAT) for *D. dermatoglyphus* Verschelde and Vincx, 1993. Also, the SEM observed disc-like structure on the sublateral PAT tip needs a

keen observation with the light microscope. Particularly if one knows that this tiny structure is often in a reflexed backward position within permanent slides.

No type material was deposited by Gerlach (1954) nor by Allen and Noffsinger (1978) for *D. simplex* (i.e., no holotype or syntype material is available). The original type locality is Banyuls-sur-Mer or Cannes, France (Gerlach, 1954). To our knowledge, the oldest available *D. simplex* material (2 females) was collected near Tirrenia by Dr. D. J. Raski in 1962, i.e., not far from the original type locality area (see also Allen & Noffsinger, 1978 p. 118; note: Tirrenia was reported as Terrenia). Based on the observed uniformity of the studied Mediterranean *D. simplex* material and the lack of any types, we propose slide GC 91 3e (1 female), deposited in the Department of Nematology, University of California, Davis, as a neotype.

The thorough emended description of *D. simplex sensu* Decreamer & Gourbault (1986) was based on material collected near Guadeloupe, Martinique, and its satellite islands. This Caribbean material differs from the Mediterranean *D. simplex* in number of CAT, number and position of copulatory thorns, number and composition of PAT, the lack of any disc-like structure on sublateral PAT tip, strongly reduced cloacal flaps and ventral spicules alae development. Based on these differences and the wide geographical separation, we consider the Caribbean *D. simplex* as a new species and rename it herein as *Dracognomus americanum* n. sp.

We have not observed *D. simplex* material from the Celtic Sea in the British Isles, as reported by Platt and Warwick (1988), and consider this report as conspecific with the Mediterranean material. An additional study, however, is needed to prove if this is correct.

*Dracognomus americanum* n. sp.

Syn. *Dracognomus simplex* (Gerlach, 1954). *sensu* Decreamer & Gourbault, 1986

#### Description

*Dracognomus simplex*: Decreamer & Gourbault (1986): 112–115, Figs. 4–6.

#### Type locality and habitat

Anse de la Gourde, Grande Terre, Guadeloupe. Marine, in coarse, calcareous sandy beach interstitial water. Also detected on the following localities: Anse Laborde, Grande Terre and Anse Dupuy, Basse Terre, Guadeloupe and at Anse Petite Rivière, La Désirade; Plage de Pompierre and Grosse Pointe, Iles des Saintes; Anse l'Etang, Martinique.

#### Type specimens

Holotype male, collected on 4 August 1982, slide RI 62 and paratypes, slides RI 59–85 (41 males, 36 females, and 45 juveniles), deposited in Department of Invertebrates, Koninklijk Belgisch Instituut voor Natuurwet-

schappen, Brussels, Belgium, and paratypes, slides AN 466–470, 474, 479–482 (8 males, 12 females, and 5 juveniles), deposited in Muséum National d'Histoire Naturelle, Paris, France.

#### Diagnosis

Eight modified cephalic adhesion tubes present, just posterior to helmet. Amphids small, 4–5 µm long, extending into first two body annules. Two sublateral rows of 7 modified adhesion tubes between pharyngeal region and posterior adhesion tubes. No posterior adhesion tubes (PAT) present posterior to anus/cloaca. All subventral PAT without differentiated tip, first 2–3 pairs slightly bending in males, other 3–4 pairs less robust. Four pairs of sublateral PAT, without differentiated tip, in males intermingled with somatic setae. Male with 4 pairs of copulatory thorns, between first pairs of subventral PAT and cloaca, additionally a single thorn present in front of cloacal opening. Cloaca provided with two small flaps, including a single transverse annule. Ventral spicules alae weakly developed. Male with two small cuticularized protuberances on non-annulated tail part.

#### Relationships

*Dracognomus americanum* n. sp. is similar to *D. simplex* in having most modified cephalic adhesion tubes not positioned on helmet; no posterior adhesion tubes present posterior to anus/cloaca; males with precloacal copulatory thorns and females with large vulvar sclerotized pieces. They differ in that *D. americanum* n. sp. has 8 CAT all just posterior to helmet, while *D. simplex* has 12 CAT mostly posterior to helmet; *D. americanum* n. sp. has 9 copulatory thorns, evenly distributed, while *D. simplex* has 11 thorns, mainly in one cluster; PAT in *D. americanum* n. sp. clearly less robust and sublateral PAT tip without any differentiation (i.e., disc-like shaped tip); *D. americanum* n. sp. has small cloacal flaps provided with a single transverse annule and weakly developed ventral alae, while *D. simplex* has well-developed cloacal flaps with 3–4 transverse annules and well-developed ventral alae.

#### DISCUSSION

Two types of posterior adhesion tubes have been observed within the Draconematidae so far, i.e., typical or modified tubes. The typical or normal tube terminates in a characteristic open bell-shaped enlargement, containing a tongue-like triangular structure. The modified tube, within the Draconematidea restricted to the genus *Dracognomus*, has no tip differentiation and is gradually tapering toward a narrow open tip. The modified tubes resemble long somatic setae but differ from them by their slightly swollen base and narrowing tip (Allen and Noffsinger, 1978; Decreamer et al., 1997). The observed unusual flexible, thin disc-like



structure on the sublateral PAT tips is clearly different from the known typical and modified adhesion tubes. We have not observed this structure in the other studied *Dracognomus* species. Allen and Noffsinger (1978) marked these tubes as typical and illustrated the characteristic tip, but without a tongue-like structure and not enlarged as in other drawn typical tubes. We propose to rename these tubes as typical disc-shaped tubes as in contradistinction to typical bell-shaped tubes.

All described *Dracognomus* species were observed for the preparation of an identification key, based on males and females. Particularly *D. marioni*, *D. nothohalensis*, *D. dermatoglyphus*, and *D. annae* are difficult to identify with females only. Although morphologically closely related, we were able to separate these males rather easily. Particularly spiculum and gubernaculum morphology and morphometrics are useful for identification of these species. For correct genus identification we recommend using Allen and Noffsinger (1978) and Decraemer et al. (1997).

Our conclusion that the western North Atlantic specimens are a different species from Mediterranean specimens of *D. simplex* may have implications for all American marine nematodes, routinely identified as conspecific with European species. Particularly SEM studies, or other detailed comparative studies, could be worthwhile to aid in marine nematode studies.

#### KEY TO THE *DRACOGNOMUS* SPECIES

- 1) Male with pre-cloacal copulatory thorns; female with large sclerotized vulvar pieces .....2.  
Male without copulatory thorns; female without sclerotized vulvar pieces .....3.
- 2) Head with 12 CAT, sublateral PAT tip disc-like shaped; male with 11 copulatory thorns and pronounced cloacal flaps with 3–4 transverse annules .....*D. simplex*.  
Head with 8 CAT, sublateral PAT tip not differentiated; male with 9 copulatory thorns and small cloacal flaps with a single transverse annule .....*D. americanum* n. sp.
- 3) Male amphid length  $\geq 9 \mu\text{m}$ ; female amphid length  $\geq 7 \mu\text{m}$ .....*D. tinae*.  
Male amphid length  $< 9 \mu\text{m}$ , female amphid length  $< 7 \mu\text{m}$  .....4.
- 4) Male with cloacal flap, spicules without capitulum .....5.  
Male without cloacal flap, spicules with capitulum .....6.
- 5) Male subventral PAT not broad-based and spurred, gubernaculum  $\pm 9 \mu\text{m}$  .....*D. nothohalensis*.  
Male subventral PAT broad-based and spurred, gubernaculum  $\pm 18 \mu\text{m}$ .....*D. marioni*.
- 6) Helmet dorsally clearly indented, amphid nearly transverse, capitulum hook-shaped .....  
.....*D. dermatoglyphus*.  
Helmet dorsally rounded, amphid longitudinal, capitulum rounded.....*D. annae*.

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