Three new species of Carcinonemertes (Nemertea, Carcinonemertidae) from the southeastern coast of Brazil

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Abstract
Three new species of Carcinonemertes from the southeastern coast of Brazil are described: Carcinonemertes divae new species, Carcinonemertes caissarum new species and Carcinonemertes sebastianensis new species. They were found, respectively, associated with the crabs Libinia spinosa, Hepatus pudibundus and Menippe nodifrons, each a newly recorded host for Carcinonemertes. Characters not previously used to describe members of the family Carcinonemertidae, such as distance from ovaries to tip of head, distance from brain to tip of head and distance from stylet to tip of head are included in the descriptions of the new species and are discussed. The locations of these new carcionemertid worms in their respective hosts are presented in detail and a novelty regarding the infestation site is registered.

Keywords: Carcinonemertidae, decapod crustacean, symbiosis, predation, Carcinonemertes divae, Carcinonemertes caissarum, Carcinonemertes sebastianensis, Brazil

Introduction
Nemertean worms of the family Carcinonemertidae are symbiotic egg predators of many decapod crustaceans. Owing to their life cycle, intimacy and use of chemically mediated cues from their hosts, their biology is effectively akin to parasitism; their ecological impact, however, is that of a predator because they kill individual embryos (Kuris 1993; Torchin et al. 1996).

The family Carcinonemertidae comprises two genera, Carcinonemertes (Kölliker 1845), which lacks accessory stylets, and Ovicides Shields, 2001, the latter with a single species, O. juliae Shields, 2001, which has accessory stylets. Ten species of Carcinonemertes have been described so far: C. carcinophila (Kölliker 1845), with two recognized subspecies, C. c. carcinophila (Kölliker 1845) and C. c. imminuta Humes, 1942; C. epialti Coe ,1902; C.
To date only *Carcinonemertes carcinophila imminuta* has been reported for Brazil, in the portunid crabs *Callinectes danae* Smith and *Callinectes ornatus* Ordway (Santos and Bueno 2001). In the present paper, three new species of the genus *Carcinonemertes* are described, obtained from the crabs *Libinia spinosa* H. Milne Edwards (Decapoda, Pisidae), *Hepatus pudibundus* (Herbst) (Decapoda, Hepatidae) and *Menippe nodifrons* Stimpson (Decapoda, Menippidae) collected on the southeastern coast of Brazil. The locations of these new worms in their respective hosts are presented in detail. The distinctive morphological characters of the new species are compared to the previously described ones; new characters are included in the descriptions and discussed.

**Material and methods**

Decapod crustaceans were collected by trawling on the southeastern coast of Brazil along the following beaches: Anchovas Beach, São Sebastião Island, (23°55′S, 45°19′W); Enseada Beach, São Sebastião, (23°43′S, 45°25′W), and Poço Beach, São Sebastião Island, (23°45′S, 45°16′W). Crabs were also collected by hand at Figueira Beach, São Sebastião (23°44′56″S, 45°24′34″W).

Crabs were transported to the nearby laboratory facilities at the Centro de Biologia Marinha, Universidade de São Paulo, São Sebastião, where they were kept alive in tanks with a flow-through seawater system until dissection. Identification of crabs followed Melo (1996).

The exoskeleton surface and the arthrodial membranes of the crabs were macroscopically examined for nemerteans. The dorsal carapace was removed to expose the branchial chambers. Gills and pleopods were removed with the aid of forceps and were examined under a dissecting microscope.

The nemerteans were collected from crabs and placed in Petri dishes filled with seawater until the moment of taking measurements, pictures and notes on characters. Nemerteans were relaxed in a 1:1 solution of 7.5% MgCl₂ (prepared with bottled drinking water) and seawater for 15–30 min, after which, length and width of body were determined with the aid of an ocular micrometer in a dissecting microscope. Measurements of internal features were made with the aid of an ocular micrometer in a compound microscope after covering the worms with a coverslip. Photomicrographs were made with a Canon PowerShot A10 digital camera. Holotype and paratypes of each new species are deposited at the Museu de Zoologia da Universidade de São Paulo, Brazil (Abbreviation: MZUSP).

Some mucus sheaths produced by *Carcinonemertes sebastianensis* n. sp were prepared for scanning electron microscopy to obtain more details on their morphology. The mucus sheaths were cleaned in a 1:1 solution of 7.5% MgCl₂ (prepared with bottled drinking water) and seawater and were fixed in a 10% seawater formalin solution. Mucus sheaths were dehydrated in a series of graded ethanol (50, 70, 95, 100 and 100% for 10 min each), dried by the critical point method with CO₂ and coated with gold, then examined with a JEOL 6400 Visions scanning electron microscope.
Systematics

**Family Carcinonemertidae** Sumner, Osburn & Cole, 1913

*Diagnosis*

Modified by Shields et al. (1989) from Humes (1942): Monostiliferous hoplonemerteans living as symbionts (egg predators) on the gills, under the abdomen, on the apodemes, and axillae, and in or on the egg masses of decapod crustaceans. Short proboscis, reaching scarcely beyond the posterior end of the muscular portion of the esophagus. Lateral nerves lie internal to the well-developed submuscular glands. Cephalic glands well developed, with cephalic muscle fibers present. Cerebral organs lacking. Takakura’s duct system present in males. Internal fertilization and oviparity occur commonly. In most species, adult worms occupy, at least temporarily, mucus sheaths secreted and attached to the setae on the pleopods and hairs of endopodites of ovigerous decapods. Embryos hatch as hoplonemertean larvae.

**Genus Carcinonemertes** (Kölliker, 1845)

*Diagnosis*

From Coe (1902) and amended here (*italic*): Nemerteans living as symbionts (egg predators) on various species of Crustacea. Proboscis but little developed, very small in size, and extremely short, without lateral pouches of reserve stylets, but armed with central stylet and basis only; anterior proboscis very short, without distinct muscular layers, without distinct nerves, and without a thickened glandular epithelium. Cerebral organs lacking. Two ocelli. Cephalic glands massively developed. Usually oviparous, though fertilization often takes place internally.

*Carcinonemertes divae* new species

(Figure 1A–F)

*Diagnosis*

Body color varies from translucent white to orange. Two eyes, black. Anterior end of body rounded or pointed; posterior end pointed. Worms 1.3–4.3 mm long. Accessory stylets absent. Ovaries arranged in one row on each side of intestine. Takakura’s duct present. Ornamented and filiform mucus sheath attached to pleopods of hosts may be present in adult worms.

*Material examined*

Fourteen females, 16 males and one larva were examined. Holotype: male, from the egg mass of *Libinia spinosa*; type locality: Poço Beach, São Sebastião Island, Brazil, (23°45’S, 45°16’W), 18 Jul 2003; Coll. Cynthia Santos; MZUSP No. 001. Paratype: female, from the egg mass of *L. spinosa*; type locality: Poço Beach, São Sebastião Island, Brazil (23°45’S, 45°16’W); 18 Jul 2003; Coll. Cynthia Santos; MZUSP No. 002.
Etymology

The species name is a noun in the genitive singular and honors Dr Diva D. Corrêa, a Brazilian nemertean specialist, formerly from the Universidade de São Paulo, who dedicated almost 40 years of her life to the study of nemerteans and greatly contributed to the knowledge of this phylum.

Etymology

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**Description**

The description is based on living adults and one larva. The latter was obtained from crabs collected at Anchovas Beach. Measurements are given as mean ± SE (range, number of specimens observed).

**Female.** Body color varied from translucent white to orange; gut orange; gonads translucent white. Two eyes, black, irregular, circular or elliptical; elliptical eyes the most common shape. Found free among egg mass of host or in a filiform, ornamented mucus sheath attached to the pleopods. Lapilli homogeneously distributed on mucus sheath and 4 ± 1 µm (3–5 µm; n = 2) in height. Anterior end of body rounded or pointed (Figure 1A); posterior end pointed. Dimensions of relaxed worms 2.6 ± 0.1 mm (1.3–3.2 mm; n = 14) long and 332 ± 17 µm (200–480 µm; n = 14) wide. Eye 26 ± 1 µm (20–36 µm; n = 13) long and 16 ± 1 µm (10–20 µm; n = 13) wide. Distance between eyes 103 ± 4 µm (80–130 µm; n = 12). Distance from eyes to tip of head 143 ± 9.5 µm (100–204 µm; n = 12). Brain 117 ± 9 µm (92–150 µm; n = 7) long and 57 ± 3 µm (45–70 µm; n = 7) wide. Distance from brain to tip of head 171 ± 7 µm (140–200 µm; n = 8). Proboscis aligned in a straight line (Figure 1B). Anterior proboscis chamber 75 µm (n = 1) long and 12 µm (n = 1) wide. Diaphragm 53 ± 2 µm (42–80 µm; n = 14) long and 54 ± 3 µm (45–78 µm; n = 14) wide. Single stylet on basis, 10 ± 1 µm (8–12 µm; n = 13) long. Stylet basis 25 ± 1 µm (22–30 µm; n = 14) long and 7 ± 0 µm (5–8 µm; n = 14) wide (Figure 1C). Stylet:basis ratio 0.387 (0.250–0.500; n = 13). Distance from central stylet to tip of head 221 ± 20 µm (130–400 µm; n = 12). Accessory stylets absent. Proboscis bulb 36 ± 2 µm (22–48 µm; n = 12) long and 45 ± 2 µm (38–56 µm; n = 14) wide. Posterior proboscis 57 ± 6 µm (30–72 µm; n = 6) long and 51 ± 5 µm (35–68 µm; n = 7) wide. Ovaries arranged in one row on each side of the intestinal diverticula. Number of ovaries 31 ± 4 (13–53; n = 11). Distance from first gonad to tip of head 428 ± 17 µm (348–540 µm; n = 12).

**Male.** Body color cream; gut orange; gonads translucent white. Two eyes, black, irregular, circular or elliptical; elliptical eyes the most common shape. Found free among egg mass of host or in a filiform, ornamented mucus sheath attached to the pleopods. Lapilli homogeneously distributed on mucus sheath and 4 µm (n = 1) in height. Anterior end of body rounded or pointed (Figure 1D); posterior end pointed (Figure 1E). Dimensions of relaxed worms 2.6 ± 0.2 mm (1.3–4.3 mm; n = 14) long and 316 ± 32 µm (170–720 µm; n = 16) wide. Eye 29 ± 2 µm (22–48 µm; n = 16) long and 17 ± 1 µm (10–25 µm; n = 16) wide. Distance between eyes 96 ± 3 µm (75–112 µm; n = 15). Distance from eyes to tip of head 160 ± 10 µm (110–230 µm; n = 16). Brain 118 ± 9 µm (90–175 µm; n = 9) long and 52 ± 3 µm (45–70 µm; n = 7) wide. Distance from brain to tip of head 212 ± 5 µm (200–260 µm; n = 13). Proboscis aligned in a straight line. Anterior proboscis chamber 46 ± 6 µm (40–52 µm; n = 2) long and 18 µm (n = 2) wide. Diaphragm 48 ± 1 µm (45–55 µm; n = 14) long and 50 ± 2 µm (38–60 µm; n = 14) wide. Single stylet on basis 10 ± 0 µm (8–12 µm; n = 13) long. Stylet basis 28 ± 1 µm (22–30 µm; n = 15) long and 7 ± 0 µm (5–10 µm; n = 15) wide. Stylet:basis ratio 0.371 (0.250–0.500; n = 13). Distance from central stylet to tip of head 231 ± 14 µm (160–310 µm; n = 13). Accessory stylets absent. Proboscis bulb 35 ± 2 µm (25–48 µm; n = 13) long and 39 ± 2 µm (25–48 µm; n = 13) wide. Posterior proboscis 72 ± 11 µm (40–100 µm; n = 5) long and 48 ± 1 µm (25–50 µm; n = 6) wide. Distance from first gonad to tip of head 475 ± 9 µm (370–500 µm; n = 12). Seminal vesicle (Figure 1E) easily visible under stereomicroscope in the majority of adult male worms.
**Larva.** Body ciliated with anterior and posterior ciliary tuft. Body shape spherical or ovoid. Two eyes. Length 110 μm (n=1). Width 70 μm (n=1). Anterior tuft 20 μm (n=1) in length. Posterior tuft 24 μm (n=1) in length. Eye 12 μm (n=1) long and 6 μm (n=1) wide. Distance between eyes 22 μm (n=1). Distance from eyes to tip of head 33 μm (n=1).

**Infestation site**

Adult worms were found in the host’s egg mass (Figure 1F) (ovigerous females with eggs in initial, intermediate and final stages of development). Immature worms were found on the abdomen of juvenile male crabs; on the abdomen and at the arthrodial membrane of pereopods of juvenile female crabs; at the base of pereopods, on the ventral and dorsal sides of the abdomen of adult male crabs; on the ventral and dorsal sides of the abdomen of non-ovigerous adult female crabs; at the base of pleopods, on the abdomen and on the eggs of ovigerous females; at the base of pleopods and on the ventral side of the abdomen of post-ovigerous females.

Figure 2. *Carcinonemertes caissarum* n. sp. (A) Filiform mucus sheath of adult worm; (B) male with rounded posterior end; (C) stylet and basis (arrow) in a male worm; (D) juvenile worm encysted on pleopod setae of *Hepatus pudibundus*. 
Carcinonemertes caissarum new species
(Figure 2A–D)

Diagnosis

Body color varies from translucent white to cream; male with a red spot at posterior end. Two eyes, black. Anterior end of body rounded, posterior end rounded or truncated (males). Worms 1.2–11.0 mm long. Accessory stylets absent. Ovaries arranged in one row on each side of intestine. Takakura’s duct present. Ornamented and filiform mucus sheath may be present in adult worms.

Material examined

Nine females and eleven males were examined. Holotype: female, from the egg mass of Hepatus pudibundus; type locality: Poço Beach, São Sebastião Island, Brazil, (23°45’S, 45°16’W), 16 September 2002; Coll. Cynthia Santos; MZUSP No. 003. Paratype: female, from the egg mass of H. pudibundus; type locality Poço Beach, São Sebastião Island, Brazil, (23°45’S, 45°16’W), 16 September 2002; Coll. Cynthia Santos; MZUSP No. 004.

Etymology

The species name is a noun in the genitive plural and is in recognition of the human communities known as “caicaras” (a tupi-guarani word), which are found in the localities where the crabs were collected. The people of these communities depend mainly on fisheries; therefore, this name honors the fishermen who helped us collect, Rogério dos Santos Júnior and his father, Rogério dos Santos.

Description

The description is based on living adults. Measurements are given in mean ± SE (range, number of specimens observed).

Female. Body color varied from translucent white to cream; gut varied from orange to brownish; gonads translucent white. Two eyes, black, round or cup-shaped; round eyes the most common shape. Found free among egg mass of host or in a filiform, ornamented mucus sheath (Figure 2A). Lapilli 7 ± 0 μm (7–8 μm; n = 2) in height, larger at center of sheath and smaller and scarce at extremities. Anterior and posterior end of body rounded. Dimensions of relaxed worms 5.5 ± 1.0 mm (2.6–11.0 mm; n = 7) long and 282 ± 20 μm (210–376 μm; n = 8) wide. Eye 11 ± 9 μm (8–15 μm; n = 9) long and 9 ± 1 μm (5–12 μm; n = 9) wide. Distance between eyes 112 ± 11 μm (75–160 μm; n = 7). Distance from eyes to tip of head 115 ± 6 μm (100–155 μm; n = 9). Brain 97 ± 6 μm (88–120 μm; n = 5) long and 46 ± 2 μm (40–52 μm; n = 7) wide. Distance from brain to tip of head 132 ± 6 μm (120–160 μm; n = 6). Anterior proboscis chamber could not be distinguished. Diaphragm 38 ± 2 μm (30–48 μm; n = 8) long and 46 ± 2 μm (38–55 μm; n = 7) wide. Single stylet on basis 8 ± 1 μm (5–10 μm; n = 8) long. Stylet basis 22 ± 1 μm (20–25 μm; n = 9) long and 7 ± 0 μm (5–8 μm; n = 9) wide. Stylet:basis ratio 0.378 (0.250–0.500; n = 8). Distance from central stylet to tip of head 160 ± 8 μm (122–190 μm; n = 8). Accessory stylets absent. Proboscis bulb 26 ± 2 μm (20–38 μm; n = 8) long and 30 ± 2 μm (22–38 μm; n = 8) wide.
Posterior proboscis 70 ± 4 μm (55–88 μm; n=4) long and 59 ± 4 μm (52–68 μm; n=4) wide. Ovaries arranged in one row on each side of the intestinal diverticula. Number of ovaries 158 ± 30 (76–252; n=5). Distance from first gonad to tip of head 568 ± 56 μm (430–900 μm; n=8).

**Male.** Body color cream with a red spot at the posterior end; gut varied from orange to brownish; gonads translucent white. Two eyes, black, irregular, circular or cup-shaped. Found free among egg mass of host or in a filiform, ornamented mucus sheath. Lapilli 4 μm (n=1) in height. Anterior end of body rounded, posterior end rounded (Figure 2B) or truncated. Dimensions of relaxed worms 2.0 ± 0.3 mm (1.2–4.0 mm; n=9) long and 400 ± 25 μm (230–520 μm; n=11) wide. Eye 11 ± 1 μm (8–15 μm; n=10) long and 10 ± 0 μm (8–12 μm; n=10) wide. Distance between eyes 107 ± 9 μm (62–150 μm; n=8). Distance from eyes to tip of head 106 ± 7 μm (60–130 μm; n=10). Brain 110 ± 6 μm (86–125 μm; n=8) long and 54 ± 4 μm (38–75 μm; n=8) wide. Distance from brain to tip of head 135 ± 9 μm (80–170 μm; n=8). Anterior proboscis chamber could not be distinguished. Diaphragm 42 ± 4 μm (30–60 μm; n=7) long and 42 ± 2 μm (32–50 μm; n=7) wide. Single stylet on basis 8 ± 1 μm (5–10 μm; n=9) long. Stylet basis 21 ± 1 μm (18–25 μm; n=10) long and 7 ± 0 μm (5–8 μm; n=9) wide (Figure 2C). Stylet:basis ratio 0.372 (0.250–0.571; n=9). Distance from central stylet to tip of head 172 ± 13 μm (110–216 μm; n=8). Accessory stylets absent. Proboscis bulb 22 ± 2 μm (20–25 μm; n=2) long and 30 μm (n=1) wide. Posterior proboscis could not be observed. Distance from first gonad to tip of head 325 ± 46 μm (230–510 μm; n=5). Seminal vesicle easily visible under stereomicroscope in the majority of adult male worms.

**Infestation site**

Adult worms were found in the host’s egg mass (ovigerous females with eggs in initial, intermediate and final stages of development). Immature worms were found on the ventral side of the abdomen of adult male crabs; on the abdomen, at the gonopores, at the base of pleopods and encysted on the setae of pleopods (Figure 2D) of non-ovigerous adult female crabs; encysted on the setae of pleopods of ovigerous females; on the abdomen, at the gonopores, on the arthrodial membrane of pleopods, and encysted on the setae of pleopods of post-ovigerous females.

**Carcinonemertes sebastianensis** new species

(Figures 3, 4A–E and 5A, B)

**Diagnosis**

Body color translucent white. Two eyes, brown. Anterior and posterior end of body rounded. Worms 2.0–11.0 mm long. Accessory stylets absent. Ovaries arranged in one row on each side of intestine. Distance from first ovary to tip of head 1250–2000 μm. Takakura’s duct present. Ornamented and filiform mucus sheath may be present in adult worms.

**Material examined**

Four females, six males, 12 eggs and one larva were examined. Holotype: male, from the egg mass of *Menippe nodifrons*; type locality: Figueira Beach, São Sebastião, Brazil,
Paratype: female, from the egg mass of *M. nodifrons*; type locality: Figueira Beach, São Sebastião, Brazil, (23°44′56″S, 45°24′34″W); 22 January 2004; Coll. Cynthia Santos; MZUSP No. 005.

**Etymology**

The species name is an adjective in feminine singular and is in recognition of the locality where the host crab was found, São Sebastião.

**Description**

The description is based on living adults, eggs and larva. Measurements are given in mean ± SE (range, number of specimens observed).

*Female.* Body color translucent white; gut varied from orange to brownish; gonads translucent white. Two eyes, brown, circular or elliptical. Found free among egg mass of host or in a filiform, ornamented mucus sheath. Irregular lapilli on the mucus sheath (Figure 3); lapilli with concentric lines (Figure 4A). Anterior (Figure 4B) and posterior end of body rounded. Dimensions of relaxed worms were $7.0 \pm 4.0 \text{ mm} \ (3.0–11.0 \text{ mm}; \ n=2)$ long and $405 \pm 62 \mu m \ (230–500 \mu m; \ n=4)$ wide. Eye $14 \pm 2 \mu m \ (10–18 \mu m; \ n=4)$ long and $9 \pm 1 \mu m \ (8–12 \mu m; \ n=4)$ wide. Distance between eyes $131 \pm 14 \mu m \ (102–160 \mu m; \ n=4)$. Distance from eyes to tip of head $110 \pm 18 \mu m \ (70–150 \mu m; \ n=4)$. Brain $99 \pm 6 \mu m \ (88–110 \mu m; \ n=3)$ long and $64 \pm 4 \mu m \ (58–72 \mu m; \ n=3)$ wide. Distance from brain to tip of head $150 \pm 23 \mu m \ (100–210 \mu m; \ n=4)$. Proboscis bent on itself. Anterior proboscis chamber could not be distinguished. Diaphragm $51 \pm 2 \mu m \ (48–58 \mu m; \ n=4)$ long and $56 \pm 3 \mu m \ (50–62 \mu m; \ n=4)$ wide. Single stylet on basis $9 \pm 1 \mu m \ (8–10 \mu m; \ n=4)$ long. Stylet basis $22 \pm 1 \mu m \ (20–25 \mu m; \ n=4)$ long and $8 \pm 1 \mu m \ (8–10 \mu m; \ n=4)$ wide (Figure 4C). Stylet:basis ratio $0.416 \ (0.375–0.444; \ n=4)$. Distance from central stylet to tip of head $208 \pm 28 \mu m \ (170–290 \mu m; \ n=4)$. Accessory stylets absent. Proboscis bulb
34 ± 4 μm (28–42 μm; n=4) long and 38 ± 4 μm (30–42 μm; n=4) wide. Posterior proboscis could not be visualized. Ovaries arranged in one row on each side of the intestinal diverticula. Distance from first gonad to tip of head 1700 ± 167 μm (1250–2000 μm; n=4) (Figure 4B).

Male. Body color translucent white; gut yellow; gonads translucent white. Two eyes, brown, circular or elliptical; elliptical eyes the most common shape. Found free among
egg mass of host or in a filiform, ornamented mucus sheath. Irregular lapilli on mucus sheath (Figure 3); lapilli with concentric lines (Figure 4A). Anterior and posterior end of body rounded (Figure 4D, E). Dimensions of relaxed worms 6.2 ± 1.4 mm (2.0–11.0 mm; n = 6) long and 362 ± 28 µm (280–480 µm; n = 6) wide. Eye 15 ± 1 µm (12–18 µm; n = 6) long and 10 ± 1 µm (5–12 µm; n = 6) wide. Distance between eyes 110 ± 5 µm (90–120 µm; n = 6). Distance from eyes to tip of head 150 ± 13 µm (112–200 µm; n = 6). Brain 101 ± 10 µm (75–125 µm; n = 5) long and 52 ± 3 µm (42–62 µm; n = 5) wide. Distance from brain to tip of head 206 ± 24 µm (150–280 µm; n = 5). Proboscis bent in on itself. Anterior proboscis chamber could not be distinguished. Diaphragm 46 ± 2 µm (40–50 µm; n = 6) long and 48 ± 1 µm (45–50 µm; n = 6) wide. Single stylet on basis 11 ± 0 µm (10–12 µm; n = 6) long. Stylet basis 25 ± 1 µm (22–30 µm; n = 6) long and 10 ± 0 µm (8–10 µm; n = 6) wide (Figure 4C). Stylet:basis ratio 0.431 (0.333–0.556; n = 6). Distance from central stylet to tip of head 206 ± 24 µm (150–280 µm; n = 5). Accessory stylets absent. Proboscis bulb 30 ± 2 µm (25–38 µm; n = 6) long and 36 ± 2 µm (32–40 µm; n = 6) wide. Posterior proboscis 128 ± 18 µm (100–160 µm; n = 3) long and 43 ± 9 µm (25–55 µm; n = 3) wide. Seminal vesicle not visible under stereomicroscope, but could be seen under compound microscope in some specimens.

Egg. Color white. Egg strings (Figure 5A) found in a firm and transparent sheath. Developing eggs 86 ± 2 µm in diameter (75–100 µm; n = 12). Egg strings 232 ± 14 µm (180–260 µm; n = 5) wide. Number of eggs per egg string 386 ± 66 (320–451; n = 2). Number of rows of eggs per egg string 3 ± 0 (n = 5).

Larva. Body ciliated with anterior and posterior ciliary tufts. Body shape ovoid. Two eyes. Body length 92 µm (n = 1). Body width 62 µm (n = 1).

Infestation site

Adult worms were found in the host’s egg mass (ovigerous females with eggs in initial, intermediate and final stages of development). All immature worms were found encysted in a mucus sheath without ornamentation in the following sites: on the ventral side of the abdomen, on the setae that follow the border of the abdomen, on the central axis of

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**Figure 5.** *Carcinonemertes sebastianensis* n. sp. (A) Egg strand (arrow) on *Menippe nodifrons* eggs. (B) Juvenile worms encysted (arrows) on the axis of the pleopod of *Menippe nodifrons.*
pleopods (Figure 5B), and on the setae of pleopods of non-ovigerous adult female crabs; on the ventral side of the abdomen, on the central axis of pleopods, and on the setae of pleopods of ovigerous females; at the ventral side of the abdomen, on the central axis of pleopods, and on the setae of pleopods of post-ovigerous females. No immature worms were found on male crabs.

Discussion

The three species described here are gonochoric, do not present accessory stylets and, thereby, conform to the diagnosis of Carcinonemertidae provided by Humes (1942) and modified by Shields et al. (1989) and to the diagnosis of Carcinonemertes provided by Coe (1902). The genus Ovicides is characterized by being hermaphroditic and by having two accessory stylets (Shields 2001).

Carcinonemertes divae n. sp., C. caissarum n. sp. and C. sebastianensis n. sp. can be distinguished from each other by color of the body, shape of the posterior region of body, eye length, and by distance between the first ovary and tip of head (Table I).

Carcinonemertes pinnotheridophila is found on the floor and wall of branchial chambers as well as on eggs of its hosts (McDermott and Gibson 1993). Juvenile worms of C. c. carcinophila, C. c. imminuta and C. mitsukurii are found between the gills of their hosts (Humes 1942; Shields 1992; Santos and Bueno 2001; Table II). None of the three species described here was found in the branchial chamber or between the gills of their hosts. The three species described here also can be distinguished from C. pinnotheridophila by the presence of eyes in the adult worms, shape of the egg mucus sheath and by the number of rows of ovaries on each side of the intestine (McDermott and Gibson 1993) (Table II). Carcinonemertes coei also has two rows of ovaries on each side of the intestine (Humes 1942; Table III).

Carcinonemertes divae n. sp., C. caissarum n. sp. and C. sebastianensis n. sp. can be distinguished from C. c. carcinophila by body length (Humes 1942), and from C. c. imminuta by body length and by eye width (Humes 1942; Table II).

Adult worms of C. errans do not have a mucus sheath (Wickham 1978), whereas adult worms of C. regicides have a mucus sheath with no lapilli (Shields et al. 1989). The adult worms of the three species described here have a mucus sheath with lapilli, which are, according to Humes (1942), irregularly shaped, raised concretions, present on the surface of the sheath. In addition, C. errans can be distinguished from C. divae n. sp., C. caissarum n. sp. and C. sebastianensis n. sp. by diaphragm length and by stylet basis length (personal

Table I. Morphological and ecological differences that distinguish the species Carcinonemertes divae n. sp., Carcinonemertes caissarum n. sp. and Carcinonemertes sebastianensis n. sp. (measurements in μm).

| Characters                  | C. divae n. sp. | C. caissarum n. sp. | C. sebastianensis n. sp. |
|-----------------------------|-----------------|---------------------|--------------------------|
| Body color                  | White to orange | White to cream, red spot (male) | White                   |
| Shape of posterior end of body | Pointed        | Truncated on males  | Rounded                 |
| Eye length                   | 20–48           | 8–15                | 10–18                    |
| Distance 1st ovary–tip of head | 348–540        | 430–900             | 1250–2000               |
| Infestation site            | e, ab, per, pl | e, ab, gon, pl, sepl | e, ab, seab, axpl, sepl |

Abbreviations for infestation site: ab, abdomen; axpl, axis of pleopod; bc, branchial chamber; e, eggs; gl, gill lamellae; gon, gonopores; per, pereiopods; pl, pleopods; seab, setae of abdomen; sepl, setae of pleopods.
Table II. Morphological and ecological differences that distinguish the species *Carcinonemertes carcinophila*, *Carcinonemertes carcinophila imminuta*, *Carcinonemertes mitsukurii* and *Carcinonemertes pinnotheridophila* from the species described here (body length in mm; other measurements in μm).

| Characters                     | *C. c. carcinophila* | *C. c. imminuta* | *C. mitsukurii* | *C. divae* n. sp. | *C. caissarum* n. sp. | *C. sebastianensis* n. sp. |
|-------------------------------|----------------------|------------------|-----------------|-------------------|-----------------------|-----------------------------|
| Body length                   | 20.0–70.0            | 4.0–15.0         | 1.0–16.0        | 0.9–15.0          | 1.3–4.3               | 1.2–11.0                    |
| Eye width                     | ?                    | 20–70            | ?               | eyeless           | 10–25                 | 5–12                        |
| Number of rows of ovaries     | 1                    | 1                | 1               | 2                 | 1                     | 1                           |
| Shape of egg mucus sheath     | Filiform             | Filiform         | Filiform        | Oval              | Filiform              | Filiform                    |
| Infestation site              | e, gl                | e, gl            | e, gl           | e, bc             | e, ab, per, pl        | e, ab, seab, axpl, sepl     |
|                               |                      |                  |                 |                   |                       |                             |
| Table III. Morphological and ecological differences that distinguish the species *Carcinonemertes coei*, *Carcinonemertes errans*, *Carcinonemertes regicidae* and *Carcinonemertes australiensis* from the species described here (measurements in μm).

| Characters                     | *C. coei* | *C. errans* | *C. regicidae* | *C. australiensis* | *C. divae* n. sp. | *C. caissarum* n. sp. | *C. sebastianensis* n. sp. |
|-------------------------------|-----------|-------------|----------------|---------------------|-------------------|-----------------------|-----------------------------|
| Presence of mucus sheath      | Present   | Absent     | Present        | ?                   | Present           | Present               | Present                     |
| Mucus sheath ornamentation    | ?         | N/A         | No lapilli      | ?                   | Ornamented        | Ornamented            | Ornamented                  |
| Diaphragm length              | ?         | 62–125     | 64–96          | 75                  | 42–80             | 30–60                 | 40–58                       |
| Stylet basis length           | 21–23     | 30–40       | 38–44          | 40                  | 22–30             | 18–25                 | 20–30                       |
| Stylet basis width            | 6–8       | 10–13       | 9–17           | 15–16               | 5–10              | 5–8                   | 8–10                        |
| Stylet length                 | 8–9       | 12 (no variation) | 14–19     | 15–18               | 8–12              | 5–10                  | 8–12                        |
| Number of rows of ovaries     | 2         | 1           | 1              | 1                   | 1                 | 1                     | 1                           |

Abbreviations for infestation site: ab, abdomen; axpl, axis of pleopod; bc, branchial chamber; e, eggs; gl, gill lamellae; gon, gonopores; per, pereiopods; pl, pleopods; seab, setae of abdomen; sepl, setae of pleopods.
the eye length (Coe 1902; Table IV). The eye is 8–15 mm long in *C. caissarum* n. sp. and 10–18 mm long in *C. sebastianensis* n. sp. *Carcinonemertes epialti* also can be distinguished from *C. caissarum* n. sp. by stylet length (personal observation) and from *C. sebastianensis* new species by the distance between the eyes and tip of head (personal observation; Table IV).

Juvenile worms of some nemerteans of the genus *Carcinonemertes* were previously found on the pleopods of their hosts (at the base or on the arthrodial membranes). A novelty regarding the infestation site by nemerteans of the genus *Carcinonemertes* is registered in the present study for *C. sebastianensis* n. sp., whose juveniles can be found encysted on setae of pleopods (a site also used by juveniles of *C. caissarum* n. sp.) or on the axis of pleopods, or even on setae that follow the border of the abdomen of the hosts.

In addition to the diagnostics mentioned above, the three new species differ from other carcinonemertids in that they infest, respectively, *L. spinosa*, *H. pudibundus* and *M. nodifrons*, all registered here for the first time as hosts for nemerteans of the genus *Carcinonemertes*.

Specimens of *H. epheliticus* and *M. mercenaria* collected at Grand Isle, Louisiana, USA, were registered by Humes (1942) as hosts for *C. c. imminuta*. Humes (1942) found juvenile worms on gills of *H. epheliticus*, which makes us believe he could be right about the identification of these specimens of *Carcinonemertes*. Nevertheless, the following facts suggest that there is a chance the worms found in *H. epheliticus* and *M. mercenaria* were misidentified as *C. c. imminuta*: (1) only one worm was found on the gill of *H. epheliticus* from Louisiana (Humes 1942); this infestation could be casual, not a regular pattern; (2) an adult male of *H. epheliticus* was collected in Florida and none of the 90 juvenile worms infesting the crab was found on gills (personal observation); (3) Humes (1942) did not find any worms on the gills of *M. mercenaria* and stated that “in the non-portunid species infested at Grand Isle, the worms were all minute, though were sexually mature”; it seems that the worms found in *H. epheliticus* and in *M. mercenaria* by Humes (1942) were smaller than *C. c. imminuta*.

There are some factors that can lead to misidentifications among the species belonging to the family Carcinonemertidae: (1) the small size of worms, (2) morphological simplification apparently resulting from their parasite-like life style, and (3) the consequent morphological ambiguity of structures and morphological similarity between species. For example, the structure referenced as anterior proboscis chamber in some publications (Shields et al. 1989; Shields and Kuris 1990) appears to us to be the proboscis diaphragm and that is how
we treat it. Similarly, the region designated as the stylet bulb, appears to include the diaphragm in a number of publications (Gibson 1972; Campbell et al. 1989; Gibson and Jones 1990; McDermott and Gibson 1993); we distinguish between the two.

The stifling morphological homogeneity of Carcinonemertes species led us to search for other, relatively practical characters, such as distance from first gonad to tip of head, distance from brain to tip of head and distance from stylet to tip of head. Distance from first gonad to tip of head enabled us to distinguish among the three species described here. We believe that information, such as infestation site and infestation according to sex and maturity of host, may also provide valuable diagnostic and systematic data. With this information in hand from previously described species it may be possible to conduct phylogenetic analyses with useful results that will lead to understanding of the diversification and host relationships of this potentially damaging group of nemerteans.

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