

## A NEW SPECIES OF *CUCULLANUS* (NEMATODA: CUCULLANIDAE) PARASITIZING *CONGER ORBIGNIANUS* (PISCES: CONGRIDAE) FROM ARGENTINEAN WATERS

Juan T. Timi and Ana L. Lanfranchi

Laboratorio de Parasitología, Departamento de Biología, Facultad de Ciencias Exactas y Naturales, Universidad Nacional de Mar del Plata, Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Funes 3350, 7600 Mar del Plata, Argentina. e-mail: jtimi@mdp.edu.ar

**ABSTRACT:** In total, 17 specimens of *Conger orbignianus* Valenciennes, 1847 from waters off Mar del Plata, Argentina (38°08' S, 57°32' W) were examined for parasitic nematodes. A new nematode species, *Cucullanus pedroi* n. sp., is described (prevalence 76.5%,  $\bar{x}$  intensity  $\pm$  SD = 3.8  $\pm$  2.7). The new species closely resembles some species parasitizing other anguilliform fishes; however, it can be distinguished from most of its congeners by the distribution pattern of caudal papillae (particularly fourth and eight pairs) and phasmids. Those congeners with similar pattern of papillae differ from the new species by the length of the spicules and gubernaculum and by the position of the excretory pore and deirids.

*Cucullanus* Müller, 1777 comprises approximately 100 species (Petter, 1974; Hasegawa et al., 1991), 3 of which have so far been described in a series of recent studies carried out in Argentinean Atlantic waters, i.e., *Cucullanus genypteri* Sardella, Navone and Timi, 1997, a parasite of *Genypterus blacodes* Schneider, 1801 and *Genypterus brasiliensis* Reagan, 1903 (Ophidiidae) (Sardella et al., 1997); *Cucullanus marplatensis* Daniel, Timi and Sardella, 2002, a parasite of *Odontesthes argentinensis* (Valenciennes, 1835) (Atherinidae) (Daniel et al., 2002); and *Cucullanus bonaerensis* Lanfranchi, Timi and Sardella, 2003 from *Urophycis brasiliensis* (Kaup, 1858) (Phycidae) (Lanfranchi et al. 2004). A fourth species, reported from the Argentine conger, *Conger orbignianus* Valenciennes, 1847 (Anguilliformes: Congridae), from Bahía Blanca (38°45' S, 62°15' W) (Tanzola and Guagliardo, 2000), remains undescribed. During a parasitological survey carried out on samples of *C. orbignianus* landed by fishermen at Mar del Plata port, parasitic nematodes were found in the large intestine of fish: These parasites are herein described and a new species of *Cucullanus* is proposed.

### MATERIALS AND METHODS

In total, 17 specimens of *C. orbignianus* caught by commercial trawlers at the Mar del Plata port (38°08' S, 57°32' W) during March–April 2004 were examined for nematodes immediately after capture. Fish were dissected, and the intestines were removed and examined under a stereoscopic microscope. In total, 49 cucullanid nematodes were collected and fixed in 4% hot formaldehyde, preserved in 70% alcohol, cleared in lactophenol, and then studied and measured by light microscopy. Drawings were made using a drawing tube. For scanning electron microscopy (SEM), specimens were dehydrated using a series of ethanol washes, dried by evaporation with hexamethyldisilazane, coated with gold palladium, and scanned in a JEOL JSM 6460-LV SEM. All measurements are given in millimeters, unless otherwise indicated, with mean followed by standard deviation and range in parentheses. Caudal papillae nomenclature follows Petter (1974). Prevalence and mean intensity were calculated according Bush et al. (1997). The studied material was deposited in the Helminthological Collection of the Museo de La Plata (CHMLP), La Plata, Argentina.

### DESCRIPTION

#### *Cucullanus pedroi* n. sp.

(Figs. 1–14)

*General* (10 males and 10 females measured): Medium-sized nematodes. Body slender. Cuticle finely striated throughout. Lateral alae absent. Anterior end rounded, dorsoventrally expanded. Cephalic ex-

tremitry with usual features of *Cucullanus*, with 2 pairs of prominent cephalic papillae and a pair of amphids; inner ring of 3 pairs of small labial papillae not seen. Mouth slit-like dorsoventrally, surrounded by a collarete armed with numerous triangular denticles on each side. Pseudobuccal cavity well developed with internal cuticular lining, esophagus long and narrow, expanded at both extremities, opening into intestine through small valve; pseudobuccal capsule wider than posterior end. Nerve ring surrounding esophagus at approximately one-third of its length. Deirids located at the posterior third of esophagus. Excretory pore variable in position, slightly anterior to, at level of or posterior to distal end of esophagus. Right post-deirid preequatorial, left one post-equatorial. Anus with prominent anterior lip. Tail conical.

*Male:* Body length 13.26  $\pm$  2.23 (9.86–16.14) long, width at mid-body 0.38  $\pm$  0.05 (0.28–0.46). Esophagus length 1.44  $\pm$  0.12 (1.28–1.62), 11.05  $\pm$  1.45 (7.80–13.18)% of body length, width at base 0.20  $\pm$  0.03 (0.14–0.24); width of pseudobuccal capsule 0.22  $\pm$  0.03 (0.15–0.26). Distance of nerve ring from anterior extremity 0.46  $\pm$  0.02 (0.42–0.49), of excretory pore 1.43  $\pm$  0.23 (1.09–1.80), of deirids 1.01  $\pm$  0.15 (0.72–1.22), of right post-deirid 6.38  $\pm$  1.26 (4.82–8.44), and of left post-deirid 9.78  $\pm$  2.00 (7.48–13.04). Preecloacal sucker, distance from center to posterior body end 1.29  $\pm$  0.26 (0.86–1.80). Caudal papillae consisting of 1 medial adcloacal papilla and 11 pairs of papillae, 3 pairs precloacal (pair 1 anterior and pair 2 posterior to ventral sucker, respectively; pair 3 between sucker and cloaca but closer to latter), 4 pairs adcloacal (3 subventral [5–7], 1 lateral [4] situated at level of, or more frequently, slightly posterior to pair 7) and 4 pairs of postcloacal (pairs 9 and 10 subventral; pair 8 subdorsal, at level of pair 9; lateral phasmids, slightly anterior to or at level of pairs 8 and 9, and situated at 0.16  $\pm$  0.02 [0.14–0.18] from posterior extremity). Spicules broadly alate, subequal with pointed distal end, left spicule length 1.36  $\pm$  0.23 (0.94–1.60), right spicule length 1.31  $\pm$  0.22 (0.90–1.52), 10.15  $\pm$  1.45 (7.80–12.58)% of body length. Gubernaculum Y-shaped, length 0.22  $\pm$  0.02 (0.18–0.24); tail length 0.27  $\pm$  0.04 (0.20–0.33).

*Female:* Body length 14.70  $\pm$  1.74 (12.70–17.52), width at mid-body 0.39  $\pm$  0.05 (0.32–0.46). Esophagus length 1.47  $\pm$  0.10 (1.28–1.68), 10.07  $\pm$  1.24 (8.22–11.88)% of body length, width at base 0.20  $\pm$  0.03 (0.15–0.25); width of pseudobuccal capsule 0.23  $\pm$  0.01 (0.21–0.25). Distance of nerve ring from anterior extremity 0.48  $\pm$  0.02 (0.46–0.51), of excretory pore 1.44  $\pm$  0.21 (1.18–1.84), of deirids 0.99  $\pm$  0.09 (0.86–1.12), of right post-deirid 6.39  $\pm$  0.81 (4.64–7.70), and of left post-deirid 10.57  $\pm$  0.99 (8.62–11.88) (posterior to vulva). Vulva prominent, postequatorial, distance from anterior body end 8.63  $\pm$  0.90 (7.50–10.00), 58.88  $\pm$  4.15 (50.34–65.35)% of body length. Ovipositor directed anteriorly from vulva. Uteri amphidelphic. Egg length 0.085  $\pm$  0.004 (0.076–0.090), width 0.048  $\pm$  0.002 (0.044–0.052). Tail length 0.36  $\pm$  0.05 (0.30–0.46), with a pair of caudal papillae (phasmids) situated at 0.22  $\pm$  0.03 (0.17–0.26) from posterior extremity.

### Taxonomic summary

*Type host:* *C. orbignianus*

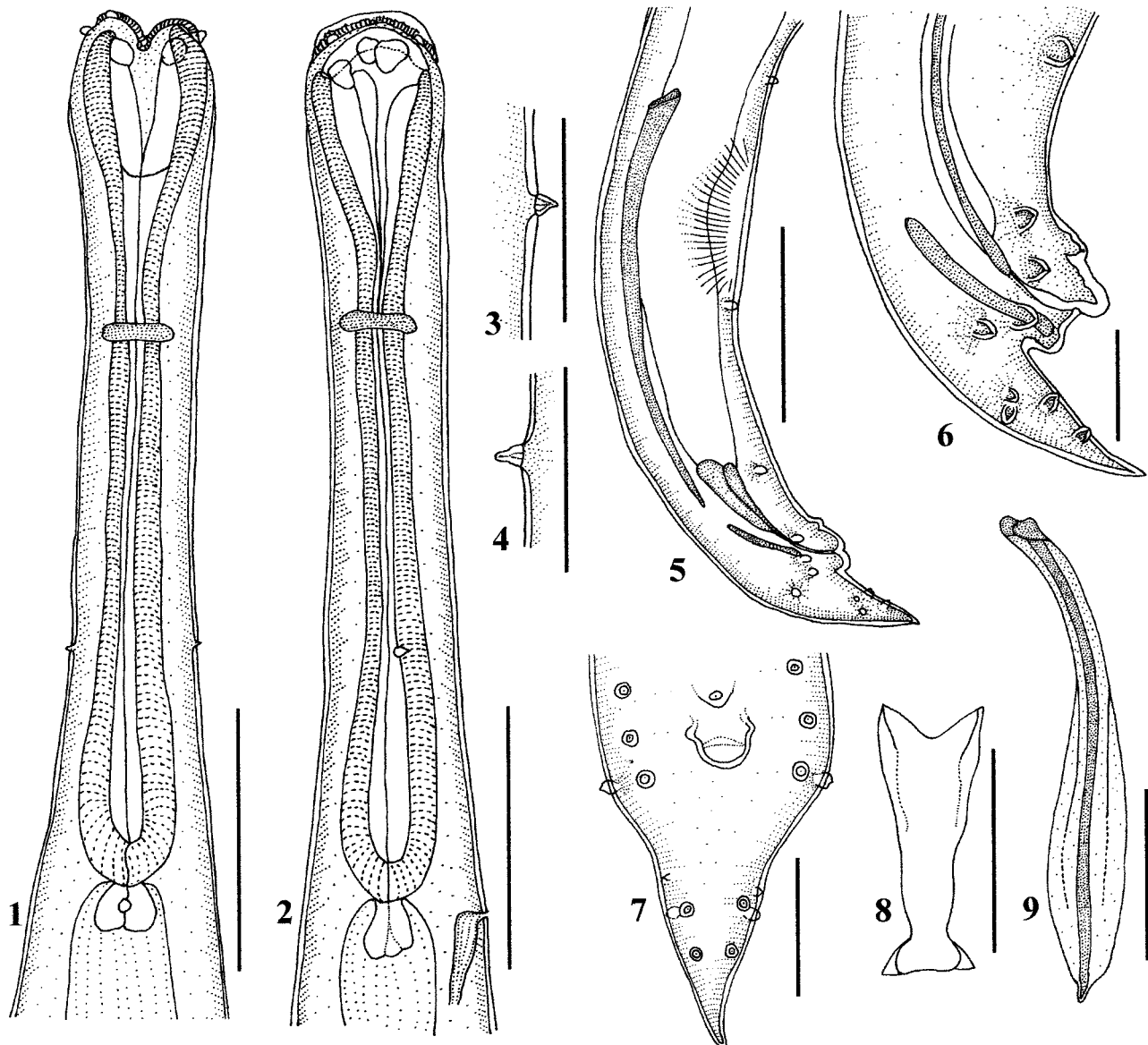
*Site:* Posterior end of intestine.

*Type locality:* Mar del Plata (coastal area of Buenos Aires Province, Argentina, 38°08' S, 57°32' W).

*Date of collection:* March–April 2004.

*Type specimens:* Holotype: 1 male (CHMLP coll. nr 5452); allotype:

Received 3 March 2005; revised 10 June 2005; accepted 10 June 2005.



FIGURES 1–9. *C. pedroi* n. sp. 1. Anterior end, ventral view. 2. Anterior end, lateral view. 3. Deirid. 4. Post-deirid. 5. Posterior end of male, lateral view. 6. Tail of male, lateral view. 7. Tail of male, ventral view. 8. Gubernaculum. 9. Evaginate spicule. Bars = 0.4 mm (1, 2, 5, 9); 0.1 mm (3, 4, 6, 7); 0.05 mm (4, 5); and 0.15 mm (8).

1 female (CHMLP coll. nr 5453); paratypes: 5 males and 5 females (CHMLP coll. nr 5454).

Prevalence: 76.5%.

Mean intensity  $\pm$  SD: 3.8  $\pm$  2.7 (1–10).

Etymology: The new species is named after our son Pedro Timi.

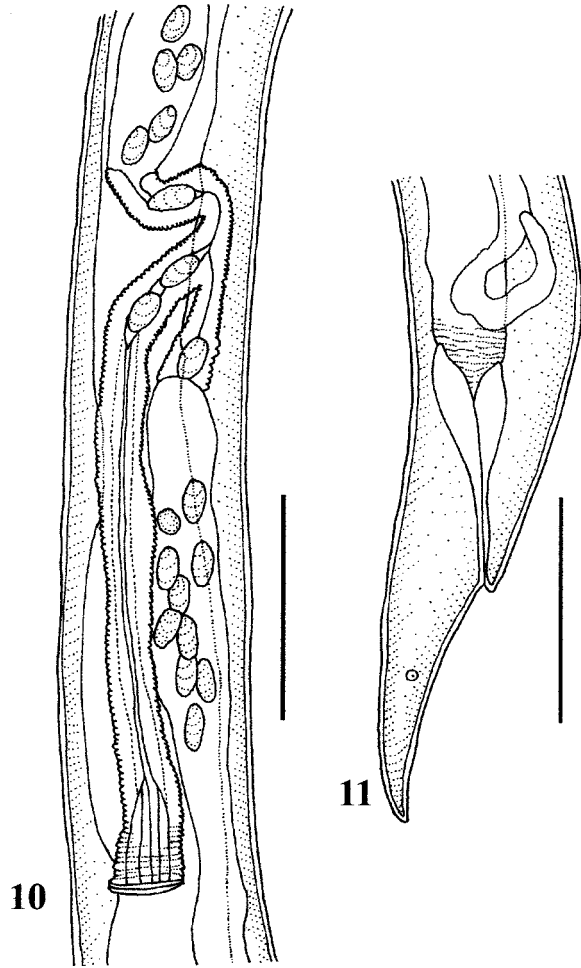
**Remarks**

In lacking lateral alae and in having a precloacal sucker, 11 pairs of caudal papillae, alate spicules, and a protuberant anus as well as similar general morphometric features, *Cucullanus pedroi* n. sp. most closely resembles *C. hians* (Dujardin, 1845) and the related species *Cucullanus australiensis* Baylis, 1927 and *Cucullanus filiformis* Yamaguti, 1935, all parasites of anguilliform fish (Campana-Rouget and Chabaud, 1956; Rasheed, 1966).

*Cucullanus hians* is a common parasite of the conger *Conger conger* (L.) from European and North African waters (Campana-Rouget and Chabaud, 1956; Campana-Rouget, 1957; Muñoz et al., 1988; Petter and Radujkovic, 1989; Saraiva et al., 2000); it differs from *C. pedroi* n. sp.

mainly by having pair 4 of male caudal papillae situated laterally between pairs 6 and 7, pair 8 in a lateral position (instead of subdorsal), and the phasmids displaced to the cloaca; *C. hians* also lacks the medial adcloacal papilla and has a longer tail (Campana Rouget and Chabaud, 1956, Plate 1, Figs. 3, 5; Saraiva et al., 2000). A description of extended spicules has not been provided in previous descriptions of *C. hians*; however, in the revision of Cucullanidae by Barreto (1922), broadly alate evaginate spicules, similar to those of the *C. pedroi* n. sp., are depicted for *C. praecinctus* (Drasche, 1882) from a conger from the Adriatic. This species was later regarded as a junior synonym of *C. hians* by Campana-Rouget and Chabaud (1956).

Rasheed (1966) described *C. hians* from 5 fish hosts belonging to 4 families, from Karachi, west Pakistan. These nematodes show similar general morphometric features to both those of *C. hians* from European waters and those of the new species. Nevertheless, their distribution pattern of caudal papillae in males differs from the new species by having the pair 4 situated laterally between pairs 6 and 7, pair 8 situated laterally between pairs 9 and 10, and ventral phasmids anterior to pair



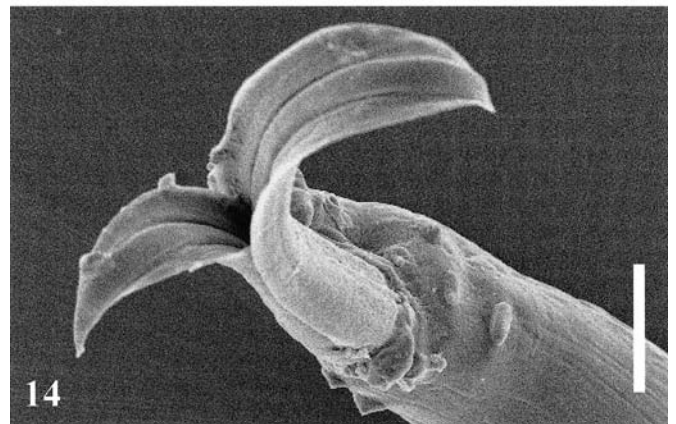
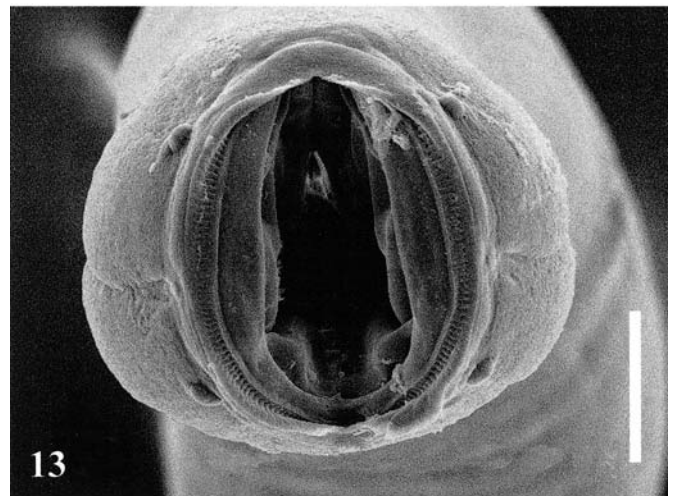
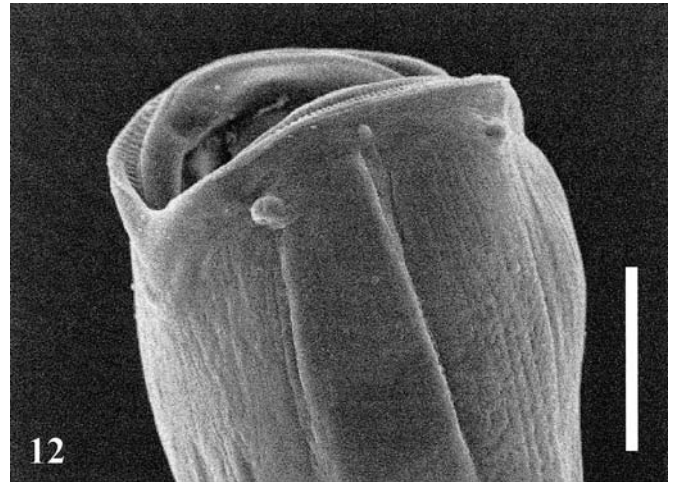
FIGURES 10–11. *C. pedroi* n. sp. **10.** Vulva and vagina, lateral view. **11.** Tail of female, lateral view. Bars = 0.4 mm.

9. The position of both pair 8 and phasmids also distinguishes specimens of *C. hians* from Pakistan and Europe. Therefore, the specific status of parasites described by Rasheed (1966) should be revised. Indeed, Petter and Le Bel (1992) considered the specimens described by Rasheed (1966) as a possible synonym of *C. bourdini* Petter and Le Bel, 1992, because in the tables of measurements the deirids are, generally, posterior or only slightly anterior to the distal end of esophagus.

Campana-Rouget (1957) considered *C. australiensis*, a parasite of a reef eel (*Gymnothorax pictus*) from Australia (Baylis, 1927) as a variety of *C. hians*, on the basis of slight differences in the location of nerve ring, cervical papillae, and slightly posterior position of 1 of the paraoccal papillae. Because of these minor differences, Rasheed (1966) synonymized both species; this author also regarded *C. filiformis*, a parasite of *Conger myriaster* (Brevoort, 1856) from Japan (Yamaguti, 1935), as a junior synonym of *C. hians*. However, Petter (1974), in a posterior revision of the family retained the 3 species as valid.

Indeed, males of *C. filiformis* display a pattern of caudal papillae similar to those of the new species, differing consequently from *C. hians*. However, *C. filiformis* can be distinguished from the new species by having a shorter esophagus, tail, spicules, and gubernaculum; smaller eggs; and the deirids situated at the end of esophagus (Yamaguti, 1935). Therefore, *C. filiformis* is considered as a distinct, valid species.

In contrast, the original description of *C. australiensis* by Baylis (1927) is brief and lacks a number of diagnostic features (e.g., esophagus width and the position of excretory pore and phasmids in males). Nevertheless, *C. australiensis* differs from *C. pedroi* n. sp. by having pair 4 of caudal papillae in males very large and pair 7 in preoccal



FIGURES 12–14. *C. pedroi* n. sp. **12.** Anterior end, lateral view. **13.** Anterior end, apical view. **14.)** Tail of male with evaginate spicules. Bars = 50  $\mu$ m (12, 13); and 100  $\mu$ m (14).

position as well as shorter esophagus and nerve ring and deirids located more anteriorly.

The new species also resembles *Cucullanus robustus* Yamaguti, 1935, another parasite of *C. myriaster* from Japan (Yamaguti, 1935). Nevertheless, both species can be distinguished by having *C. robustus* shorter esophagus and gubernaculum and wider body as well as pair 4 of caudal papillae situated at the level of pair 5 and pair 8 between pairs 9 and 10.

More recently, Morand and Rigby (1998) described a new species, *Cucullanus faliexae*, from another Anguilliform fish, *Gymnothorax javanicus* (Bleeker, 1859) (Muraenidae), from French Polynesia. This nematode also resembles *C. pedroi* n. sp., especially in the distribution pattern of caudal papillae in males. However, they can be distinguished by having smaller phasmids, shorter spicules and gubernaculum, shorter tail in males, and deirids and excretory pore located more anteriorly.

#### ACKNOWLEDGMENTS

We thank Roberto Mazzella and Hugo Mazzella from the fish market Albatros, Mar del Plata, for kindly providing fish samples and František Moravec, Academy of Sciences of the Czech Republic, for providing part of the bibliography. The present study was funded by grants from Universidad Nacional de Mar del Plata (15 E /146) and FONCYT (PICT 15192).

#### LITERATURE CITED

- BARRETO, A. L. 1922. Revisão da família Cucullanidae Barreto, 1916. (1). Memórias do Instituto Oswaldo Cruz **14**: 68–87.
- BAYLIS, H. A. 1927. Some new parasitic nematodes from Australia. Annals and Magazine of Natural History, sér. 9 **20**: 214–225.
- BUSH, A. O., K. D. LAFFERTY, J. M. LOTZ, AND A. W. SHOSTAK. 1997. Parasitology meets ecology on its own terms: Margolis et al. revisited. Journal of Parasitology **83**: 575–583.
- CAMPANA-ROUGET, Y. 1957. Parasites de poissons de mer puestafricans récoltés par J. Cadenat. Nématodes (4e note). Sur quelques espèces de Cucullanidae. Révision de la sous-famille. Bulletin de l'Institut français d'Afrique noire, sér. A **19**:
- , AND A.-G. CHABAUD. 1956. Helminthes des environs de banylus III. Sur trois espèces de *Cucullanus* (Camallanoidea, Nematoda) parasites des poissons. Vie et Milieu **7**: 267–279.
- DANIEL, V. I., J. T. TIMI, AND N. H. SARDELLA. 2002. *Cucullanus marplatensis* sp. nov. (Nematoda, Cucullanidae) parasitizing *Odontesthes argentinensis* (Valenciennes, 1835) (Pisces: Atherinidae) from Argentinean waters. Acta Parasitologica **47**: 41–46.
- HASEGAWA, H., E. H. WILLIAMS, JR., AND L. BUNKLEY-WILLIAMS. 1991. Nematode parasites from marine fishes of Okinawa, Japan. Journal of the Helminthological Society of Washington **58**: 186–197.
- LANFRANCHI, A. L., J. T. TIMI, AND N. H. SARDELLA. 2004. *Cucullanus bonaerensis* n. sp. (Nematoda: Cucullanidae) parasitizing *Urophycis brasiliensis* (Pisces: Phycidae) from Argentinean waters. Journal of Parasitology **90**: 808–812.
- MORAND, S., AND M. C. RIGBY. 1998. Cucullanin nematodes from coral reef fishes of French Polynesia, with a description of *Cucullanus faliexae* n. sp. (Nematoda: Chitwoodchabaudiidae). Journal of Parasitology, **84**: 1213–1217.
- MUÑOZ, M. V., J. P. FERNÁNDEZ, M. E. ORTS, AND E. CARBONELL. 1988. Sobre algunos nematodos cucullanidos parásitos de *Conger conger* (L.) en aguas del litoral valenciano. Revista Ibérica de Parasitología **48**: 275–281.
- PETTER, A. J. 1974. Essai de classification de la famille des Cucullanidae. Bulletin du Muséum national d'Histoire naturelle, 3° sér., **225**, Zoologie **117**: 1469–1490.
- , AND J. LE BEL. 1992. Two new species in the genus *Cucullanus* (Nematoda-Cucullanidae) from the Australian region. Memórias do Instituto Oswaldo Cruz **87**: 201–206.
- , AND B. M. RADUJKOVIC. 1989. Parasites des poissons marins du Montenegro: Nematodes. Acta Adriatica **30**: 195–236.
- RASHEED, S. 1966. The nematodes of the genus *Cucullanus* Mueller, 1777, from the marine fish of Karachi coast. Anales de la Escuela Nacional de Ciencias Biológicas, México **15**: 23–59.
- SARAIVA, A., C. CRUZ, M. J. SANTOS, AND S. FERREIRA. 2000. Nematode Parasites from the digestive tract of conger *Conger conger* L. from the northwest coast of the Iberian Peninsula. Bulletin of the European Association of Fish Pathologists **20**: 163–166.
- SARDELLA, N. H., G. T. NAVONE, AND J. T. TIMI. 1997. A new species of *Cucullanus* (Nematoda, Cucullanidae) parasite of *Genypterus blacodes* and *G. brasiliensis* (Pisces, Ophidiidae) in the South West Atlantic. Parasite **4**: 41–47.
- TANZOLA, R. D., AND S. E. GUAGLIARDO. 2000. Helminth fauna of the Argentine conger, *Conger orbignianus* (Pisces: Anguilliformes). Helminthologia **37**: 229–232.
- YAMAGUTI, S. 1935. Studies on the helminth fauna of Japan. Part 9. Nematodes of fishes, 1. Japanese Journal of Zoology **6**: 337–386.