


Description of a new species of *Engraulicola* George 1960 (Monogenea: Gastrocotylidae) parasitizing gills of *Chloroscombrus chrysurus* (Carangidae) from Veracruz, southern Gulf of Mexico

Descrição de uma nova espécie de *Engraulicola* George 1960 (Monogenea: Gastrocotylidae) parasito de brânquias de *Chloroscombrus chrysurus* (Carangidae) de Veracruz, Sul do Golfo do México

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Abstract

Engraulicola lamothei n. sp. (Monogenea: Gastrocotylidae) is described from the gills of *Chloroscombrus chrysurus* (Teleostei: Carangidae) from Veracruz, southern Gulf of Mexico. This monogenean is 1110-1670 (1328) μm long and 260-550 (363) μm wide. The haptor is asymmetrical, the long side has several clamps (35-42), and the short side has a single clamp; the posterior end has a small lappet armed with two dissimilar pairs of hooks. Male and female genital pores open near the anterior body end; the male genital pore is ventral, armed with a corona of 12 spines; the female pore is dorsal and unarmed. The new species differs from *Engraulicola forcipopenis* George, 1960 and from *Engraulicola micropharyngella* Unnithan, 1967 by lacking a pair of pointed forceps-like spines at the end of the penis amid the corona spines, and from *Engraulicola thrissocles* Tripathi, 1959 by the number of clamps on both sides, the number the hook pairs on the terminal lappet and the testis number. This is the first record of the genus *Engraulicola* George, 1960 on the Atlantic West Coast.

Keywords: *Engraulicola lamothei*, marine fishes, Monogenea, morphology, parasites, Gulf of Mexico.

Resumo

Engraulicola lamothei n. sp. (Monogenea: Gastrocotylidae) é descrito a partir das brânquias de *Chloroscombrus chrysurus* (Teleostei: Carangidae) de Veracruz, Sul do Golfo do México. Este monogêneo tem 1110-1670 (1328) μm de comprimento e 260-550 (363) μm de largura. O "haptor" é assimétrico, o lado longo tem vários grampos (35-42), e o lado curto tem um único grampo, e a extremidade posterior tem um pequeno "lappet" armado com dois pares de ganchos diferentes. Os poros genitais masculino e feminino são abertos perto da extremidade anterior do corpo; o poro genital masculino é ventral, armado com uma coroa de 12 espinhos; o poro feminino é dorsal e desarmado. A nova espécie difere de *Engraulicola forcipopenis* George, 1960 e de *Engraulicola micropharyngella* Unnithan, 1967, pela ausência de um par de fórceps pontiagudos como espinhos localizados na extremidade do pênis, em meio aos espinhos da coroa; e de *Engraulicola thrissocles* Tripathi, 1959, pelo número de pinças em ambos os lados, pelo número de pares de ganchos no "lappet" terminal, e pelo número de testículos. Este é o primeiro registro do gênero *Engraulicola* George, 1960 para a costa oeste do Atlântico.

Palavras-chave: *Engraulicola lamothei*, peixe marinho, Monogenea, morfologia, parasitos, Golfo do México.

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Introduction

During the helminthological analyses of Sciaenidae and Carangidae marine fishes off the coast of Veracruz, southern Gulf of Mexico, we collected ten monogeneans identified as *Engraulicola* cf. *thrissocles* (Tripathi, 1959), from *Chloroscombrus chrysurus* Linnaeus 1766 (Teleostei: Carangidae) (Montoya-Mendoza et al., 2008). Although the general morphology of our specimens closely resembled the species alluded to, other characteristics, further elaborated upon later, made us consider that they might be an as yet undescribed species in this genus. We performed another round of samplings between 2008 and 2012, aiming to collect additional specimens of this monogenean species; despite collecting only a few, we were able to confirm that it is indeed a new taxon, which we describe here. Besides our previous record, only one additional record of monogeneans in the Atlantic bumper (*C. chrysurus*) from the Gulf of Mexico is available. The parasite was *Amphipolycotyle chloroscombrus* Hargis, 1957, in spite of this, Yamaguti (1963) proposed the synonymy of *Engraulicola* and *Amphipolycotyle*, although *A. chloroscombrus* is still referred to as a valid species and has been recorded in the general geographic area of our specimen collection (Bravo-Hollis, 1984; Costa & Chellappa, 2016). Currently, the genus *Engraulicola* George, 1960 (Monogenea: Gastrocotylidae: Gastrocotylinae) includes three valid species parasitizing Engraulidae fishes from the Indo-Pacific: *E. forcipopenis* George, 1960, *E. micropharyngella* Unnithan, 1967, and *E. thrissocles* (Tripathi, 1959) Lebedev, 1971 (George, 1960; Unnithan, 1967; Lebedev, 1972, 1986). In this study we describe the fourth species in this genus and provide prevalence and infection intensity data in *C. chrysurus* in the southern Gulf of Mexico.

Materials and Methods

We examined ten *C. chrysurus* specimens in October 2008 and a further ten in March 2012. All specimens were captured off Las Barrancas beach (18°59'31"N, 95°57'83"W), Alvarado, Veracruz, Mexico, in the southern Gulf of Mexico. Captured fish were transported live to the laboratory and examined during the subsequent 24 hours. We examined gills, opercula, nostrils, skin, and fins under a stereo microscope. Monogeneans were fixed in hot 4% formalin and stored in 70% ethanol. The specimens were later stained with Mayer's paracarmine and Gomori's triple stain, gradually dehydrated in ethanol, cleared in clove oil, and mounted as whole specimens.

Some monogenean specimens were mounted in temporary preparations using ammonium picrate to examine sclerotized structures (Vidal-Martínez et al., 2001). Line drawings were made using an Olympus BX51 compound microscope and a camera lucida. Voucher specimens of Montoya-Mendoza et al. (2008), deposited in the Colección Nacional de Helmintos (CNHE; CNHE-6197, four specimens), were re-examined. Measures are given in micrometers with the range followed by the average in parentheses. We calculated the prevalence (percentage of infected hosts) and mean intensity (mean number of monogeneans per host), according to Bush et al. (1997). The holotype (CNHE no. 11439) and the paratype specimens (n = 9; CNHE no. 11440, 11441) were deposited in the Colección Nacional de Helmintos (CNHE), Instituto de Biología, Universidad Nacional Autónoma de México, Mexico City, Mexico.

Results

Class: Monogenea van Beneden, 1858

Order: Mazocraeidea Price, 1936

Family: Gastrocotylidae Price, 1943

Genus: *Engraulicola* George, 1960

Engraulicola lamothei n. sp. (Figures 1-2).

Description (based on ten specimens). Lanceolate body narrowing toward the anterior end, length 1110–1670 (1328) μm from apex to posterior end; maximum width at ovary level 260–550 (363) μm . Anterior end rounded (Figure 1). Smooth tegument. Lacking eyespots. Terminal mouth. With oral spheroid-shaped suckers anterior and lateral to the pharynx; left sucker 22–25 (24) μm long, 25–30 μm (27) wide; right sucker 22–27 (25) μm long, 25–27 (26) μm wide.

Ovoid pharynx 25–37 (31) μm x 30–37 (35) μm . Simple esophagus, long and narrow, without ramifications; intestinal crura anterior to the male genital pore. Caeca simple, terminating near the posterior end of the body, not confluent. Asymmetric haptor, with a long row of 32–42 clamps on one side and a single clamp on the opposite side; the longest part measures 740–1050 (897) μm , extending as an adhered lateral expansion over ~50% of the body length. Amphipathic haptor; when the parasite was found in the host's right gill chamber, the long part of

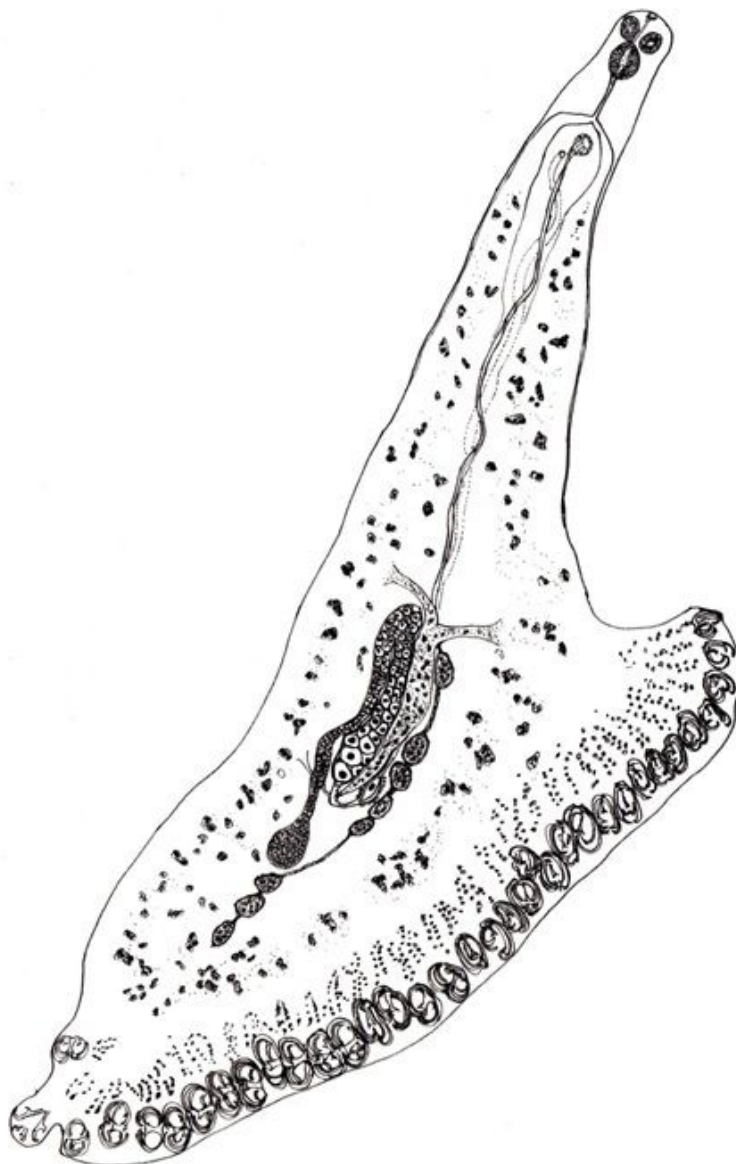
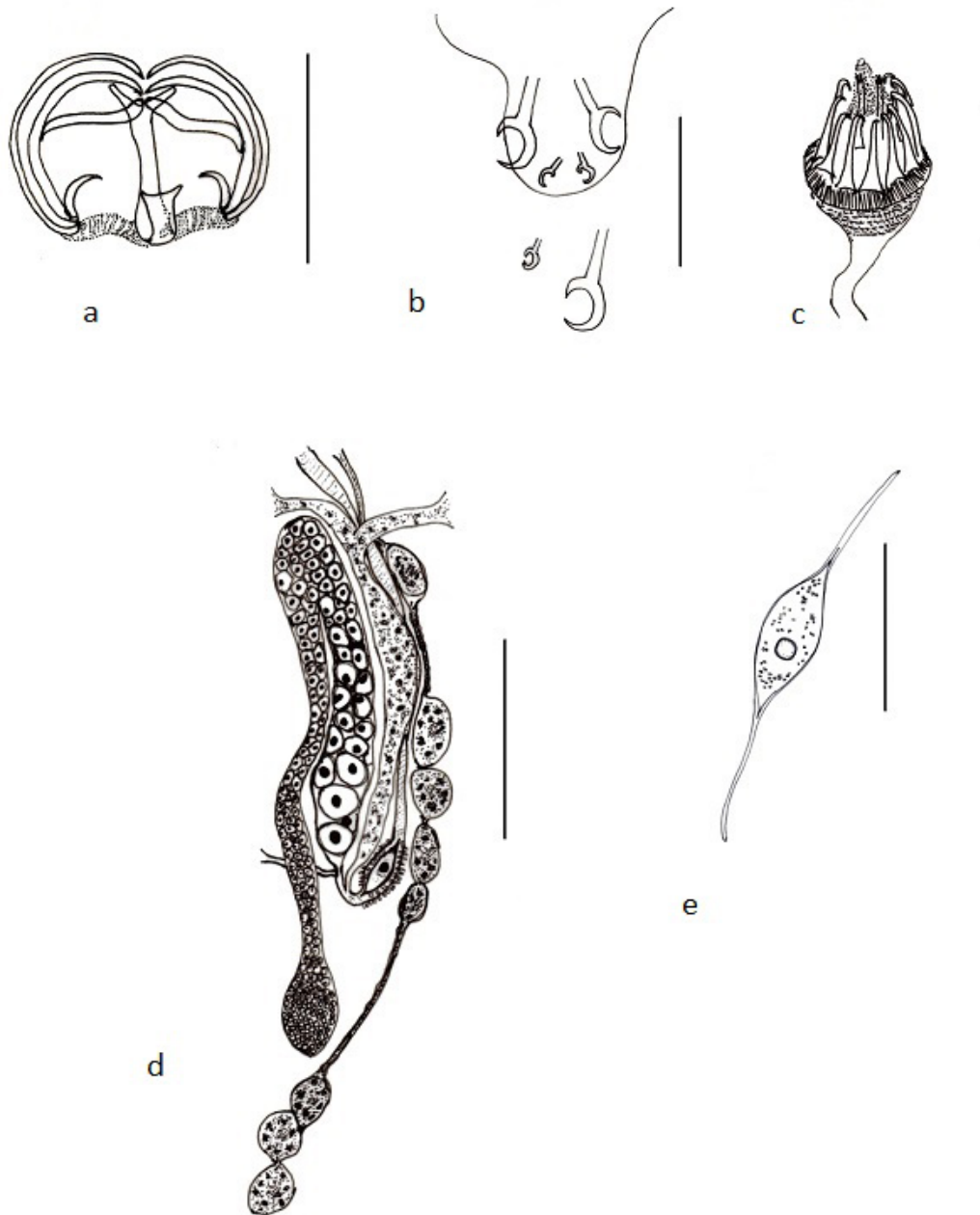


Figure 1. *Engraulicola lamothei* n. sp. from *Chloroscombrus chrysurus*. Entire worm, dorsal view; holotype (CNHE 11439).

the haptor was at the right side; the opposite occurred when the parasite was in the left gill chamber. All clamps are similar in shape with small variations in size (Figure 2a). The distal clamps measure 25–30 (27) μm long x 30–42 (39) μm wide; medium clamps 27–35 (30) μm x 37–50 (44) μm ; proximal clamps 20–30 (25) μm x 25–37 (32) μm ; the single clamp of the short side of the haptor is 18–35 (25) long x 27–42 (34) wide. The clamps present the typical structure of the Gastrocotyloidea family with a very short dorsal arm in the median spring and a V-shaped distal end; the ventral arm of the median spring is long and thin, with a bifurcated distal end extending to the superior level of the jaw sclerites; the base of each clamp shows a cuticularized ligament at both sides; the ventral and dorsal jaws are symmetrical; the dorsal arm of the ventral jaw extends to the terminal level of the dorsal arm of the median jaw; the oblique jaw sclerites are long and thin, with the distal end in contact with the median line (Figure 2a). The terminal lappet is trapezoid in shape, measuring 35–50 (43) μm x 30–40 (34) μm with two pairs of hooks; the proximal pair is larger, measuring 24–28 (27) μm , and the distal pair is smaller, measuring 8–10 (9) μm (Figure 2b). Spheroid testes, 7–10 (8), arranged in a row, forming a string running more or less parallel to the anteroposterior axis of the body (Figures 1, 2d); the proximal testes measure 25–30 (26) μm long x 25–50 (33) μm wide; the distal testes measure 25–35 (28) μm x 27–40 (31) μm . Straight *Vas deferens*, opening at the base of the male copulating organ. Median genital atrium, located ventrally, posterior to the caecal bifurcation, opening at

150–200 (168) μm from the anterior end of the body. The penis is a bulbous muscle crowned with 12 spines with converging uncinated tips; spines measure 11–18 (13) μm long x 13–21 (16) μm wide (Figure 2c). The ovary is an elongated, inverted U-shaped, located almost symmetrically to the testes; the ovary has a short oviduct that in



Figures 2. *Engraulicola lamothei* n. sp. from *Chloroscombrus chrysurus*. Figure 2a. Clamps. Figure 2b. Hook. Figure 2c. Male copulatory organ. Figure 2d. Ovarian region. Figure 2e. Eggs. Scale bars: Figure 1 - 200 μm . Figure 2a, 2b, 2e - 50 μm ; Figure 2c - 20 μm ; Figure 2d - 100 μm .

turn opens into the ootype. The uterus originates from the ootype, extending upwards and opening in proximity to the male genital pore (Figure 1). The vitelline glands extend through the lateral fields from the caecal branching zone almost to the terminal end of the body, converging toward the final region of the intestinal caeca. The vitelline receptacle is conspicuous, V-shaped, located between the ovary and the string of testes. Unarticulated vaginal pore, opening in the dorsal surface posterior to the genital atrium. The genito-intestinal canal connects the ootype with an intestinal caecum (Figure 2d). The eggs are elliptical in shape, measuring 157–166 (161) μm x 45–50 (47) μm with two short polar filaments (Figure 2e).

Locality: Off Las Barrancas beach (18°59'31"N, 95°57'83"W) Alvarado, Veracruz, Mexico, southern Gulf of Mexico.

Site of infection: gills.

Dates of collection: October 2008 and March 2012.

Prevalence, 40% (4/10 hosts), 7 monogeneans collected, mean intensity 1.75, range 1-3 (October 2008); 20% (2/10 hosts), 3 monogeneans collected, mean intensity 1.5, range 1-2 (March 2012).

Etymology: this species is named in honor of the late Mexican helminthologist Dr. Marcos Rafael Lamothe y Argumedo (1932-2013), Instituto de Biología, Universidad Nacional Autónoma de México, México.

Remarks

The analyzed material shows the identifying characteristics of the genus *Engraulicola* according to George (1960) and Lebedev (1972, 1986), including: the presence of an asymmetrical haptor with one well-developed side with numerous clamps and an opposite sub-developed side with a single clamp; a terminal caudal lappet in the shape of a small digitiform projection; two pairs of differently sized hooks; clamps are long rather than wide; testes as rounded follicles, arranged in a row forming a string; vagina dorsal and unarticulated; eggs possessing polar filaments.

The new species, *Engraulicola lamothei*, is more similar to *E. thrissocles*, given a number of characteristics, including: general anatomical structure, the lack of forceps-like spines in the spine crown surrounding the penis, the number of spines constituting this crown, the shape of the clamps in the haptor and the possession of an inverted U-shaped ovary (see Tripathi, 1959). However, it can be distinguished from the latter species by the total number of clamps in the haptor, 33–43/1 in the new species compared with 46–59/1–2 in *E. thrissocles*; the caudal lappet is smaller (30–40 μm x 35–50 μm) and has two pairs of hooks in the new species, whereas in *E. thrissocles* it is larger (75 μm x 77 μm), with three pairs of hooks. Moreover, *E. lamothei* has fewer testes (7 a 10) than *E. thrissocles* (20–40); the proximal clamps are smaller in *E. lamothei* (25–30 μm x 30–42 μm) compared to *E. thrissocles* (43–52 μm x 39–60 μm), and *E. lamothei* n. sp. is smaller overall (1110–1670 μm) than *E. thrissocles* (2600–3500 μm) (see Table 1).

The other two species of *Engraulicola*, *E. forcipopenis* and *E. micropharyngella*, show a characteristic pair of forceps-like spines emerging from the central part of the spine crown surrounding the penis, making them different from the new species (George, 1960; Unnithan, 1967). Furthermore, *E. lamothei* n. sp. possesses fewer testes (8–10) than *E. forcipopenis* (14–18), and its eggs are smaller (157–166 μm x 45–50 μm) than those of *E. forcipopenis* (168–210 μm x 50–84 μm). The pharynx of *E. lamothei* n. sp. is larger (25–37 μm x 30–37 μm) than that of *E. micropharyngella* (14); the number of spines in the crown of the penis (12), as well as the number of clamps in the haptor (33–43), is also greater in *E. lamothei* n. sp. than in *E. micropharyngella* (8–10 spines; 27 clamps), whereas the number of testes (7–10) is fewer in the new species than in *E. micropharyngella* (27 testes). Finally, the size of the eggs varies widely, 157–166 μm x 45–50 μm in the new species compared with 24 μm x 40 μm in *E. micropharyngella* (Table 1).

The proposed new species *E. lamothei* is similar to the currently valid species *Amphipolycotyle chloroscombrus* Hargis, 1957 because of the presence of a terminal lappet and larval hooks and because both species have a genital pore surrounded by a crown of 12 hooks. However, these species can be differentiated by the number of clamps (33–43/1 in *E. lamothei* n. sp. vs 15–18 on the long side, plus 9–16 on the short side in *A. chloroscombrus*); the number and length of the larval hooks on the terminal lappet (2 pairs in *E. lamothei* n. sp., the proximal 24–28 μm and the posterior pair 8–10 μm , compared to 3 pairs of larval hooks in *A. chloroscombrus*, the anterior pair 34–36 μm in length, the medium pair 24 μm and the posterior ones 20 μm); and the number of testes (7 to 10 in *E. lamothei* n. sp. and 2 in *A. chloroscombrus*).

Table 1. Comparative morphological characteristics of the species of the genus *Engraulicola*; measurements are presented as length (l) x width (w) in micrometers.

	<i>E. forcipopenis</i>	<i>E. micropharyngella</i>	<i>E. thrissocles</i>	<i>E. lamothei</i> n. sp.
Hosts	<i>Anchoviella bataviensis</i>	<i>Anchoviella commersonii</i>	<i>Thryssa mystax</i>	<i>Chloroscombrus chrysurus</i>
Location	India, Arabian Sea, NW Indian Ocean	India, Arabian Sea, NW Indian Ocean	India, NW Indian Ocean, Vietnam, NW Pacific	Mexico, Gulf of Mexico, NW Atlantic Ocean
Length	1900-2200	1300-1900	2600-3500	1110-1670
Width	520-820	420-640	370-380	260-550
Number of clamps	25-44 / 1	27/jan	46-59 / 1-2	35-42 / 1
Proximal (l x w)	21-33 x 33-50	16 x 20	43-52 x 39-60	20-30 x 25-37
Medial	21-42 x 50-84	28 x 40		27 x 35 x 37-50
Opposite	25-33 x 30-42	25-33 x 30-42	26-36 x 34-45	18-35 x 27-42
Oral suckers (l x w)	25-37 x 37-50	16-20	19	22-27 x 25-30
Pharynx (l x w)	38-42 x 50-59	14	26 x 45	25-37 x 30-37
Number corona spines	12	08/oct	12	12
Diameter corona spines	16-18	-	-	13-21
Terminal lappet	--	36 x 21	75 x 77	43 x 34
Pairs hooks	2	2	3	2
Proximal	25.4-29.2	24	35	24-28
Medial	12.6	8	14	08/oct
Distal	-	-	02/apr	-
Number of testes	14-18	27	20-40	07/oct
Eggs	168-210 x 50-84	40 x 24	-	157-166 x 45-50
References	George (1960)	Unnithan (1967)	Lebedev (1972)	Present study

Discussion

The study of additional specimens allowed us to confirm that the specimens, previously reported by us as *E. cf. trissocles* (CNHE 6197) (Montoya-Mendoza et al., 2008), are indeed a new species currently identified as *Engraulicola lamothei* n. sp. This new species is morphologically well differentiated from the three previously known species of *Engraulicola* and also from the currently valid species *A. chloroscombrus*; furthermore, we include a key to species in *Engraulicola* to differentiate between these species.

The three previously known species of the genus *Engraulicola* are parasites of the fish family Engraulidae in the Indo-Pacific (Table 1). The data hereby demonstrate that this monogenean group can be found parasitizing other fish families, such as Carangidae in this study, even in locations far from the Indo-Pacific such as the Gulf of Mexico. This discontinuity of geographical distribution is remarkable; however, engraulid fish are widely distributed, and the data presented here suggest that additional *Engraulicola* species might be encountered in other fish families. Thus, the real geographical distribution of this group of monogeneans is far from completely known, as is also true for most parasites. Therefore, additional sampling is needed to understand the question posed by our current report.

The present finding of a new species of *Engraulicola* parasitizing marine carangid fish in the southern Gulf of Mexico highlights the discussion regarding the supposed synonymy of *Engraulicola* and *Amphipolycotyle* (Yamaguti, 1963). From our point of view, and as far as the present comparison between structural characteristics of our species versus those of *A. chloroscombrus*, there are sufficient differences between both genera, including the number of clamps and larval hooks on lappet and the number of testes. These differences allow us to consider them different genera.

Key to species of *Engraulicola*

- 1 Pair of forceps-like spines emerging from the central part of male copulatory organ 2
- No forceps-like spines from the central part of male copulatory organ 3
- 2 With 8 -10 spines in the genital corona; testes, 27; clamps, 27-32/1... *E. micropharyngella*
- With 12 spines in the genital corona; testes, 14-18; clamps, 33-44/1 *E. forcipopenis*
- 3 Three pairs of hooks on the caudal lappet; testes, 20-40 *E. thrissocles*
- Two pairs of hooks on the caudal lappet; testes, 7-10 *E. lamothei* n. sp.

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