

# A new species of *Acanthobothrium* (Eucestoda: Onchobothriidae) in *Aetobatus* cf. *narinari* (Myliobatidae) from Campeche, México

Uma nova espécie de *Acanthobothrium* (Eucestoda: Onchobothriidae) em  
*Aetobatus* cf. *narinari* (Myliobatidae) de Campeche, México

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## Abstract

The helminthological examination of nine individuals of *Aetobatus* cf. *narinari* (spotted eagle ray; raya pinta; arraia pintada) revealed the presence of an undescribed species of cestode of the genus *Acanthobothrium*. The stingrays were collected from four locations in México: Laguna Términos, south of Isla del Carmen and the marine waters north of Isla del Carmen and Champotón, in the State of Campeche, and Isla Holbox, State of Quintana Roo. The new species, nominated *Acanthobothrium marquesi*, is a category 3 species (i.e. the strobila is long, has more than 50 proglottids, the numerous testicles greater than 80, and has asymmetrically-lobed ovaries); at the present, the only category 3 species that has been reported in the Western Atlantic Ocean is *Acanthobothrium tortum*. *Acanthobothrium marquesi* n. sp. can be distinguished from *A. tortum* by length (26.1 cm vs. 10.6 cm), greater number of proglottids (1,549 vs. 656), a larger scolex (707 µm long by 872 µm wide vs. 699 µm long by 665 µm wide), larger bothridia (626 µm long by 274 µm wide vs. 563 µm long by 238 µm wide). This is the first report of a species of *Acanthobothrium* from the Mexican coast of the Gulf México.

**Keywords:** Onchobothriidae, *Acanthobothrium*, Chondrichthyes, Myliobatiformes, helminth, México.

## Resumo

O exame helmintológico do trato digestivo de nove espécimes de *Aetobatus* cf. *narinari* (arraia pintada) revelou a presença de uma nova espécie de cestódeo do gênero *Acanthobothrium*. As arraia foram coletadas de quatro locais no México: Laguna Términos, ao sul de Isla del Carmen e nas águas marinhas ao norte de Isla del Carmen e Champotón, no estado de Campeche, e Isla Holbox, estado de Quintana Roo. A nova espécie foi denominada *Acanthobothrium marquesi*, pertencente a uma espécie da categoria 3 (estrobilo longo, tendo mais de 50 proglotes, numerosos testículos, superiores a 80, e ovários assimetricamente lobados). Apenas outra espécie deste gênero, *Acanthobothrium tortum* pertence a categoria 3, no Oceano Atlântico Ocidental. *Acanthobothrium marquesi* n. sp. se distingue de *A. tortum* por ser mais longo (26,1 cm vs. 10,6 cm), possuir maior número de proglotes (1.549 vs. 656), ter um escolex maior (707 µm de comprimento por 872 µm de largura vs. 699 µm de comprimento por 666 µm de largura), e botridias maiores (626 µm de comprimento por 274 µm de largura vs. 563 µm de comprimento por 238 µm de largura). Este é o primeiro relato de uma espécie de *Acanthobothrium* do Golfo do México.

**Palavras-chave:** Onchobothriidae, *Acanthobothrium*, Chondrichthyes, Myliobatiformes, helminto, México.

## Introduction

*Acanthobothrium* is one of the richest genera within Onchoproteocephalidea, currently comprised of more than 188 nominal species (CAIRA & JENSEN, 2017). Only a few occurrences

have been documented in México, and most for the Pacific coast. In the first report of this genus in Mexican waters, Monks et al. (1996) described *A. cleofanus* Monks, Brooks, and Pérez-Ponce de León, 1996, from *Hypanus longus* (Garman, 1880). In three studies derived from the same project, Ghoshroy & Caira (2001), Caira & Burge (2001) and Caira & Zahner (2001) described

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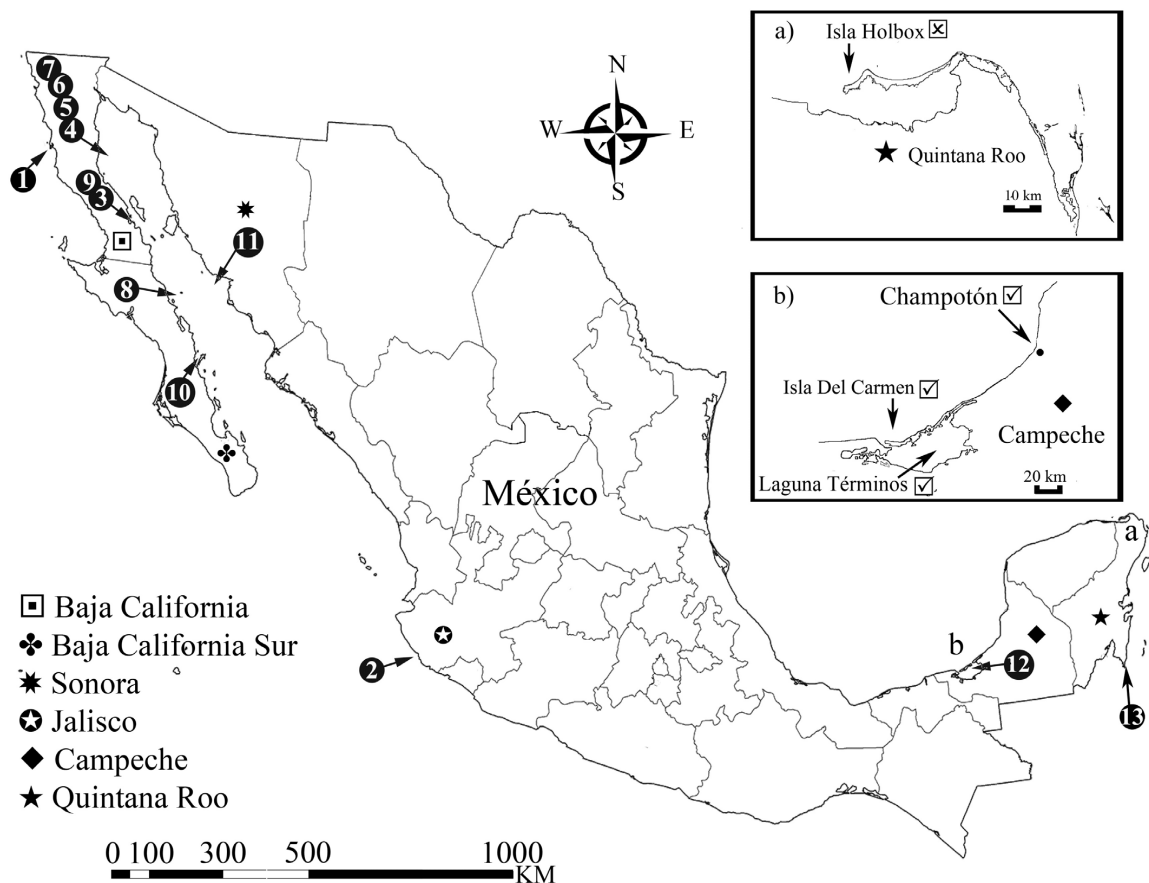


nine species, all from the Gulf of California (Figure 1; Table 1). Only *A. cartegenensis* Brooks and Mayes, 1980 has been reported from the eastern coast of México (BROOKS & MAYES, 1980; MONKS et al., 2015). To date, no members of the genus have been reported from the Mexican coasts of Gulf of Mexico, but Linton (1908), Goldstein (1964), Goldstein et al. (1969) and Vardo-Zalik & Campbell (2011) collectively have reported nine species of *Acanthobothrium* from the northern coast of the Gulf of Mexico in waters of the USA. This suggests that members of the genus likely occur in more southern parts of the Gulf. As part of a parasitological survey of elasmobranchs in the Gulf of Mexico, adult cestodes were found in the spiral valve of *Aetobatus cf. narinari* (Euphrasen, 1790) that could be assigned to *Acanthobothrium* but not to any known species; this species is described herein. It represents the first record of the genus from the Mexican coast of the Gulf of Mexico.

## Materials and Methods

From 1999 to 2005 the digestive tracts of nine stingrays, *Aetobatus cf. narinari* (Euphrasen, 1790) (spotted eagle ray; raya pinta; arraia pintada) were purchased from local fishermen at four

localities: five rays from Isla del Carmen, Campeche (three from open water North of Ciudad del Carmen and two rays from Laguna de Términos) (18°36' N, 91°33' W and 18°37'58"N; 91°49'57"W, respectively) (2000 and 2005); three specimens from Champotón, Campeche, (19°21'N; 90°54'W) (1999); and one specimen from Holbox, Quintana Roo, northwest of the island, (21°34'N; 86°14'W) (2000) (Insets a and b in Figure 1, Table 2). Cestodes were removed from the spiral valve of the host, killed with hot tap water, transferred immediately to AFA (alcohol-formalin-acetic acid) for 24-48 hr, and then stored in 70°GL ethanol. Specimens were stained either with Mayer's carmalum or Ehrlich's hematoxylin and mounted in Canada balsam for examination as whole mounts; worms that were too large to be mounted on a single slide were cut into sections and mounted sequentially on separate slides. Stained specimens were examined using a compound photomicroscope equipped with Nomarski differential interference contrast optics (Leica DM LB2). Taxonomic information for the host was taken from McEachran & Dunn (1998), White et al. (2010), Richards et al. (2009), Froese & Pauly (2017). Some specimens were prepared for scanning electron microscopy (SEM) by dehydration in a graded ethanol series and low-vacuum dried. Dried specimens were mounted on aluminum stubs with carbon tape and



**Figure 1.** The type localities of the species of *Acanthobothrium* reported from the coasts of Mexico (see Table 1); a and b refers to the sampling areas reported in this study, where ☐ is assigned to individuals of *Aetobatus cf. narinari* negative for *A. marquesi* n. sp. and ☑ for individuals of *Aetobatus cf. narinari* that were positive for the new species.

**Table 1.** Species of *Acanthobothrium* reported from the Eastern Pacific Ocean and from the Western Atlantic Ocean of the Americas (amphi-American species). Numbers in parentheses for each taxon name refer to localities in Figure 1.

Host species	<i>Acanthobothrium</i> spp.	Type locality	Source	Category designation
<b><i>Acanthobothrium</i> spp. reported from the eastern Pacific Ocean</b>				
<i>Dasyatis</i>				
<i>D. brevis</i>	<i>A. bullardi</i> Ghoshroy and Caira, 2001 (3*)	Bahía de Los Ángeles, Gulf of California, México	Ghoshroy & Caira (2001)	2
<i>D. brevis</i>	<i>A. dasi</i> Ghoshroy and Caira, 2001 (4)	Puertecitos, Gulf of California, México	Ghoshroy & Caira (2001)	2
<i>D. brevis</i>	<i>A. rajivi</i> Ghoshroy and Caira, 2001 (5)	Puertecitos, Gulf of California, México	Ghoshroy & Caira (2001)	2
<i>D. brevis</i>	<i>A. soberoni</i> Ghoshroy and Caira, 2001 (6)	Puertecitos, Gulf of California, México	Ghoshroy & Caira (2001)	6
<i>Hypanus</i>				
<i>H. longus</i>	<i>A. cleofanus</i> Monks, Brooks, and Pérez-Ponce de León, 1996 (2)	Chamela Bay, México	Monks et al. (1996)	3
<i>Diplobatis</i>				
<i>D. ommata</i>	<i>A. dolhya</i> Caira and Burge, 2001 (9)	Bahía de Los Ángeles, Gulf of California, México	Caira & Burge (2001)	1
<i>D. ommata</i>	<i>A. maryanskii</i> Caira and Burge, 2001 (10)	Loreto, Gulf of California, México	Caira & Burge (2001)	5
<i>D. ommata</i>	<i>A. royi</i> Caira and Burge, 2001 (11)	Punta Arena, Gulf of California, México	Caira & Burge (2001)	1
<i>Heterodontus</i>				
<i>H. francisci</i>	<i>A. bajaensis</i> Appy and Dailey, 1973 (1)	Bahía de San Quintin, México	Appy & Dailey (1973)	4
<i>H. francisci</i>	<i>A. puertecitense</i> Caira and Zahner, 2001 (7)	Puertecitos, Gulf of California, México	Caira & Zahner (2001)	4
<i>H. mexicanus</i>	<i>A. santarosaliense</i> Caira and Zahner, 2001 (8)	Santa Rosalia, Gulf of California, México	Caira & Zahner (2001)	3
<b><i>Acanthobothrium</i> spp. reported from the western Atlantic Ocean</b>				
<i>Aetobatus</i>				
<i>A.cf. narinari</i>	<i>A. marquesi</i> n. sp. (12)	Laguna de Términos, Gulf of México, México	This study	3
<i>Urobatis</i>				
<i>U. jamaicensis</i>	<i>A. cartagenensis</i> Brooks and Mayes, 1980 (13)	Xcalak, Quintana Roo, Caribbean Sea, México	Brooks & Mayes (1980), Monks et al. (2015)	9

\*Reference number for locations on the map (see Figure 1).

grounded with carbon paint before being sputter-coated with approximately 100 Å of gold/palladium (about two minutes). Specimens were examined using a Jeol JSM-6300 scanning electron microscope. Illustrations were made with the aid of a drawing tube. Measurements are in micrometers unless specified otherwise, and expressed as length by width. For most characters, ranges are given, followed in parentheses by the sample mean and sample size (n). Mean values  $\pm$  1 standard deviation is provided for some characters (i.e., mean  $\pm$  sd; n = sample size). Hook measurements follow Euzet (1959) as modified by Monks et al. (1996) (i.e., mean value  $\pm$  1 standard deviation is given followed by, in parentheses, the range).

The categorical method suggested by Ghoshroy & Caira (2001) and by Fyler & Caira (2006) was used to facilitate comparisons among species from the same geographic area in conjunction with the current literature. This method was proposed for the

comparison and differentiation of species by categories based upon the combination of four variables: total length  $\leq$  15 mm = S (short) or  $>$  15 mm = L (long); number of proglottids comprising the strobili  $\leq$  50 = F (few) or  $>$  50 = M (many); number of testes per proglottid  $\leq$  80 = F (few) or  $>$  80 = M (many); and ovarian lobes symmetrical = S or asymmetrical = A (GHOSHROY & CAIRA, 2001). Of the possible combinations the following 10 categories currently are recognized and coded as follows: 1 = SFFS; 2 = SFFA; 3 = LMMA; 4 = LMMS; 5 = LMFS; 6 = LMFA; 7 = LFFA; 8 = SMFS; 9 = LFFS; 10 = SMMS.

Specimens from three collections were examined: CNHE refers to the Colección Nacional de Helmintos, Instituto de Biología, Universidad Nacional Autónoma de México, Mexico City, México; CHE refers to the Colección de Helmintos, Centro de Investigaciones Biológicas, Universidad Autónoma del Estado de Hidalgo, Pachuca, México; and HWML refers to the Harold W.

Manter Laboratory of Parasitology, University of Nebraska-Lincoln, Lincoln, Nebraska, USA.

## Results

### Survey data

Nine stingrays, all large adults, were collected from the four localities (Figure 1, insets a and b). Of these, two of three rays from Champotón (in 1999) were infected with two and six individuals of *Acanthobothrium marquesi* n. sp., two rays from south of Isla del Carmen in Laguna de Términos (collected in 2005) were

infected with 10 and 12 worms, and one ray from north of Isla del Carmen was infected with 64 worms (Table 2). One ray collected in 2000 from Isla Holbox and two rays from the marine waters north of Isla del Carmen (2005) were uninfected.

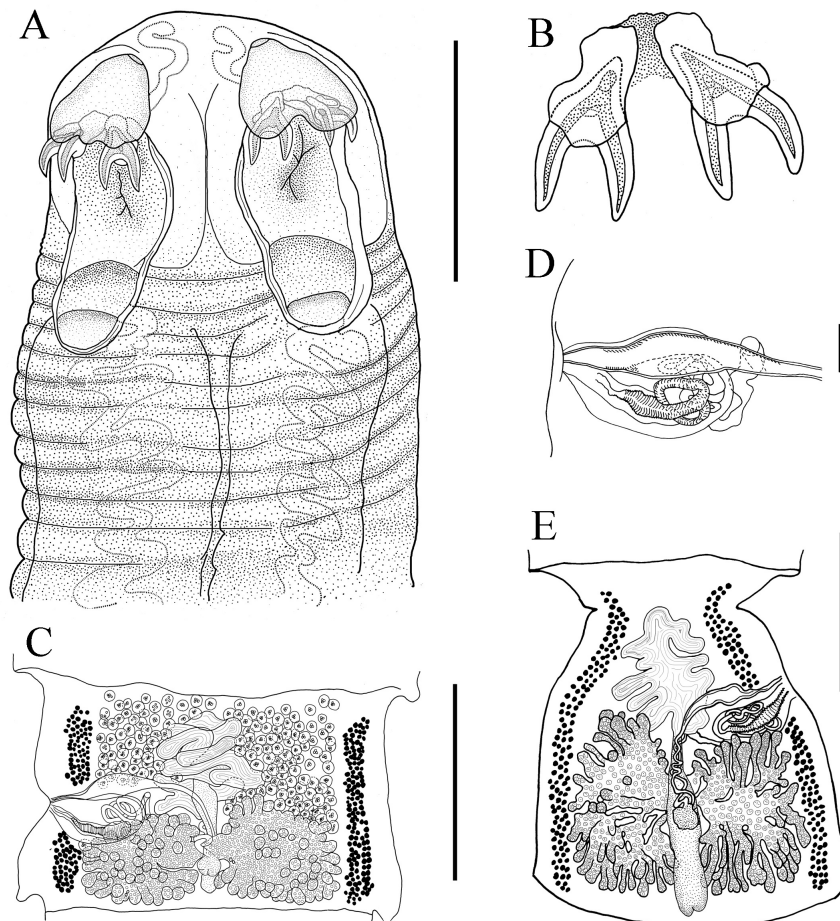
### Description

*Acanthobothrium marquesi* n. sp.  
(Figures 2-4)

*Description:* Based upon measurements of 10 complete worms and 20 partial worms mounted on slides, and two scoleces mounted on stubs for SEM. Strobila craspedote, anapolytic,

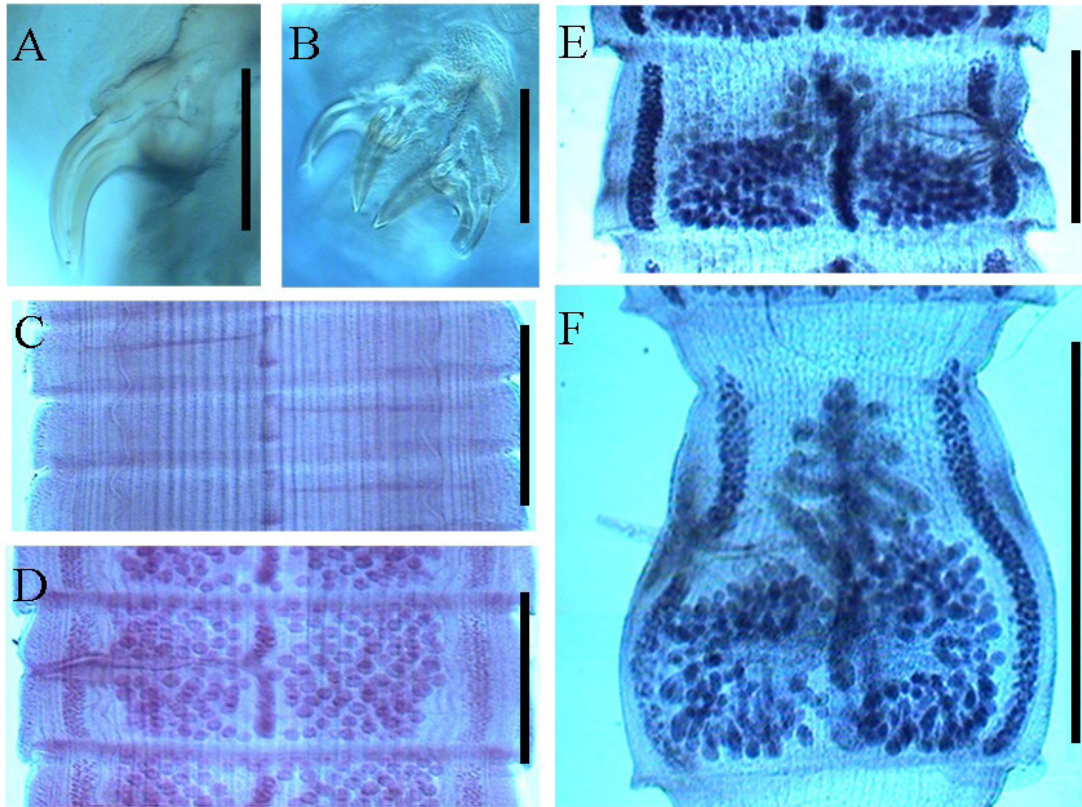
**Table 2.** Localities where individuals of *Aetobatus* cf. *narinari* were collected for this study.

State	Location	No. of rays collected	Geographic coordinates	Infection parameters of <i>Acanthobothrium marquesi</i> n. sp.
Quintana Roo	Isla Holbox (Open water North of island)	1	21°34'N; 86°14'W	☒
Campeche	Isla del Carmen (Open water North of Ciudad del Carmen)	3	18°83' N, 91°49' W	☒, ☑ (28), ☒
	Campotón (Open water North of Ciudad)	3	19°21' N, 90°54' W	☑ (2), ☑ (4), ☒
	Laguna Términos (South of Ciudad del Carmen and center of lagoon)	2	18°36' N, 91°33' W	☑ (64), ☑ (12)

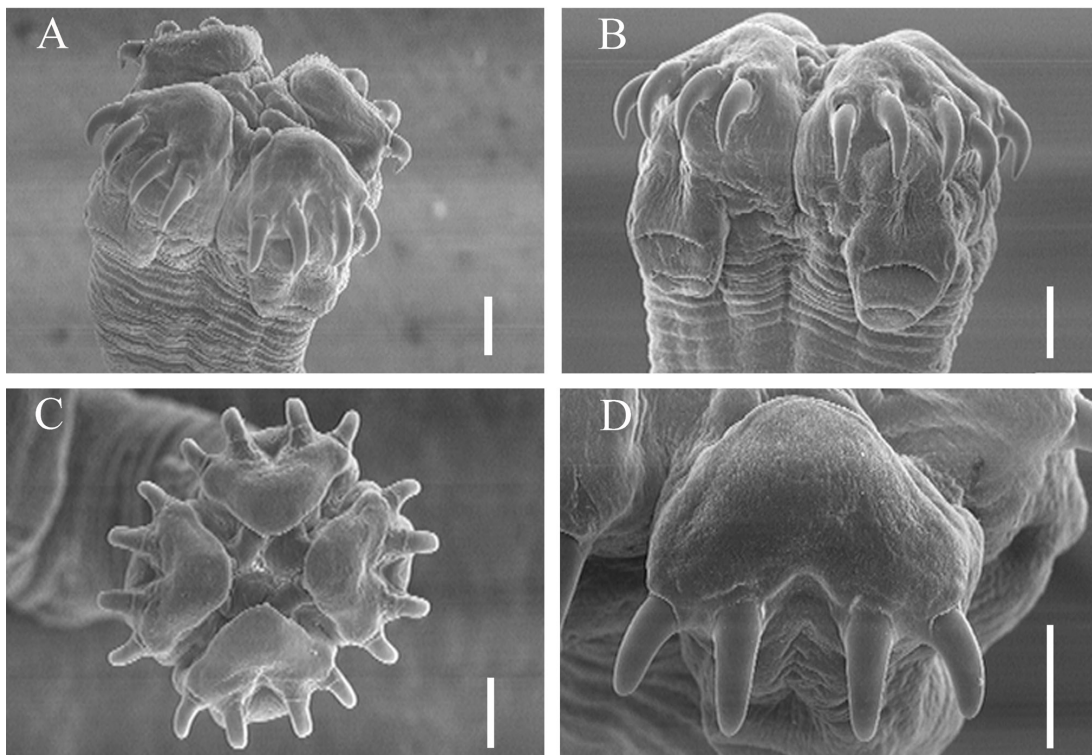


**Figure 2.** *Acanthobothrium marquesi* n. sp. A. Scolex. B. Hooks. C. Mature proglotid. D. Cirrus sack. E. Terminal proglotid. Scale bar: A. 750  $\mu$ m; B. 150  $\mu$ m; C. 1000  $\mu$ m; D. 133  $\mu$ m; E. 880  $\mu$ m.





**Figure 3.** Photographic images of *Acanthobothrium marquesi* n. sp. from *Aetobatus cf. narinari* taken using a compound microscope equipped with normal light and Nomarski differential optics. A. Abaxial view of hook (lateral prong). B. Frontal view of hooks with sclerotic plaques. C. Immature proglottid. D. Mature proglottid showing testicles. E. Mature proglottid. F. Terminal proglottid. Scale bars: A. 115  $\mu$ m; B. 160  $\mu$ m; C. 300  $\mu$ m; D. 535  $\mu$ m; E. 1170  $\mu$ m; F. 1725  $\mu$ m.



**Figure 4.** Scanning electron micrographs of *Acanthobothrium marquesi* n. sp. from *Aetobatus cf. narinari*. A. Scolex. B. Front view of pad, hooks and bothridia. C. Superficial apical view. D. Front view of pad and hooks. Scale bars: A, B, C, D 100  $\mu$ m.

7.9 cm to 51.0 cm ( $26.1 \text{ cm} \pm 17.1 \text{ cm}$ ,  $n = 10$ ) long; composed of 903-2,379 ( $1,549 \pm 491$ ,  $n = 11$ ) proglottids. Scolex proper 500-1,000 ( $707 \pm 151$ ;  $n = 20$ ) long by 670-1,130 ( $872 \pm 122$ ;  $n = 19$ ) wide, composed of 4 sessile trilobulate bothridia; each bothridium with apical sucker and pad, armed with pair of bifid hooks (Figures 2-4). Bothridia with thick muscular walls; 490-915 ( $626 \pm 136$ ;  $n = 19$ ) long by 233-338 ( $274 \pm 27$ ,  $n = 20$ ) wide. Locular wall dividing anterior and middle loculi relatively thick and muscular; wall between middle and posterior loculi relatively thin. Anterior loculus 263-480 ( $373 \pm 72$ ,  $n = 20$ ) long, middle loculus 75-150 ( $113 \pm 22$ ,  $n = 20$ ) long, posterior loculus 88-140 ( $110 \pm 15$ ,  $n = 19$ ) long (Figures 2-4). Ratio of lengths of loculi 1:0.3:0.3.

Apical suckers not strongly defined, 118-170 ( $143 \pm 15$ ,  $n = 18$ ) in diameter; pads muscular, 258-365 ( $309 \pm 32$ ,  $n = 19$ ) wide. Posterior edge of apical muscular pads extending posteriorly over handles of hooks, divided in middle to form two straight extensions; posterior edge not well defined. Hooks bipronged, handle stout, extended almost directly opposite prongs (Figures 2-4). Hook formula for external hooks ( $n = 18$ ):

$$\frac{99 \pm 4 \ (93 - 108) \ 100 \pm 6 \ (90 - 110) \ 100 \pm 5 \ (93 - 108)}{196 \pm 9 \ (183 - 213)} \quad (1)$$

Hook formula for internal hooks ( $n = 16$ ):

$$\frac{99 \pm 5 \ (90 - 110) \ 99 \pm 7 \ (90 - 113) \ 98 \pm 6 \ (85 \text{v} 110)}{193 \pm 12 \ (175 - 213)} \quad (2)$$

Cephalic peduncle unspined, 5,750-13,650 ( $9,255 \pm 1,945$ ,  $n = 21$ ) long by 510-1,290 ( $742 \pm 235$ ,  $n = 21$ ). Immature proglottids wider than long, 200-950 ( $468 \pm 193$ ,  $n = 23$ ) long by 220-2,720 ( $1,655 \pm 617$ ,  $n = 24$ ) wide (Figure 3); mature proglottids, 300-1,450 ( $763 \pm 296$ ,  $n = 23$ ) long by 830-2,000 ( $1,405 \pm 327$ ,  $n = 23$ ) wide (Figures 2 and 3); terminal proglottids 980-2,250 ( $1,388 \pm 381$ ,  $n = 13$ ) long by 780-1,900 ( $1,152 \pm 324$ ,  $n = 12$ ) wide (Figures 2 and 3). Testes 109-171 ( $138 \pm 17$ ,  $n = 18$ ) in number, 25-45 ( $33 \pm 7$ ,  $n = 17$ ) preporal, 14-39 ( $27 \pm 9$ ,  $n = 18$ ) postporal, 62-95 ( $81 \pm 8$ ,  $n = 18$ ) antiporal. Testes 43-150 ( $75 \pm 26$ ,  $n = 21$ ) long by 40-130 ( $71 \pm 24$ ,  $n = 21$ ) wide (Figures 2 and 3). Cirrus sac slightly posterior to mid-proglottid, extending medially to mid-proglottid, curved posteriorly in terminal proglottids, 325-750 ( $425 \pm 90$ ,  $n = 22$ ) long, 100-450 ( $197 \pm 76$ ,  $n = 22$ ) wide (Figures 2 and 3). Cirrus 556  $\mu\text{m}$  long when extended, slightly swollen at base. Cirrus sac containing spined eversible cirrus. Genital atrium shallow. Genital pore located 41%-59% ( $51\% \pm 6\%$ ,  $n = 21$ ) of total length of proglottid from anterior end, irregularly alternating. Vagina anterior to cirrus sac, vaginal wall glandular. Vagina canal can form loops, descends on the aporal, relatively to the midline of proglottid. Ovary near posterior end of proglottid, with foliose lobes. Shape in frontal view changing as proglottids mature: nearly U-shaped in immature proglottids, H-shaped in mature proglottids, and inverted A- or V-shaped in terminal proglottids. Ovary 460-1400 ( $974 \pm 305$ ,  $n = 22$ ) wide at isthmus; arms unequal in length; in proglottids mature, and terminal the isthmus often located in middle position regarding the uterus, gravid proglottid not observed (Figures 2 and 3). Aporal arm 250-810 ( $435 \pm 158$ ,  $n = 15$ ) long, extending to

anterior margin of cirrus sac; poral arm 250-920 ( $473 \pm 180$ ,  $n = 15$ ) long. Mehlis' gland immediately posterior to ovarian isthmus; external seminal receptacle at level of or slightly anterior to isthmus (Figures 2 and 3). Vitelline follicles elongate oval in shape, 120-640 ( $305 \pm 146$ ,  $n = 21$ ) wide, 160-640 ( $323 \pm 136$ ,  $n = 21$ ) long, extending as narrow bands on each side of proglottid. Uterus saccate, narrowly elongate in terminal attached proglottids. Eggs not observed.

## Taxonomic Summary

Type host: *Aetobatus* cf. *narinari* (Euphrasen, 1790); the host belongs to a currently unresolved species complex (RICHARDS et al., 2009; WHITE et al., 2010).

Site of infection: spiral valve.

Type locality: Laguna de Términos, Ciudad del Carmen, Campeche, México ( $18^{\circ}35'19''\text{N}$ ;  $91^{\circ}33'30''\text{W}$ ).

Other locality: Champotón, Campeche, México ( $19^{\circ}21'\text{N}$ ;  $90^{\circ}54'\text{W}$ ).

Holotype: CNHE-10554.

Paratypes: CNHE-10555 to 10556; HWML-139377 to 139384; CHE-P00061 to P00063.

Zoobank registration: B4DAA0D5-284B-4B66-9477-CDDEAA98B7BE (ZOOBANK, 2018)

Etymology: the species is named in honor of Dr. Fernando Marques (Departamento de Zoologia, Universidade de São Paulo, São Paulo, Brazil) for his friendship and for his contributions to our knowledge of the helminths of stingrays.

**Remarks:** There are 11 species reported from the Pacific Coast of México (Figure 1; Table 1); *A. bajaensis* Appy and Dailey, 1973; *A. bullardi* Ghoshroy and Caira, 2001; *A. cleofanus* Monks, Brooks, and Pérez-Ponce de León, 1996; *A. dasi* Ghoshroy and Caira, 2001; *A. dollyae* Caira and Burge, 2001; *A. maryanskii* Caira and Burge, 2001; *A. puertecitense* Caira and Zahner, 2001; *A. rajivi* Ghoshroy and Caira, 2001; *A. royi* Caira and Burge, 2001; *A. santarosaliense* Caira and Zahner, 2001; *A. soberoni* Ghoshroy and Caira, 2001 (CAIRA & BURGE, 2001; CAIRA & ZAHNER, 2001; GHOSHROY & CAIRA, 2001; MONKS et al., 1996). None of these have been reported from *Aetobatus* cf. *narinari* in Mexican waters.

*Acanthobothrium cartagenensis* Brooks and Mayes, 1980 is the only species that has been reported from the Eastern Coast of México (includes the Gulf of Mexico and Caribbean Sea). It can be distinguished from the new species because it is a Category 9 species (LFFS: L, >15 mm; F,  $\leq 50$  segments; F,  $\leq 80$  testes; S, left and right lobes symmetrical) (categories *sensu* GHOSHROY & CAIRA, 2001).

The new species is the third member of the genus reported from *Aetobatus* cf. *narinari* in the greater Atlantic Ocean. The species that have been reported from *Aetobatus* cf. *narinari* from the eastern Pacific Ocean are *A. monksi* Marques, Brooks, and Barriga, 1997, and *A. nicoyaense* Brooks and McCorquodale, 1995 (both are Category 1 species; i.e., much smaller than the new species). None of these species have been reported from Mexican waters (MARGUEE et al., 1997; BROOKS & MCCORQUODALE, 1995). The species of *Acanthobothrium* that have been reported



from *Aetobatus* cf. *narinari* from the western Atlantic Ocean are, *A. colombianum* Brooks and Mayes, 1980 and *A. tortum* (Linton, 1916) Baer and Euzet, 1962.

*Acanthobothrium marquesi* is a Category 3 species (LMMA) (*sensu* GHOSHROY & CAIRA, 2001): total length > 15 mm (specimens of the new species are 26,100 mm long); number of proglottids > 50 (average of 1,549 proglottids); number of testes per proglottid > 80 (138 testes per proglottid); and ovarian lobes asymmetrical (aporal lobe reaches anteriorly from the posterior end of the proglottid to about the anterior margin of the cirrus sac and the poral lobe reaches anteriorly to the posterior margin of the cirrus sac).

*Acanthobothrium colombianum* is a Category 9 species (L = >15 mm; F = ≤50 proglottids; F = ≤80 testes per proglottid; S = ovarian lobes symmetrical), differing from the new species in the number of proglottids, number of testes, and the symmetry of the ovarian lobes. *Acanthobothrium monksi* and *A. nicoyaense* are Category 1 species (S = ≤15 mm; F = ≤50 proglottids; F = ≤80 testes per proglottid; S = ovarian lobes symmetrical).

*Acanthobothrium tortum*, as described by Campbell (1970) is a Category 3 species and is the most similar species in the Western Atlantic Ocean (Eastern coast of North America). *Acanthobothrium marquesi* n. sp. can be distinguished from *A. tortum* by being longer (26.1 cm vs. 10.6 cm), having a greater number of proglottids (1,549 proglottids vs. 656), having a larger scolex (872 long by 872 wide vs. 699 long by 665 wide), larger bothridia (626 long by 274 wide vs. 563 long by 238 wide). In specimens of the new species the sizes and relative proportions of the bothridia are different from those of *A. tortum* (373:113:110 = 1:0.3:0.3 vs. 259:87:117 = 1:0.3:0.5, respectively). Finally, the cirrus sac is larger in the new species (425 long by 197 wide vs. 350 long by 122 wide) and the number of testes is less (138 total = 33 preporal, 27 postporal, and 81 aporal vs. 163 total = 44 preporal, 32 postporal, and 87 aporal).

Other amphi-American Category 3 species are *A. holorbini* Alexander, 1953 and *A. maculatum* Riser, 1955, both from *Myliobatis californica* Gill; neither has been reported from México. These two species are smaller than the new species in all structures and the number of testes is less in the former two species than in the new species. Finally, these two species and their hosts are known only from the Pacific coast.

## Discussion

Only seven species of *Acanthobothrium* have been reported previously from México, six species from the Pacific coast (*A. bajaensis* Appy and Dailey, 1973; *A. bullardi* Ghoshroy and Caira, 2001; *A. cleofanus*; *A. dasi* Ghoshroy and Caira, 2001; *A. rajivi* Ghoshroy and Caira, 2001; and *A. soberoni* Ghoshroy and Caira, 2001) and one from the Caribbean coast (*A. cartagenensis* Brooks and Mayes, 1980) (APPY & DAILEY, 1973; MONKS et al., 1996, 2015; GHOSHROY & CAIRA, 2001); *A. marquesi* is the eleventh species reported from México and the second species from the eastern coast of México.

The nominal species *Aetobatus narinari* (*sensu* Euphrasen) has classically been thought to be a globally-distributed species inhabiting

tropical seas, but recent studies have suggested that it represents a species complex (RICHARDS et al., 2009; WHITE et al., 2010; WHITE et al., 2013), an hypothesis supported by parasitological studies (MARIE & JUSTINE, 2005; MARIE & JUSTINE, 2006). No molecular data exist for the particular populations of *Aetobatus* cf. *narinari* that are hosts to *A. tortum* and *A. marquesi* n. sp., but they are on the opposite north-south coasts of the Gulf of México; the presence of different species of *Acanthobothrium* in each suggests that these two population of *Aetobatus* cf. *narinari* do not mix. The study of divergence of cryptic populations of species that serve as hosts and that of their parasites is in initial processes, but studies of water currents in the Gulf of México/Caribbean Sea region suggest that more isolated species of helminths are waiting to be discovered (SHULMAN & BERMINGHAM, 1995; CARTON & CHAO, 1999; SANDOVAL-CASTILLO & ROCHA-OLIVARES, 2011).

Members of *Aetobatus* Blainville, 1816 traditionally have been considered to be part of Myliobatidae (FROESE & PAULY, 2017). However, White & Naylor (2016) recently moved the genus to Aetobatidae. Although this arrangement has begun to be followed (LAST et al, 2016), we chose to follow the most widely accepted classification (FROESE & PAULY, 2017) pending further evaluations of the status of the family.

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