



# A new species of *Heterocotyle* (Monogenea: Monocotylidae), a gill parasite of the diamond stingray *Hypanus dipterurus* (Myliobatiformes: Dasyatidae) from the Peruvian coastal zone

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## Abstract

**Introduction** A new monogenean, *Heterocotyle margaritae* n. sp. (Monocotylidae: Heterocotylinae), is described based on specimens collected from the gills of the diamond stingray, *Hypanus dipterurus* (Jordan & Gilbert, 1880) (Myliobatiformes: Dasyatidae) captured off the South Pacific coast, Lima Region, Peru.

**Materials and methods** Monogeneans were fixed in hot 4% formalin, observed and measured as permanent mounts stained with Gomori's trichrome, and mounted in Canada balsam. Drawings were made with the aid of a drawing tube.

**Results** The new species is distinguished from all congeners mainly by having a funnel-shaped male copulatory organ, spatulate distally with lateral folds and by its club-shaped accessory piece. The most similar congener, *H. americana* Hargis, 1955, resemble *H. margaritae* n. sp. regarding the general structure of the male copulatory organ (funnel-shaped with an accessory piece). However, besides other features, the new species differ from *H. americana* by having an accessory piece without a dorsal button (vs an accessory piece with a dorsal button in *H. americana*).

**Conclusions** This is the first report of a member of *Heterocotyle* Scott, 1904 in the Southeastern Pacific Ocean. The present finding brings to 20 the number of known species of *Heterocotyle*, and represents the fifth described monogenean species from *Hypanus dipterurus*.

**Keywords** Monogenea · Elasmobranch · Taxonomy · Marine fish · Peru

## Introduction

Members of *Heterocotyle* Scott, 1904 (Monogenea: Monocotylidae) are branchial parasites that infect mainly dasyatids [1]. 21 nominal species have been described, but only 19 of them are considered valid, namely *H. americana* Hargis, 1955; *H. armata* Timofeeva, 1983; *H. capapei* Neifar, Euzet & Ben Hassine, 2000; *H. capricornensis* Chisholm & Whittington, 1996; *H. chinensis* Timofeeva, 1983; *H. confusa* Timofeeva, 1983; *H. dasyatis* (Yamaguti, 1965); *H. forcifera* Neifar, Euzet & Ben Hassine, 1999; *H. granulatae* Young, 1967; *H. minima* (MacCallum, 1916) Price, 1938; *H. mokhtarai* Neifar, Euzet & Ben Hassine, 1999; *H. pastinacae* Scott, 1904; *H. pseudominima* Hargis, 1955; *H. scotti* Neifar, Euzet & Ben Hassine, 1998; *H. similis* Neifar, Euzet & Ben Hassine, 1998; *H. striata* Neifar, Euzet & Ben Hassine, 1999; *H. sulamericana* Santos, Santos, Cunha & Chisholm, 2012; *H. taeniuropi* Cao, Ding, Zhang & Liu, 2010; and *H. tokoloshei* Vaughan & Chisholm, 2010 [1–7]. Three of these species have

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been described in the Americas: *H. americana* from the Southern stingray *Hypanus americanus* (Hildebrand & Schroeder, 1928) in the United States; *H. pseudominima* from *Hypanus* sp., probably either *Hy. americanus* or the bluntnose stingray *Hy. say* (Lesueur, 1817), in the United States and *H. sulamericana* from the longnose stingray *Hy. guttatus* (Bloch & Schneider, 1801) in Brazil [2–5].

*Hypanus dipterurus* (Jordan & Gilbert, 1880) (Myliobatiformes: Dasyatidae) called diamond stingray, is a demersal fish endemic to Eastern Pacific Ocean, which inhabits sandy areas, often around rocks and kelp beds [8, 9]. This species is distributed from southern California, USA to Peru, including the Galapagos Islands [10]. To date, four monogenean species have been described

Key to the species of *Heterocotyle* (modified from Nitta and Nagasawa [7])

1. Haptor with single sinuous ridge surmounting the septa ..... 2  
Haptor with 1/2/3 sinuous ridge arrangement ..... 7
2. Accessory piece of male copulatory organ (MCO) absent ..... 3  
Accessory piece of MCO present ..... 4
3. MCO 77–106 µm long; testis with three posteriorly-directed finger-like lobes ..... *H. capricornensis*  
MCO 23–27 µm long; testis tubular forming complete loop ..... *H. sulamericana*
4. Accessory piece with spines ..... 5  
Accessory piece without spines ..... 6
5. Accessory piece with two branches fused to base of the MCO ..... *H. forcifera*  
Accessory piece unbranched with distinct spines distally ..... *H. armata*
6. Accessory piece small, associated with the distal end of the MCO ..... *H. confusa*  
Accessory piece larger, not associated with the distal end of the MCO ..... *H. granulatae*
7. Accessory piece of MCO present ..... 8  
Accessory piece of MCO absent ..... 13
8. Accessory piece of MCO composed of 10 sclerotized spines ..... *H. dasyatis*  
Accessory piece of MCO composed of a single piece ..... 9
9. Accessory piece associated with proximal part of the MCO ..... 10  
Accessory piece associated with distal part of the MCO ..... *H. minima*
10. MCO more than 180 µm long, slender or J-shaped ..... 11  
MCO less than 92 µm long, with funnel-shaped ..... 12
11. MCO 193–330 µm long, slender and narrow tube-shaped; accessory piece small with jagged edge ..... *H. taeniuropi*  
MCO 189–260 µm long, J-shaped, distal region with approximately 20 small spines; comb-like accessory piece ..... *H. tokoloshei*
12. MCO with spatulate distal end and lateral folds; accessory piece club-shaped without dorsal bottom; vagina sclerotized ..... *H. margaritae* n. sp.  
MCO with straight and tubular distal end, without lateral folds; accessory piece claw-like with a dorsal bottom; vagina muscular ..... *H. americana*
13. Distal portion of the MCO with long points ..... 14  
Distal portion of the MCO without points ..... 15
14. Distal portion of the MCO with 3 long points; vagina with rows of sclerotized bars armed with spines ..... *H. pastinacae*  
Distal portion of the MCO with one long points; vagina without sclerotized structures ..... *H. capapei*
15. Distal end of MCO hook-shaped ..... *H. similis*  
Distal end of MCO straight ..... 16
16. MCO with two rows of filaments ..... *H. striata*  
MCO without rows of filaments ..... 17
17. Ejaculatory bulb larger than the MCO ..... 18  
Ejaculatory bulb of approximately same length as MCO ..... 19
18. Ejaculatory bulb 3 times as long as MCO ..... *H. mokhtarai*  
Ejaculatory bulb 1–2 times as long as MCO ..... *H. chinensis*
19. MCO 36–54 µm long, curved ..... *H. pseudominima*  
MCO 115–155 µm long, sinuous ..... *H. scotti*

parasitizing *H. dipterurus* in the Pacific Ocean: *Listrocephalos kearni* Bullard, Payne & Braswell, 2004 (Cap-salidae) in Mexico, *Hypanocotyle bullardi* Chero, Cruces, Sáez, Camargo, Santos & Luque, 2018 (Hexabothriidae), *Monocotyle luquei* Chero, Cruces, Iannacone, Sanchez, Minaya, Sáez & Alvariano, 2016 and *Peruanocotyle chisholmae* Chero, Cruces, Sáez & Luque, 2018 (Monocotylidae) in Peru [5, 11–15].

During a parasitological survey of Peruvian marine fishes carried out from July 2018 through November 2018, several specimens of monogeneans were found on the gill filaments of *Hy. dipterurus* caught near Chorrillos, central Peru. Detailed morphological examination revealed that the parasites represent a new species of *Heterocotyle*, which is described and illustrated herein. In addition, Nitta and Nagasawa's [7] key to the species of *Heterocotyle* is revised to include the new species.

## Materials and methods

Stingrays were collected during July 2018 through November 2018 from the coastal zone of Chorrillos, Lima, Peru (12° 09' S, 77° 01' W), using gillnets, euthanized immediately after capture, and dissected. The gills were excised and placed in Petri dishes with seawater and examined for monogeneans with the aid of a stereomicroscope. Monogeneans were collected alive, washed in seawater, fixed in hot 4% formalin under slight coverslip pressure. They were then stained with Semichon's carmine or Gomori's trichrome, clarified in Eugenol, and mounted in Canada balsam to study internal organs. Other specimens were mounted directly in Gray and Wess medium for study of sclerotized structures [16]. Specimens were examined using a compound Olympus™ BX51 light photomicroscope equipped with Nomarski Differential Interference Contrast (DIC) optics and drawings were made with the aid of a drawing tube. Unless otherwise stated, measurements are in micrometers, representing straight-line distances between extreme points of the structures measured and are expressed as the range followed by the mean and number (*n*) of specimens measured in parentheses. Body length represents the length of the body proper including the haptor. Scientific names, including taxonomic authorities and dates, for fishes follow Eschmeyer et al. [17]. Nomenclature and classification of Myliobatiformes, Dasyatidae and *Hypanus* spp. follow Last et al. [18]. Mean intensity of infection was calculated according to Bush et al. [9]. Specimens deposited in the Coleção Helmintológica do Instituto Oswaldo Cruz (CHIOC), Brazil of *Heterocotyle sulamericana* Santos, Santos, Cunha & Chisholm, 2012 (holotype, CHIOC 37551; paratype, CHIOC 37552), were examined. Type material was deposited in the Helminthological Collection of the Museum of Natural History at the San Marcos University (MUSM), Lima, Peru and in the CHIOC.

## Result

### Order Monocotylidea Lebedev, 1988

#### Monocotylidae Taschenberg, 1879

#### Heterocotylinae Chisholm, Wheeler & Beverley-Burton, 1995

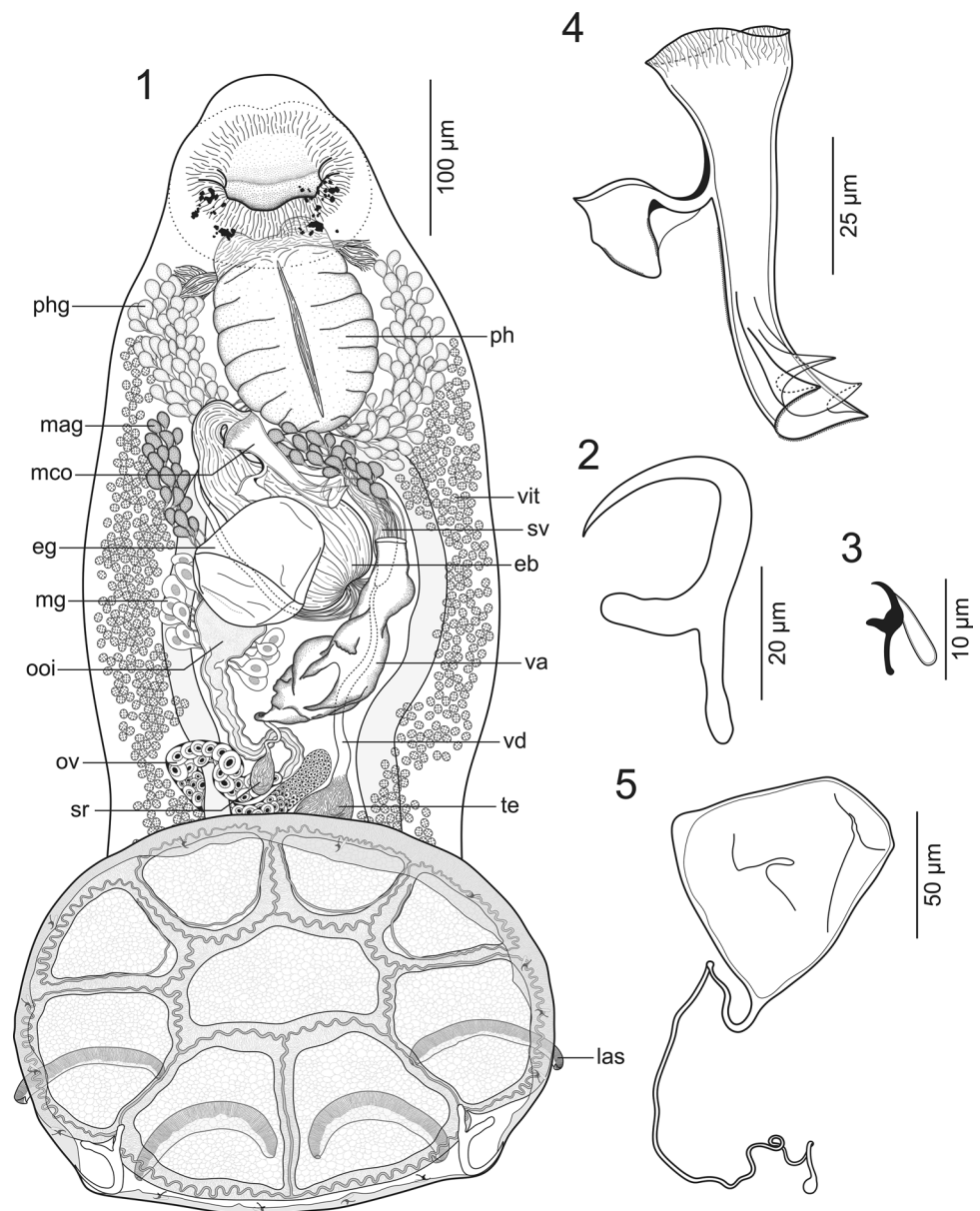
##### *Heterocotyle margaritae* n. sp.

(Fig. 1–5).

Description (based on 14 adult specimens): body 0.57–1.24 (0.83; *n* = 14) mm long, 161–364 (258; *n* = 14) wide usually at level of ejaculatory bulb, slightly constricted at level of pharynx. Tegument smooth. Haptor broadly ellipsoid 184–377 (261; *n* = 11) long, 280–542 (358; *n* = 11) wide, with one central and eight peripheral loculi; posterior loculi slightly larger than anterior ones (Fig. 1). Sinuous ridges of haptor with 1/2/3 configuration: single on inner and outer ring and three posterior septa, double on two lateral radial septa and triple on three anterior radial septa (see [1]); marginal membrane delicate, uniform, 19–32 (26; *n* = 5) wide (Fig. 1). Dorsal surface of haptor with four accessory structures, luniform, sclerotized, striated at anterior edge with lateral extremities folded around adjacent region, situated dorsal to posterior loculi (Fig. 1). Anchor 40–43 (41; *n* = 12) long, associated with posterolateral septa, with deep and superficial roots distinct; elongate and narrow deep root; thick superficial root; slightly curved shaft and recurved point, point extending beyond of tip of superficial root (Fig. 2). Fourteen similar hooks, 9–10 (9; *n* = 5) long, distributed in the marginal membrane as depicted (Fig. 1), each with short and curved point, protruding thumb, dilated shaft and uniform and narrow shank; filamentous hook (FH) loop extending to near level of proximal end of shank (Fig. 3). Eye-spots in form of dispersed pigment granules distributed in two areas (Fig. 1), one pair of dispersed granules posterolateral to oral opening, and loosely concentrated granules at level of anterior end of pharynx. Mouth ventral, subterminal, surrounded by muscular pseudosucker. Anterior glands not observed. Pharynx muscular, barrel-shaped, 73–131 (105; *n* = 9) long, 57–124 (88; *n* = 9) wide, comprising short anterior part surrounded by weak sphincter and long posterior part; prominent pharyngeal glands either side of pharynx; esophagus short to indistinct. Intestinal bifurcation anterior to common genital aperture; intestinal caeca unbranched, extending from anterior to posterior end of body proper, not confluent posteriorly (Fig. 1). Common genital aperture unarmed. Testis almost reniform, limited to intercecal field. Vas deferens originating from left anterior margin of testis, running anteriorly dorsal to vagina; widening to form seminal vesicle. Seminal vesicle elongate, sinistral to ejaculatory bulb, continues as a narrow duct and loops dorsally to enter the base of ejaculatory bulb with numerous accessory glands. Ejaculatory bulb well-developed, highly muscular,

**Fig. 1–5** *Heterocotyle margari-tae* n. sp. (Monocotylidae) from the gills of the diamond stingray *Hypanus dipterurus*, holotype.

**1.** Whole worm, ventral view. *eb* ejaculatory bulb, *eg* egg, *las* luniform accessory structures, *mag* male accessory glands, *mco* male copulatory organ, *mg* Mehlis' glands, *ooi* ootype, *ov* ovary, *ph* pharynx, *phg* pharyngeal glands, *sr* seminal receptacle, *sv* seminal vesicle, *te* testis, *va* vagina, *vd* vas deferens, *vit* vitelline follicles. **2.** Anchor. **3.** Hook. **4.** Male copulatory organ. **5.** Egg



panduriform, 102–219 (174;  $n=6$ ) long, 41–129 (86;  $n=6$ ) wide, anteromedial to oötype; anterior region narrows to meet proximal region of male copulatory organ (MCO). Male copulatory organ 73–80 (76;  $n=11$ ) long, sclerotized, funnel-shaped, spatulate distally with lateral folds (Fig. 4). Accessory piece sclerotized, club-shaped, 41–45 (43;  $n=4$ ) long, originates from proximal region of MCO (Fig. 4). Ovary elongate, dextralateral to testis, encircles right intestinal caecum dorsoventrally and narrows to form oviduct, which receives seminal receptacle and vitelline ducts. Oötype 163–185 (173;  $n=4$ ) long, 61–68 (64;  $n=2$ ) wide, cup-shaped, proximal end slightly sinuous (Fig. 1). Vagina sclerotized, sac-like, lying obliquely on left side of body anterior to ovary; proximal portion of vagina connected to small narrowly ovate seminal receptacle by a thin duct

(Fig. 1); seminal receptacle 27–44 (36;  $n=3$ ) long, 16–22 (18;  $n=3$ ) wide. Vitelline follicles dense, extending from pharyngeal level to posterior end of trunk, absent in regions of reproductive organs, (Fig. 1). Vaginal pore unarmed, opening ventrally on left side, 230–447 (348;  $n=5$ ) mm from anterior end. Egg tetrahedral 74–89 (79;  $n=3$ ) long, 73–87 (79;  $n=3$ ) wide, single filament 163–253 (200;  $n=3$ ) long, with a small button 6–7 (6;  $n=3$ ) long at distal end.

### Taxonomic summary

**Type host:** *Hypanus dipterurus* (Jordan & Gilbert, 1880) (Myliobatiformes: Dasyatidae), diamond stingray.

**Site in host:** Gill filaments.



**Type locality:** Chorrillos, Lima, Peru, South Pacific Ocean (12° 09' S, 77° 01' W).

**Infection:** Two of three hosts infected with a total of 14 worms.

**Mean intensity:**  $7 \pm 2.82$  (range 5–9) monogeneans per infected fish.

**Type material:** Holotype, MUSM 4436; nine paratypes, MUSM 4437a–i; four paratypes, CHIOC 39264a–d.

**Etymology:** The new species is named in honor to Paulina Margarita de la Cruz Lavado, mother of the first author.

**Remarks:** The newly collected specimens from *Hy. dipterurus* are assigned to *Heterocotyle* by having a haptor with one central and eight peripheral loculi, sinuous ridges of types 1/2/3 and four accessory structures on the dorsal surface of the haptor. *Heterocotyle margaritae* n. sp. differs from *H. armata*, *H. capricornensis*, *H. confusa*, *H. forcifera*, *H. granulatae*, and *H. sulamericana* by having a haptor with 1/2/3 sinuous ridges arrangement, rather than having a single sinuous ridge surmounting the septa as in aforementioned species. *Heterocotyle capapei*, *H. chinensis*, *H. mokhtarae*, *H. pastinacae*, *H. pseudominima*, *H. scotti*, *H. similis*, and *H. striata* all differ from *H. margaritae* n. sp. by having a male copulatory organ lacking accessory piece (present in the new species). *Heterocotyle margaritae* n. sp. can be distinguished from *H. dasyatis*, *H. minima*, *H. taeniuropi* and *H. tokoloshei* by the morphology of the accessory piece (club-shaped in the new species vs composed of about ten sclerotized spines in *H. dasyatis*, bell-shaped in *H. minima*, small with jagged edge in *H. taeniuropi* and comb-like in *H. tokoloshei*) and by the size of the male copulatory organ (75–80 in the new species vs 32–46 in *H. dasyatis*, 110 in *H. minima*, 193–330 in *H. taeniuropi* and 189–260 *H. tokoloshei*). In the presence of a funnel-shaped MCO with an accessory piece, *H. margaritae* n. sp. resembles *H. americana*. However, *H. margaritae* n. sp. can be easily distinguished from *H. americana* by its club-shaped accessory piece (vs claw-like with a dorsal button in *H. americana*) and by having a MCO with spatulate distal end and lateral folds (vs straight and tubular distal end without lateral folds in *H. americana*). In addition, the new species differs from *H. americana* by possessing a sclerotized vagina (vs muscular in *H. americana*) and a panduriform ejaculatory bulb (vs ovoid in *H. americana*). This is the first report of a member of *Heterocotyle* in the Southeastern Pacific Ocean.

## Discussion

*Heterocotyle margaritae* n. sp. from *Hy. dipterurus* off Peru is added to the three previously described species of *Heterocotyle* from species of *Hypanus*, i.e. *H. americana*, *H.*

*pseudominima* and *H. sulamericana* (Table 1). The new species is distinguished from these species by the following combination of features: a haptor with 1/2/3 sinuous ridge arrangement, a MCO with an accessory piece, and a vagina sclerotized. According to Chisholm and Whittington [1], the morphology of the MCO is the best character to differentiate between species of *Heterocotyle*. However, this character should be studied only in mature specimens since variation in the morphology of the MCO with the age has been described in some monocotylid species, i.e., *H. granulatae* and *Dendromonocotyle ardea* Chisholm & Whittington, 1995 [1, 20]. The MCO of *H. margaritae* n. sp. (based only in mature specimens) differs from all congeners by having funnel-shaped, spatulate distally with lateral folds and by its club-shaped accessory piece.

Nineteen monogenean species from the genera *Listrocephalos* Bullard, Payne & Braswell, 2004 (3 sp.) (Capsalidae), *Dasyonchocotyle* Hargis, 1955 (2 spp.), *Hypanocotyle* Chero, Cruces, Sáez, Camargo, Santos & Luque, 2018 (1 sp.) (Hexabothriidae), *Loimopapillosum* Hargis, 1955 (1 sp.) (Loimoidae), *Dendromonocotyle* Hargis, 1955 (1 sp.), *Heterocotyle* (3 spp.), *Merizocotyle* Cerfontaine, 1894 (3 spp.), *Monocotyle* Taschenberg, 1878 (4 spp.) and *Peruanocotyle* Chero, Cruces, Sáez, Camargo, Santos & Luque, 2018 (1 sp.) (Monocotylidae) have been described or reported infecting the gills or skin of six of the eight known *Hypanus* species. From this, four species of monogeneans from five genera are known parasitizing *Hy. dipterurus* (see Table 1). The capsalid *Listrocephalos kearni* was the first species of monogenean described from this host species, which was captured in the Gulf of California, Mexico [11]. Later, three other monogenean species (a hexabothriid and two monocotylids), were described from Peru [12, 14, 15]. The new species described here increases the number of species that infect *Hy. dipterurus* to five, which is a relatively poorly studied fish host. The occurrence of different monogenean species infesting *Hy. dipterurus* is not unusual because this has been previously reported in the other *Hypanus* species, i.e. *Hy. americanus* and *Hy. say* with five species, *Hy. sabinus* with four species and *Hy. guttatus* and *Hy. longus* with two species (Table 1). The presence of multiple monogenean species infecting a single species of *Hypanus* provides evidence that the diversity of monogeneans from these hosts requires further study and probably new species can still be discovered.

The present finding brings to 20 the number of known species of *Heterocotyle* and a key to the species of *Heterocotyle* is provided.

**Table 1** Species of Monogenea infecting fishes of the genus *Hypanus* Rafinesque, 1818 (Myliobatiformes: Dasyatidae) worldwide

Species	Host	Locality	Site	References
<b>CAPSALIDAE</b> Baird, 1853				
<i>Listrocephalus corona</i> (Hargis, 1955) Bullard, Payne & Braswell, 2004	<i>Hypanus americanus</i> <i>Hypanus sabinus</i> <i>Hypanus say</i>	Alligator Harbor, Florida, Gulf of Mexico, EEUU Mississippi, Gulf of Mexico, EEUU	Skin Gills	Hargis [22], Bullard et al. [11]
<i>Listrocephalus kearnii</i> Bullard, Payne & Braswell, 2004	<i>Hypanus dipterurus</i>	Bahia de los Angeles, Gulf of California, Mexico Off Santa Rosalia, Gulf of California, Mexico	Skin	Bullard et al. [11]
<i>Listrocephalus whittingtoni</i> Bullard, Payne & Braswell, 2004	<i>Hypanus longus</i>	Bahia de los Angeles, Gulf of California, Mexico Off La Paz, Gulf of California, Mexico	Skin	Bullard et al. [11]
<b>LOIMOIDAE</b> Price, 1936				
<i>Loimopapillosum dasyatis</i> Hargis, 1955	<i>Hypanus americanus</i> <i>Hypanus say</i>	Alligator Harbor, Franklin Co., Gulf of Mexico, EEUU	Gills	Hargis [22]
<b>MONOCOTYLIDAE</b> Taschenberg, 1879				
<i>Dendromonocotyle octodiscus</i> Hargis, 1955	<i>Hypanus say</i>	Alligator Harbor, Franklin Co., Gulf of Mexico, EEUU	Skin	Hargis [22]
<i>Heterocotyle americana</i> Hargis, 1955	<i>Hypanus americanus</i>	Alligator Harbor, Franklin Co., Gulf of Mexico, EEUU	Gills	Hargis [2]
<i>Heterocotyle margaritae</i> n. sp.	<i>Hypanus dipterurus</i>	Chorrillos, Lima Region, Peru	Gills	Present study
<i>Heterocotyle pseudominima</i> Hargis, 1955	<i>Hypanus</i> sp. <sup>a</sup>	Alligator Harbor, Franklin Co., Gulf of Mexico, EEUU	Gills	Hargis [2]
<i>Heterocotyle sulamericana</i> Santos, Santos, Cunha & Chisholm, 2012	<i>Hypanus guttatus</i>	Off Angra dos Reis, Rio de Janeiro, Brazil	Gills	Santos et al. [3]
<i>Merizocotyle longicirrus</i> (Hargis, 1955) Chisholm, Wheeler & Beverley-Burton, 1995	<i>Hypanus say</i>	Alligator Harbor, Franklin Co., Gulf of Mexico, EEUU	Skin	Hargis [22]
<i>Merizocotyle retorta</i> (Hargis, 1955) Chisholm, Wheeler & Beverley-Burton, 1995	<i>Hypanus americanus</i>	Alligator Harbor, Franklin Co., Gulf of Mexico, EEUU	Gills	Hargis [22]
<i>Merizocotyle roumillati</i> (de Buron & Euzet, 2005)	<i>Hypanus sabinus</i>	Charleston, South Carolina, EEUU	Nasal fossae	de Buron and Euzet [23]
<i>Monocotyle diademalis</i> Hargis, 1955	<i>Hypanus sabinus</i> <i>Hypanus</i> sp. <sup>a</sup>	Alligator Harbor, Franklin Co., Gulf of Mexico, EEUU	Gills	Hargis [2]
<i>Monocotyle guttatae</i> Santos, Santos & Gibson, 2006	<i>Hypanus guttatus</i>	Off Angra dos Reis, Rio de Janeiro, Brazil	Gills	Santos et al. [24]
<i>Monocotyle luquei</i> Chero, Cruces, Iannacone, Sanchez, Minaya, Sáez & Alvarino, 2016	<i>Hypanus dipterurus</i>	Callao, Lima Region, Peru	Gills	Chero et al. [12]
<i>Monocotyle pricei</i> Pearse, 1949	<i>Hypanus americanus</i> <i>Hypanus say</i>	Alligator Harbor, Florida, Gulf of Mexico, EEUU	Gills	Hargis [2]
<i>Peruanocotyle chisholmae</i> Chero, Cruces, Sáez & Luque, 2018	<i>Hypanus dipterurus</i>	Callao, Lima Region, Peru	Gills	Chero et al. [15]
<b>HEXABOTHRIDAE</b> Price, 1942				
<i>Dasyonchocotyle dasyatis</i> (Yamaguti, 1968) Boeger & Kritsky, 1989	<i>Hypanus longus</i> <i>Hypanus</i> sp. <sup>b</sup>	Hawaii La Puntilla, Mazatlán, Sinaloa, Mexico Bahia Acapulco, Guerrero, Mexico	Gills	Yamaguti [25], Escorcia-Ignazio et al. [26]
<i>Dasyonchocotyle spiniphallus</i> Hargis, 1955	<i>Hypanus sabinus</i>	Alligator Harbor, Franklin Co., Gulf of Mexico, EEUU	Gills	Hargis [27]
<i>Hypanocotyle bullardi</i> Chero, Cruces, Sáez, Camargo, Santos & Luque, 2018	<i>Hypanus dipterurus</i>	Callao, Lima Region, Peru	Gills	Chero et al. [14]

<sup>a</sup>Probably either *Hy. americanus* or *Hy. say*<sup>b</sup>Probably *Hy. dipterurus*

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## Compliance with ethical standards

**Conflict of interest** The authors declare that they have no conflict of interest.

**Ethical approval** The authors declare that all applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

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