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VERSITA

Description of *Maritrema formicae* sp. nov. (Digenea, Microphallidae) parasitic in the kelp gull, *Larus dominicanus*, from the Patagonian coast, Argentina

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Abstract

Maritrema formicae sp. nov. is described from the Patagonian coast, Argentina, based on adults obtained from the kelp gull, *Larus dominicanus*. The new species fits with the "eroliae complex" and can be distinguished from other related species mainly in shape and size of body, shape, size, and pattern of distribution of cirrus spines, uterus extension, number and size of eggs, vitellarium in a complete ring in all specimens, and its Neotropical distribution. The new species is sympatric with another species of the genus, *Maritrema madrynense*, which was recorded in the same host and locality.

Keywords

Trematode digeneans, *Maritrema*, Microphallidae, *Larus*, Patagonia, Argentina

Introduction

Species of *Maritrema* Nicoll, 1907 (Microphallidae) are parasites mainly of marine birds; gastropods act as first intermediate hosts, and crustaceans primarily as second intermediate hosts (Deblock 1971; Yamaguti 1971, 1975). Three species of the genus have been previously reported from Argentinean waters, i.e., *Maritrema bonaerensis* Etchegoin et Martorelli, 1997 from *Larus atlanticus* Olrog, *Larus maculipennis* Lichtenstein, and *Larus dominicanus* Lichtenstein; *Maritrema orensensis* Cremonte et Martorelli, 1998 from *L. dominicanus* and *L. atlanticus*; and *Maritrema madrynensis* Diaz et Cremonte, 2010 from *L. dominicanus* (Etchegoin and Martorelli 1997, Cremonte and Martorelli 1998, La Sala *et al*. 2009, Diaz and Cremonte 2010).

As *Maritrema* is a neuter name (article 30.1.2 of the International Code of Zoological Nomenclature; from Greek τρήμα, neuter: a hole; type species: *Maritrema gratiosum* Nikoll, 1907), the original spelling *bonaerensis*, *orensensis*, *madrynensis*, *misenensis*, *interrupta*, *patulus*, *minuta*, *jilenensis* and *kitanensis* must be corrected to *bonaerense*, *orensense*, *madrynense*, *misenense*, *interruptum*, *patulum*, *minutum*, *jilenense* and *kitanense* for gender agreement (article 34.2).

During surveys of helminth communities in birds from the Argentinean Patagonian coast (Diaz and Cremonte 2010, Diaz *et al*. 2011), two *Maritrema* species were found parasitizing the intestine of the kelp gull, *Larus dominicanus*, *M. madrynense* and a new species which is described herein.

Materials and methods

Twenty nine kelp gulls, *Larus dominicanus* (Aves, Laridae), were collected from March 2000 to April 2004, in Peninsula Valdés and adjacent areas (24 adults from Fracasso Beach, 42°25′S, 64°07′W, San José Gulf, and 5 adults from Puerto Madryn Beach, 42°47′S, 65°02′W, Nuevo Gulf) of Chubut Province, Argentina. The birds were captured using a net, killed with ether, and transferred as soon as possible to the laboratory for dissection. The viscera were examined under a stereomicroscope. Adult digeneans belonging to the genus *Maritrema* were recovered from the intestine. The specimens were fixed in 5% hot formalin, stored in 70% ethanol, stained with Semichon's acetocarmine, cleared in methyl salicylate, mounted in Canada balsam, and studied using a light microscope (LM).

Illustrations were made with the aid of a drawing tube. Dimensions of mounted specimens are given in micrometers with the mean value followed by the range in parentheses. The forebody was measured from the anterior extremity to the anterior margin of ventral sucker. Some fixed adult specimens were dehydrated, critical point dried, gold coated, and ob-

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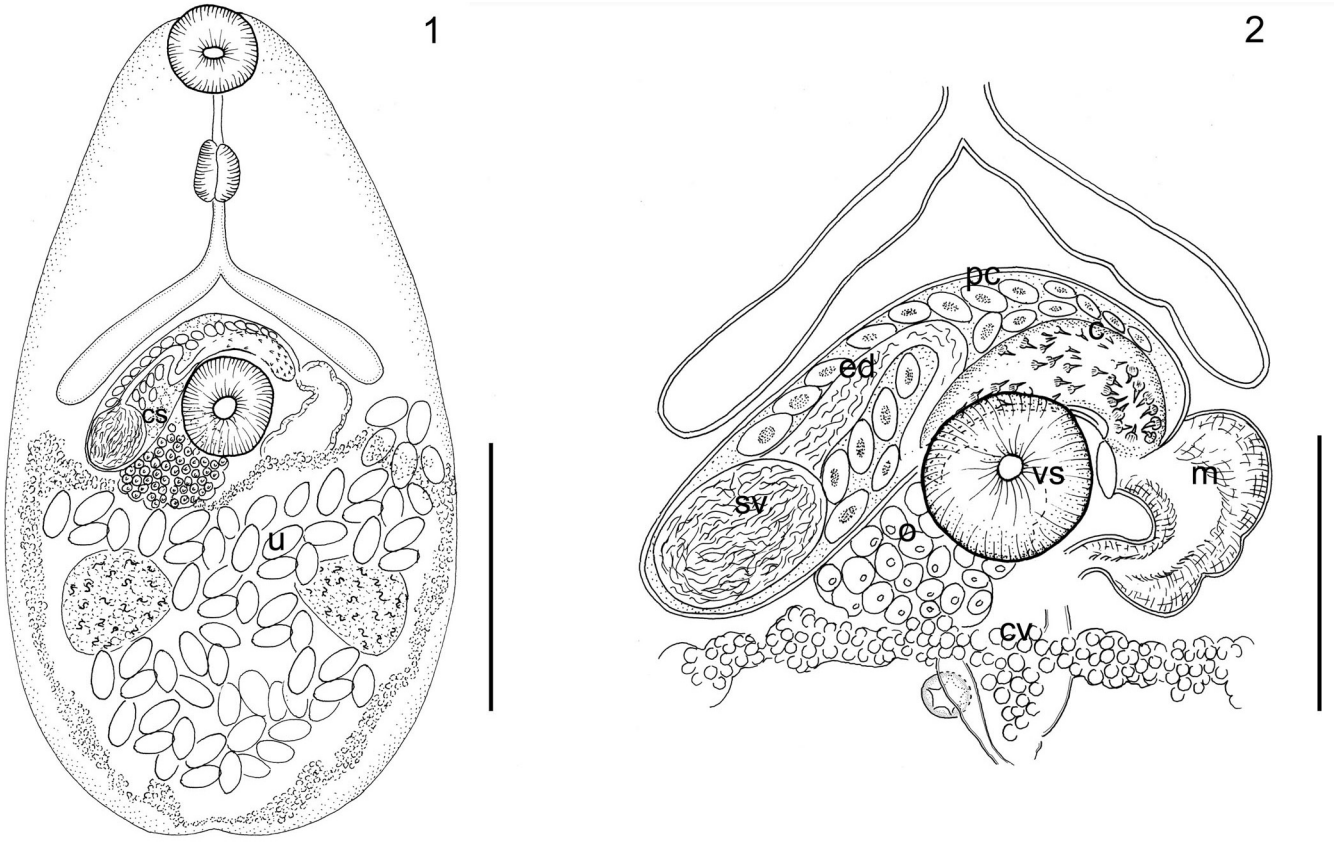
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served and photographed using a JEOL JSM 6360 LV (Jeol, Tokyo, Japan) scanning electron microscope (SEM).

Description

***Maritrema formicae* sp. nov.** (Figs 1–6)

Diagnosis (measurements based on 14 stained specimens) (Figs 1–6): Body "pyriform" (i.e. pointed at anterior extremity and rounded posteriorly), 269 (230–350) long by 130 (102–195) wide at ventral sucker level (Fig. 1). Spines transversally arranged covering entire body surface (Figs 3–5). Body spines multidentated, wider than longer (mean size 1.35 by 1.25), anterior spines compactly distributed (Figs 3–4). Oral sucker subterminal, 32 (26–43) long by 32 (26–40) wide. Ventral sucker 28 (23–40) long by 29 (24–40) wide, located in the second third of body. Forebody 135 (115–187) long. Sucker ratio (ventral sucker/oral sucker width) 0.91 (0.82–1). Prepharynx 13 (6–20) long. Pharynx 20 (17–28) long by 15 (13–18) wide. Oesophagus 27 (20–35) long. Intestinal caeca reaching the middle of ventral sucker. Testes postovarian, lateral, symmetrical, oval, smooth or slightly lobed edges (Fig. 1). Right testis 36 (30–45) long axis by 29 (23–40) short axis, left testis 36 (25–40) long axis by 29 (25–35) short axis. Vasa differentia joining anterior region of seminal vesicle. Cirrus sac shaped as inverted "J", 102 (85–130) long by 23 (18–30) maximum width (Fig. 2); located between intestinal caeca and ventral sucker; with a very thin muscular wall. Cirrus sac enclosing oval seminal vesicle, 27 (20–45) long by 20 (17–25) wide; numerous prostatic cells surrounding a curled ejaculatory duct, and a spinous cirrus (Fig. 2). Invaginated cirrus 41 (33–55) long. Cirrus spines irregularly distributed, not covering entire cirrus, densely distributed at base. Proximal cirrus-spines are rose-thorn shaped and larger than distal ones (5.5–6.5 *vs* 3.0–4.0, respectively) (Figs 2, 6). Ovary submedian, postacetabular, oval, slightly lobed, partially displaced dextrally, covered by posterior border of ventral sucker obliquely disposed, 42 (30–52) long axis by 28 (23–35) wide (Fig. 2). Oviduct short. Seminal receptacle present. Ootype and Mehlis' gland situated immediately posterior to ventral sucker. Laurer's canal not observed. Metraterm with thin muscular diverticulated wall. Genital pore simple, slightly differentiated, unarmed, located at left side of ventral sucker. Uterus in hindbody, surrounding testes, from posterior border of cirrus sac to end of body. Operculated eggs from 31 to approximately 100 in number, 16 (14–18) long by 8.5 (8–9) wide (Fig. 1). Vitellarium with numerous small follicles forming 2 symmetrical ribbons, confluent posteriorly, enclosing uterus and testes (Fig. 1). Excretory vesicle "Y" shaped.



Figs 1 and **2.** *Maritrema formicae* sp. nov. **1.** Naturally obtained adult, ventral view. **2.** Genital system, ventral view. **Abbreviations:** c, cirrus; cs, cirrus sac; cv, common vitelline duct; ed, ejaculatory duct; m, metraterm; o, ovary; pc, prostatic cells; sr, seminal receptacle; sv, seminal vesicle; u, uterus; vs, ventral sucker. Scale bars = 100 μm (1), 50 μm (2)

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Taxonomic summary

Type host: *Larus dominicanus* Lichtenstein (Aves, Laridae).

Type locality: Fracasso Beach (42°25´S, 64°07´W), Península Valdés, Chubut Province, Argentina.

Site of infection: Small intestine.

Prevalence: 17.3%.

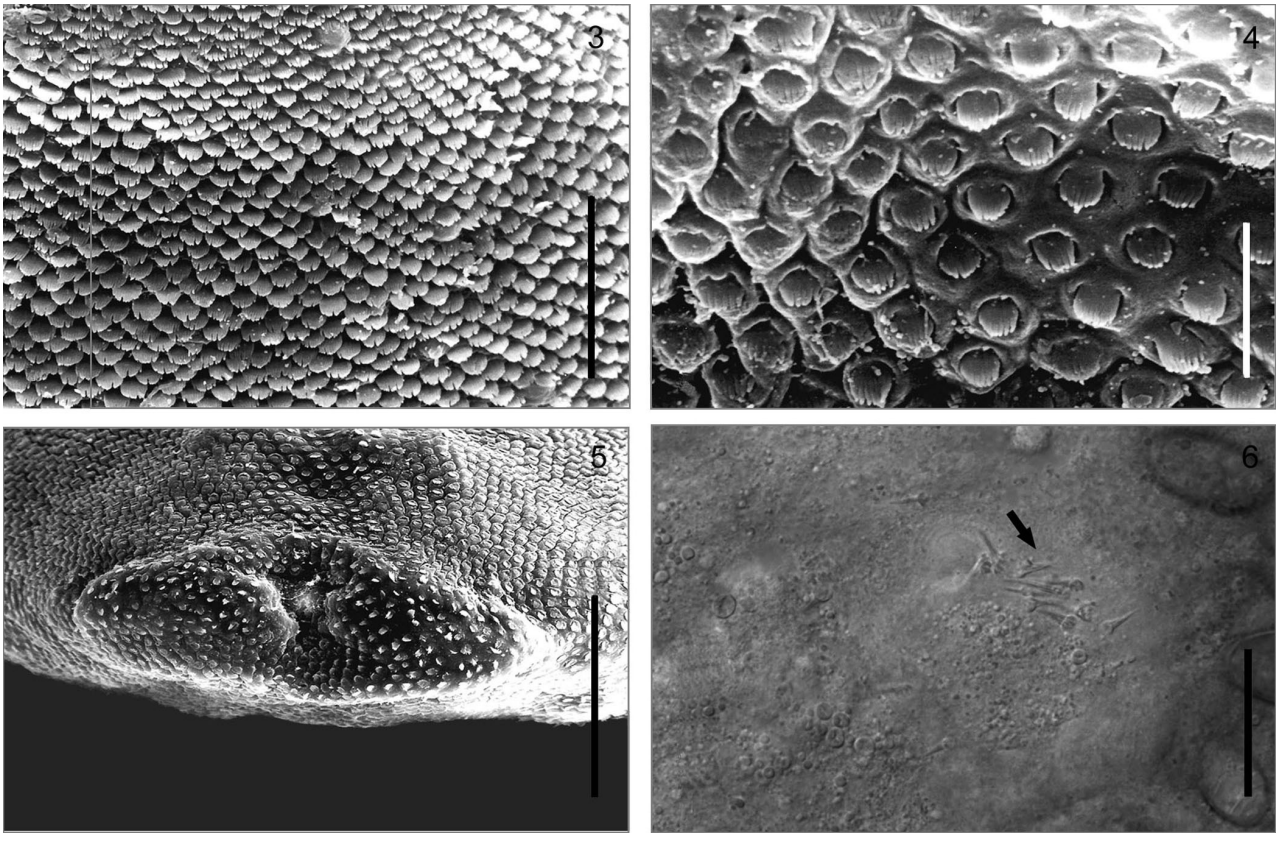
Specimens deposited: Holotype N° 6501 and paratypes N° 6502, voucher specimens N° 6503 at the Helminthological Collection of Museo de La Plata (CHMLP), Museo de La Plata, La Plata, and at the Parasitological Collection of the Centro Nacional Patagónico (CNP-Par N°13), Puerto Madryn, Argentina.

Etymology: The name of the new species is derived from the Latin for ant, "*formica*," as a tribute to "Hormiga", the nickname of our partner Miguel Angel Díaz (CENPAT-CONICET), indefatigable worker and lover of the sea. The species name is dedicated in his memory.

Remarks

The new species can be included in the genus *Maritrema* Nicoll, 1907 and in the subgenus *M*. (*Maritrema*) by the presence of postcaecal uterine coils, vitellarium in symmetrical ribbons surrounding uterine coils and testes, a cirrus pouch enclosing an invaginable cirrus like a finger of a glove turned inside out, a simple, superficial, and unarmed genital pore, and the absence of a spined acetabulo-atrial plate (Deblock 1971, 2008). The presence of a spinous cirrus enclosed in a cirrus sac places the new species closest to the "eroliae complex" proposed by Deblock and Canaris (1992), parasitizing mainly Charadriiformes. Within this complex are grouped several species, difficult to distinguish from each other, and very similar to *M. eroliae* Yamaguti, 1939. At present, twelve *Maritrema* species are included in the "eroliae complex". Deblock and Canaris (1992) grouped the species related to this complex into 3 subgroups according to body size, morphology of the vitellarium, and geographical distribution, i.e., group A = species smaller than 400 µm and with horseshoe-shaped vitellarium; group B = species larger than 600 µm and the vitellarium in a complete or subcomplete ring; and group C = some species with intermediate features between A and B.

Following this grouping, and taking account that *Maritrema formicae* sp. nov. has a vitellarium in a complete ring but they are smaller than 600 µm, it should be located near to group C, which is comprised of *Maritrema misenense* (Palombi, 1940), *M. eroliae* (sensu Deblock et Pearson, 1968), *Maritrema interruptum* Oshmarin, 1970, *Maritrema papillorobusta* Ke, 1976, and *M. madrynense* Diaz et Cremonte, 2010. In this group, only the smaller specimens of *M. eroliae*



Figs 3–6. *Maritrema formicae* sp. nov. **3–5.** Body spines (SEM). **3.** Preacetabular dorsal spines. **4.** Postacetabular dorsal spines. **5.** Posterior spines, ventral view. **6.** Details of cirrus spines (arrow) at LM. Scale bars = 10 µm (3), 5 µm (4), 30 µm (5), 20 µm (6)

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(sensu Deblock et Pearson, 1968) from *Charadrius mongolus* from Australia measure less than 400 µm like the new species. However, the Australian specimens were restudied and identified later as to *Maritrema spinosulum* Deblock et Canaris, 1996, a species that was described as having a horseshoe-shaped vitellarium, and the cirrus covered by a continuous lining of uniform small spines (Deblock and Canaris 1992, 1996). In contrast, *M. misenense* from Southern Europe, *M. interruptum* from Vietnam, *M. papillorobusta* from China, and *M. madrynense* from Argentina, measured more than 400 µm in length. In addition, *M. interruptum* has a horseshoe-shaped vitellarium and *M. papillorobusta* has the cirrus totally covered by heterogeneous spines, and a horseshoe-shaped vitellarium. Also, the eggs of the new species are smaller than those from *M. papillorobusta* (14–18 vs 25–28) (Prévot *et al*. 1976; Deblock and Canaris 1992, 1996; Diaz and Cremonte 2010). *Maritrema misenense* possesses the cirrus partially covered by spines like *M. formicae* sp. nov. (Prévot *et al*. 1976). However, *M. misenense*, which was described based only on experimental adults, has a horseshoe-shaped vitellarium and the tip of cirrus is unarmed. Also, the new species has shorter spines (5–6 vs 11) and smaller eggs (14–18 vs 19–22 long; 8–9 vs 11–12.5 wide) than *M. misenense*.

On the other hand, taking into account the taxonomic difficulties arising from incomplete descriptions, poorly illustrated, and the lack of biological knowledge of species similar to *M. eroliae*, Deblock and Canaris (1992) also considered those species similar to *M. eroliae* in 5 groups of species which are likely to be similar each other, in order to simplify their study. *Maritrema formicae* sp. nov. is similar to some species into "Group I or *patulus*" and "Group II or *eroliae*". However, all species in Group I have a horseshoe-shaped vitellarium and, with the exception of *Maritrema patulum* Coil, 1955, were found only in the Palearctic or Indomalaya Regions. The new species has smaller eggs than *Maritrema borneonse* Fischthal et Kuns, 1973 (14–18 vs 25–28). Moreover, in some of the species included in these two groups, the precise morphology, size or distribution of the cirrus spines were not described (i.e., *Maritrema minutum* Ke, 1976; *Maritrema jilenense* Zhong, Ke-Feng et Min-Zen, 1988; *Maritrema eroliae* sensu Ogata, 1951) (Deblock and Canaris 1992, 1996). *Maritrema kitanense* Shibue, 1953 and *Maritrema magnicirrus* Belopolskaia, 1952 were proposed as synonymies and/or declared invalid (Deblock 1975), and other species where restudied and found to have a smooth cirrus (i.e., *M. patulum*) (Deblock and Canaris 1996). Other species including in these groups I and II were considered and compared above (i.e., *M. misenense*, *M. interruptum* and *M. papillorobusta*).

Considering the valid species with a spinous cirrus and vitellarium in a complete ring, only *M. madrynense* has a Neotropical distribution (Diaz and Cremonte 2010). *Maritrema formicae* sp. nov. differs from *M. madrynense* in body shape (pyriform vs linguiform), size (230–350 long by 102–195 wide vs 400–690 long by 235–390 wide), cirrus size (33–55 vs 100–150), patterns of distribution of cirrus spines (partially vs totally covered), number of eggs (less to ~100 vs more than four hundred), and egg size (14–18 long by 8–9 in wide vs 18–23 long by 10–12). Size and shape of body spines in the new species differ from those of *M. madrynense*; in *M. formicae* sp. nov. they are smaller than in *M. madrynense* (average of dorsal body spines: 1.35 by 1.25 vs 2.74 by 4.52). *Maritrema formicae* sp. nov. and *M. madrynense* occur sympatrically in the same host species; however, differences between them are clear.

Discussion

Many species of *Maritrema* have been reported parasitizing avian hosts around the world (see Diaz and Cremonte 2010 and references therein). In contrast, only a few species have been reported in South American birds: *Maritrema nicolli* Travassos, 1920; *Maritrema bravoae* Caballero et Ibáñez, 1970; *Maritrema magdalenae* Werding, 1973; *M. bonaerense* Etchegoin et Martorelli, 1997; *M. orensense* and *M. madrynense* (see Deblock 1971; Caballero and Ibáñez 1970; Etchegoin and Martorelli 1997; Cremonte and Martorelli 1998; Diaz and Cremonte 2010). *Maritrema* spp. reported parasitizing birds of the genus *Larus* in Argentina are *M. bonaerense*, *M. orensense* and *M. madrynense* (Lunaschi *et al*. 2007, Diaz and Cremonte 2010, Diaz *et al*. 2011). Only *M. madrynense* fits with the "eroliae complex". Adults of this last species parasitized *L. dominicanus* and the metacercaria was found in the crab *Cyrtograpsus altimanus* Rathburn (Grapsidae) at the same locality as the new species (Diaz and Cremonte 2010, Diaz *et al*. 2011). Recently, Gilardoni *et al*. (2011) described larval stages of two different *Maritrema* species (i.e., *Maritrema* sp. 1 and *Maritrema* sp. 2) in a survey of larval digeneans in the intertidal snails from the northern Patagonian coast. Gilardoni *et al*. (2011) suggest that sporocysts and cercariae of *Maritrema* sp. 2 from the gastropod *Siphonaria lessoni* (Blainville) probably correspond to *M. madrynense*. Taking into account that only 2 adult *Maritrema* species (i.e., *M. madrynense* and *M. formicae* sp. nov.) were found in the study area, and also two different species were identified as larval stages in the same area, it is probable that *Maritrema* sp. 1 registered by Gilardoni *et al*. (2011) from *Crepipatella dilatata* Lamarck, corresponds to *M. formicae* sp. nov. Experimental and/or molecular analyses are needed to test these suggestions.

As was mentioned by Deblock and Canaris (1992, 1996) and Diaz and Cremonte (2010) there are doubts regarding the correct identification of specimens reported as *M. eroliae* by Yamaguti (1939) and subsequent authors, as well as other related species, as a result of incomplete descriptions, non or poorly illustrated specimens, and lack of knowledge about the biology of the species. Additionally, there are many descriptions and/or reports of metacercariae or immature specimens. *Maritrema formicae* sp. nov. can be distinguished from other species using a combination of features including size, shape of vitellarium, size and pattern of distribution of

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cirrus spines, number and size of eggs, and geographical distribution.

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