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Parasite communities of two sharks, *Etmopterus granulosus* (Squaliformes) and *Schroederichthys bivius* (Carcharhiniformes), from Southern Chile

Comunidades de parásitos de dos tiburones, Etmopterus granulosus (Squaliformes) y Schroederichthys bivius (Carcharhiniformes), del sur de Chile

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Abstract
The purpose of this study was to analyze the parasite community of two shark species from Southern Chile considering that the shark-parasite association in the area is poorly known. A total of 24 specimens of the southern lanternshark *Etmopterus granulosus*, and seven specimens of the narrowmouthed catshark *Schroederichthys bivius* were collected from the Strait of Magellan during January of 2017 and 2018. We recorded a total of 87 parasites associated with the examined *E. granulosus,* which belonged to seven species, and 192 parasites associated with the examined *S. bivius*, which belonged to other seven species. The higher average of parasite abundance was in *S. bivius* (27.5 ± 25.5 parasites/host) compared to *E. granulosus* (3.5 ± 3.3 parasites/host). A few parasites were prevalent and abundant; the nematodes *Pseudoterranova* sp. (41.7% prevalence; 1.46 parasites/host) and *Anisakis* sp. (29.2% prevalence; 1.21 parasites/host) present in *E. granulosus*, and the copepod *Tautochondria* sp. (85.7% prevalence; 9.14 parasites/host) and the nematode *Proleptus niedmanni* (71.4% prevalence; 14.86 parasites/host) present in *S. bivius*. Similitude in terms of parasite composition between the two shark species was low (14.6%), with two parasite species in common, the anisakid nematodes *Pseudoterranova* sp. and *Anisakis* sp. Thus, despite the sympatry of the two shark species analyzed in this study, the parasite composition was significantly different. The difference in parasite composition of the *E. granulosus* and *S. bivius* could be due to their evolutionary history, involving several differences in the ecology of the hosts.

Key words:
Parasite composition, southern lanternshark, narrowmouthed catshark, Strait of Magellan.

Resumen
En la presente investigación se analizó la comunidad de parásitos de dos especies de tiburones del sur de Chile debido al escaso conocimiento de la relación entre parásitos y tiburones de la zona. Un total de 24 especímenes del tiburón linterna *Etmopterus granulosus* y siete especímenes de la pintarroja del sur *Schroederichthys bivius* fueron recolectados en el estrecho de Magallanes en enero de 2017 y 2018. Se encontró un total de 87 individuos de parásitos pertenecientes a siete especies de parásitos en *E. granulosus* y 192 individuos de parásitos de siete especies en *S. bivius*. La abundancia promedio de parásitos fue mayor en la pintarroja del sur que en el tiburón linterna, con 27,5 ± 25,5 parásitos/hospedero y 3,5 ± 3,3 parásitos/hospedero, respectivamente. Pocos parásitos fueron prevalentes y abundantes; tales como los nematodos *Pseudoterranova* sp. (41,7% de prevalencia; 1,46 parásitos/pez) y *Anisakis* sp. (29,2% de prevalencia; 1,21

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parásitos/pez) presentes en *E. granulosus*, y el copépodo *Tautochondria* sp. (85,7% de prevalencia; 9,14 parásitos/pez) y el nematodo *Proleptus niedmanni* (71,4% de prevalencia; 14,86 parásitos/pez) presentes en *S. bivius*. La similitud en la composición de especies parásitas fue baja entre *E. granulosus* y *S. bivius* (14,6%), ya que sólo dos especies de parásitos, los anisákidos *Pseudoterranova* sp. y *Anisakis* sp., fueron compartidos entre ambos hospederos, a pesar de ser especies simpátricas. Por tanto, la desigual composición de los parásitos entre ambas especies de tiburones podría explicarse por sus historias evolutivas distintas, que implican numerosas diferencias ecológicas entre los hospederos.

Palabras clave:
Composición de parásitos, tiburón linterna, pintarroja del sur, estrecho de Magallanes.

INTRODUCTION

Parasites are often studied to understand the health conditions of their hosts, but they are also important biological elements that complement ecological studies, such as trophic webs and interactions among species. In this regard, sharks have an important ecological role in the trophic web of their habitats because they are predators of a wide range of prey, usually including crustaceans and fishes, as well as marine birds and mammals. Thus, many sharks are considered key species that regulate populations of organisms located at lower levels of the local food web (Bornatowski *et al.* 2014; Roff *et al.* 2016). As a result, changes in abundance of a certain prey in a habitat may also affect their associated parasite populations. Similarly, if the abundance of a shark species changes, the population and community of their associated parasites may also change. Thus, parasites somehow respond to changes in the host population and environment (Marcogliese, 2005). As apex predators, sharks may be the hosts for many parasites; however, very few studies have addressed this topic.

Parasite communities associated with sharks are poorly documented in Chile. For example, among 58 shark species recognized in this region (Sáez *et al.* 2012), only 11 of them contain records of ectoparasites (Muñoz & Olmos, 2007) and nine have records of endoparasites (Muñoz & Olmos, 2008). Most of these records are referenced to taxonomic studies of copepods (*e.g.*, Castro & Baeza, 1986; Véliz *et al.* 2018) and cestodes (*e.g.*, Carvajal, 1974). Also, the minimal existing information about parasites in sharks only refers to northern and central Chilean coasts. One study was focused on a nematode of *Schroederichthys chilensis* (Guichenot, 1848) from the central-south of Chile (38-41°S) (Torres & Grandjean, 1983). Meanwhile, only one study has been conducted on the parasite communities of the southern lanternshark, *Etmopterus granulosus* (Günther, 1880), and the largenose catshark, *Apristurus nasutus* de Buen, 1959, both from northern Chile (Espínola-Novelo *et al.* 2018). Although there are at least eight species distributed up to the Strait of Magellan (54-55°S) (Reyes & Hüne, 2012), there are no records of their parasites from this part of the Pacific coast. Due to this lack of information, we focus on two shark species, *E. granulosus* and the narrowmouthed catshark *Schroederichthys bivius* (Müller & Henle, 1838).

E. granulosus is distributed around the south American cone (Pacific and Atlantic coasts, including Falkland Islands), usually between 26°S and 53°S, and it may reach deeper waters, up to 950 m (Reyes & Hüne, 2012). *S. bivius* shows the same latitudinal distribution, but at depths of 45 m to 199 m (Sánchez *et al.* 2009). Both shark species prey on several kind of animals, such as polychaetes, decapod crustaceans, cephalopods, actinians, although bony fish is one of the most important food items (Reyes & Hüne, 2012; Sánchez *et al.* 2009). *E. granulosus* and *S. bivius* are non-target species for the commercial fisheries, however they are caught in the by-catch. Moreover, there are no records of parasites from *S. bivius*, and parasites from *E. granulosus* are only known in the northern Chile. Therefore, the objective of this study was to describe the parasite communities of *E. granulosus* and *S. bivius* collected from the Strait of Magellan, Pacific Coast. Additionally, morphological and morphometric data of the parasite species were also provided.

MATERIALS AND METHODS

Collection and identification of sharks and parasites

A total of 24 specimens of *E. granulosus* and seven of *S. bivius* were collected from the Strait of Magellan (53°19'44.0832''S; 70°45'30.6576''W) at south of Punta Arenas city, Southern Chile, at a depth of *ca.* 50 m. Sixteen *E. granulosus* were collected in January 2017, while eight other specimens, including all the specimens of *S. bivius*, were collected in January 2018. The spinel method was used onboard of an artisanal boat, and sharks were obtained as bycatch during the austral hake summer fishing.

All the shark specimens were frozen at -20°C, and thawed and dissected posteriorly. The identification of the sharks was confirmed using the taxonomic characteristics listed by Lamilla & Bustamante (2005) and Sáez *et al.* (2012). Total body length was recorded for each specimen. Metazoan parasites from each shark were sought out from all body parts, such as body surface, gills, nose cavities, digestive tract, gonads, liver, brain, spleen, kidney, body cavity, and muscles. The spiral valve was dissected with a longitudinal cut and then several transversal cuts were made to collect all the parasites observed. Parasites gathered were then fixed in either 5% formalin or 70% ethanol, according to the processes applied for identification.

Platyhelminthes (monogeneans, digeneans, and cestodes) were stained with haematoxylin, dehydrated in an alcohol series from 70 to 100%, cleared in methyl salicylate, and mounted in Entellan. Measurements and drawings were made with a "camera lucida" attached to a Leica DM LS2 light microscope (Leica microsystems, Wetzlar, Germany). Copepods, nematodes, and cestodes were used for scanning electron microscopy (SEM), to observe the body, and particularly the cephalic and tail characteristics of copepods and nematodes, and the scolex of cestodes. Specimens were initially fixed in 5% formalin and then dehydrated through an ethanol series (70-100%), followed by critical point drying in CO_2 using a Samdri-780A machine (Tousimis Research Corporation, Rockville, MD, USA), sputter-coated with gold using an Ion JFC-1100 Sputter machine, and examined with a JEOL T-300 SEM (JEOL, Tokyo, Japan). Nematodes were cleared with 50% glycerine (diluted in absolute ethanol) for morphometry measurements with a light microscope.

Parasites were identified to the lowest taxonomic level possible using the morphological and morphometric characteristics described by Campbell & Carvajal (1987), Chisholm *et al.* (1997), Fernández & Villalba (1985), Ho (1987), Hurst (1984), Kabata (1986), Knoff *et al.* (2001), Li *et al.* (2013), Robinson (1961), Rodríguez *et al.* (2010), Ruhnke (1996), Specian *et al.* (1975), Threlfall & Carvajal (1984) and Torres & Grandjean (1983). Because parasites of sharks in Chile were poorly known, some of their morphological aspects were recorded to contribute to the understanding of the species. Additional information, such as photographs (using optical or electronic microscopy) and drawings of parasites, were also provided.

Parasitological descriptors and statistical analyses

The abundance and prevalence of each parasite species were recorded and averaged for each shark species sample. The abundance and species richness of the parasite infracommunity were calculated for each shark specimens and then averaged for each host species sample (*sensu* Bush *et al.* 1997).

It is well known that the host sample size directly affects parasite species richness (Poulin, 1998; Zelmer & Esch, 1999), therefore three nonparametric estimators of parasite species (Chao 2, Jacknife 1 and 2) were applied, using presence-absence of parasite species, to ascertain whether the sample sizes of sharks were sufficient to know their parasite diversity. A Spearman correlation analysis (Zar, 1996) was used to verify the potential effect of fish body length on abundance and species richness of parasites.

RESULTS

Morphological characteristics of parasites

Seven parasite species were found in the southern lanternshark *E. granulosus* and seven parasite species were found in the narrowmouthed catshark *S. bivius* (Table 1). Altogether, twelve

Table 1. Composition of parasites and population descriptors (PRE%: Prevalence, and ABU±SD: average abundance ± standard deviation of individuals) and community descriptors of parasites found in two sharks from southern Chile. The total length (TL) ± standard deviation is also given for each host species.

Parasite taxa	*Etmopterus granulosus* (n= 24)		*Schroederichthys bivius* (n= 7)	
	TL: 53.5 ± 3.9 cm		TL: 40.5 ± 5.4 cm	
	PRE%	ABU±SD	PRE%	ABU±SD
COPEPODA				
Neoalbionella sp.	4.2	0.04 ± 0.20	0	0
Tautochondria sp.	0	0	85.7	9.14 ± 10.24
Lernaeopoda bivia	0	0	42.9	0.57 ± 0.79
MONOGENEA				
Calicotyle sp.	8.3	0.08 ± 0.28	0	0
Asthenocotyle kaikourensis	8.3	0.12 ± 0.28	0	0
DIGENEA				
Otodistomum veliporum	8.3	0.08 ± 0.28	0	0
CESTODA				
Phyllobothriidae gen. sp.	29.2	0.42 ± 0.78	0	0
Tetraphyllidea gen. sp.	0	0	14.3	0.14 ± 0.38
NEMATODA				
Heliconema sp.	0	0	28.6	1.42 ± 2.99
Proleptus c.f. *niedmanni*	0	0	71.4	14.86 ± 12.94
Pseudoterranova sp.	41.7	1.46 ± 2.92	14.3	0.43 ± 1.13
Anisakis sp.	29.2	1.21 ± 2.55	14.3	0.14 ± 0.38
Anisakidae undetermined	16.7	0.21 ± 0.51	0	0
COMMUNITY DESCRIPTORS				
Average abundance ± SD	3.5 ± 3.3		27.5 ± 25.5	
Average richness ± SD	1.4 ± 0.9		2.5 ± 1.8	
Total prevalence (%)	87.5		100	

parasite taxa were found in both sharks. Morphological distinctions of them are given below.

Phylum: Arthropoda
Subclass: Copepoda
Family: Lernaeopodidae
Species: *Neoalbionella* sp. (Fig. 1 A-C)

Only one specimen was found attached to a pectoral fin of *E. granulosus*. Body length, 4.54 mm × 2.14 mm wide. Cephalothorax short, 1.27 mm long. Trunk, 3.27 mm long. Second maxilla, 2.07 mm long. Antennule with four segments with setal elements formula as 0, 1, 1, 5 (from base to apex). Antenna birramous, with the exopod unsegmented and with lobated and denticulate apical part. Ovigerous sacs oval, 2.43 mm long.

Remarks: Unidentified specimens of *Neoalbionella* were recorded and characterized by Rodríguez *et al.* (2010) on *E. granulosus* from the Juan Fernández Archipelago, South Pacific (*ca.* 670 km west from the central Chilean coast).

These authors found that the armature antennules, antenna and mandible shape of its specimen was distinct from the other nine known species in the genus *Neoalbionella* (see key of species in Ruiz & Bullard, 2019): *N. globose* (Leigh-Sharpe, 1918), *N. centroscyllii* (Hansen, 1923), *N. fabricii* (Rubec & Hogans, 1988), *N. oviformis* (Shiino, 1956), *N. kabatai* (Benz & Izawa, 1990), *N. longicaudata* (Hansen, 1923), *N. etmopteri* (Yamaguti, 1939), *N. benzipirata* Ruiz & Bullard, in Ruiz, Driggers & Bullard, 2019, and *N. dannytangi* Ruiz & Bullard, 2019. The features of the *Neoalbionella* specimen in the present study agree with the distinctions reported previously by Rodríguez *et al.* (2010). Unfortunately, this species has not formally been described.

Species: *Tautochondria* sp. (Fig. 1D)

Sixty-four female specimens found in the olfactory bulbs of *S. bivius*. Four specimens measured. Total body length 5.16-5.76 mm, divided into head, short neck, trunk, and tail. Head almost oval, 1.17-1.47 mm long × 1.47-1.64 mm wide. Neck without processes. Robust trunk, 2.23-2.82 mm long × 1.38-1.76 mm wide. Tail, 1.35-1.76 mm long, with a wide base. Trunk with a pair of short posterior lateral processes, 0.352-0.470 mm long. Coiled egg sacs.

Remarks: Only one species has been described in the genus *Tautochondria*; *T. dolichoura* Ho, 1987 found on the abyssal fish *Anoplogaster cornuta* (Valenciennes, 1833) in Newfoundland, Canada (Ho, 1987). This species bears lateral processes on the neck and two long processes at the posterior side of the trunk, which are not present in the *Tautochondria* specimen from the present study. Therefore, this is a new species that requires description.

Species: *Lernaeopoda bivia* Leigh-Sharpe, 1930 (Fig. 1 E-F)

Four specimens found on the tongue of *S. bivius*. All of them measured. Total body length, 7.14-8.26 mm from head to tail. Short cephalothorax, 1.68-2.58 mm long. Trunk, 3.78-5.04 mm long × 1.82-1.960 mm maximum wide. Two posterior processes, 1.12-1.51 mm long. Second maxilla 3.78-5.04 mm long. Ovigerous sacs 2.38-3.92 mm long.

Remarks: *Lernaeopoda bivia* has been recorded from the cloaca of *S. bivius* (= *Scyllium bivium*), from Orange Bay, Argentina(?)[1] (Kabata, 1986). Most of the morphological characteristics of *L. bivia* were observed in the specimens found on *S. bivius* in the present study. This is the first record of *Lernaeopoda bivia* in fish from Chilean waters.

Phylum: Platyhelminthes
Class: Monogenea
Family: Monocotylidae
Species: *Calicotyle* sp. (Fig. 2A)

Two specimens found on flanks of body surface of *E. granulosus*. The conditions of these specimens were not good, and some morphological characteristics could not be observed. Two specimens cleared and measured. Flat and pyriform body shape, 6.86-11.66 mm long × 5.48-11.11 mm maximum wide (almost at posterior side). Haptor diameter, 1 mm. Pharynx rounded to oval, 0.360-0.638 mm long × 0.328-0.638 mm wide. Haptor with 7 loculi. Haptoral anchors not observed.

Remarks: *Calicotyle* has been recorded in several elasmobranchs worldwide (Chisholm *et al.* 1997). In Chile, unidentified *Calicotyle* specimens were also recorded in the *E. granulosus* from northern zone (Espínola-Novelo *et al.* 2018), although these authors did not mention the morphological features of this monogenean. Specimens found in the present study were similar in body shape (pyriform and a relatively small haptor) to two other species, *C. ramsayi* Robinson, 1961 and *C. splendens* (Szidat, 1970). Nevertheless, *C. ramsayi* is host-specific, but several other *Calicotyle* are not (Chisholm *et al.* 1997). *Calicotyle* sp. had a larger body size and smaller pharynx compared to the other two species mentioned before. However, haptoral anchors and specific characteristics of the reproductive system were not properly observed; therefore, we were unable to identify the specimens.

1 Kabata (1986) said that the shark sample was from Argentina, however, in the original study (Leigh-Sharpe, 1930) the country was not indicated.

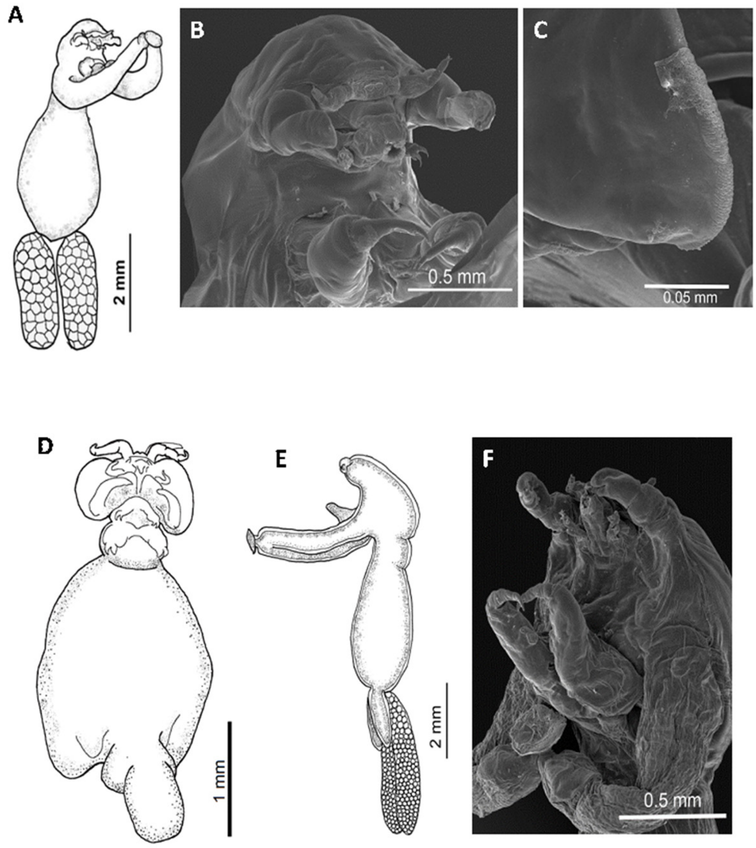

Fig. 1. Parasitic copepods: A) ventrolateral view of the whole body, B) SEM of ventral view of head, C) apex of antenna exopod of *Neoalbionella* sp., D) ventral view of the whole body of *Tautochondria* sp., E) lateral view of the body, and F) SEM of ventrolateral view of the head of *Lernaeopoda bivia*.

Family: Microbothriidae

Species: *Asthenocotyle kaikourensis* Robinson, 1961 (Fig. 2B)

Three specimens found on the host body surface of *E. granulosus*. All of them stained and measured. Body length, 8.5-9.4 mm long × 1.8-2.2 mm wide. Short prepharynx, 0.350-0.500 mm long. Pharynx oval, 0.45-0.55 mm long × 0.330-0.395 mm wide. Genital pouch slightly oval 0.675-0.912 mm long × 0.720-0.765 mm wide. Seminal vesicle external large and slightly oval, 0.650-0.700 mm long × 0.440-0.500 mm wide, posterior to germarium. Muscular ejaculatory bulb, 0.350-0.360 mm long × 0.230-0.270 mm wide connected to a sclerotized copulatory duct, 0.75-0.81 mm long. Between 46-47 testes rounded to oval, located in the central zone of the body. Germarium slightly oval, 0.237-0.240 mm long × 0.282-0.322 mm wide, located at 0.550-0.750 mm from the genital pouch. Vitellogen follicles mostly distributed along both lateral sides of the body, posterior to pharynx, up to 0.625-0.750 mm from the posterior edge. One monogenean with one oval egg, 0.350 mm long × 0.140 mm wide. No filament observed.

Remarks: *Asthenocotyle kaikourensis* has been recorded on the body surface of *Scymnodon plunketi* (Waite, 1910), from Kaikoura, New Zealand. An unidentified species of *Asthenocotyle* was found on *E. granulosus* from northern Chile (Espínola-Novelo *et al.* 2018). In the present study, specimens' morphology was consistent with that of *A. kaikourensis*, and differed from other species of the genus (*A. taranakiensis* Beverley-Burton, Klassen & Lester 1987 and *A. azorensis* Kearn, Whittington & Thomas, 2012) by the number of testes and sizes of reproductive organs (ejaculatory bulb, genital pouch, and germarium).

The wide distribution of some parasites depends on their host specificity and the swimming capacity of fish to disperse them (Terui *et al.* 2017). However, there is little information about the spatial distribution and host specificity of *Asthenocotyle kaikourensis*. Therefore, in the future, using molecular markers, it could be possible to confirm the identity of this monogenean in Chile.

Subclass: Digenea
Family: Azygiidae
Species: *Otodistomum veliporum* (Creplin, 1837) (Fig. 2C)

Two mature specimens were found in the stomach of *E. granulosus*. One specimen stained and measured. This is a large digenean, 32.0 mm length × 3.5 mm wide. Oral sucker rounded, 0.986 mm in diameter; Pharynx oval, 0.558 mm length × 0.369 mm wide. Ventral sucker, 1.23 mm length × 1.396 mm wide. Forebody, 3.32 mm length. Genital pore located 1.93 mm from the anterior edge. Terminal genitalia not observed. Ovary oval and rounded, 1.6 mm in diameter, located at equatorial zone. Two oval testes in tandem, 0.887-0.970 mm length × 0.608-0.612 mm wide. Numerous vitellogen follicles located along lateral sides, distributed from 8.4 mm, from the anterior edge to 7.4 mm from the posterior edge. Uterus long and convoluted, distributed between ventral sucker and ovary. Egg oval, 0.007-0.009 mm length × 0.005-0.006 mm wide.

Remarks: *Otodistomum veliporum* is the only valid species in the genus, and has been found in fish from the southern Atlantic and Pacific coasts of South America (Kohn *et al.* 2007). Previously, it has been recorded as *Otodistomum cestoides* (van Beneden, 1871) in batoids, such as *Psammobatis scobina* (Philippi, 1857) and *Dipturus flavirostris* (Philippi, 1892) in San Antonio, Central-Chile (Threlfall & Carvajal, 1986), and in the stomach of the fish *Helicolenus lengerichi* Norman, 1937, from Talcahuano, Central Chile (George-Nascimento & Iriarte, 1989). Recently, *Otodistomum* sp. was recorded in *E. granulosus* from Northern Chile (Espínola-Novelo *et al.* 2018).

Subphylum: Cestoda
Order: Phyllobothriidea
Family: undetermined
Species: undetermined (Fig. 2 D-E)

Ten mature specimens were found in the spiral valve of *E. granulosus*. One specimen measured and another specimen used for SEM. Body length 49.0 mm × 0.9 mm wide. Scolex length 1.75 mm, with four sessile bothridia. Apical glandular organ not observed. Sides of each bothridium folded without loculi. Each bothridium with an accessory sucker, 0.23-0.26 mm in diameter Strobila craspedote. Mature proglottids 1.62-2.25 mm long. Vitellogenic follicles located at the sides of proglottids. Lobed ovary at the posterior region. Numerous rounded testes anterior to the ovary. Uterus depleted with eggs distributed along the proglottid at the central part. Genital pore bulky at one of the lateral sides of each proglottid.

Remarks: Specimens resemble to *Phyllobothrium*, according to Ruhnke (1996). Four species of this genus have been recorded in Chile (Muñoz & Olmos, 2008): as larval stage in *P. delphini* (Bosc, 1802), and adult stages of *P. discopygi* Campbell and Carvajal, 1987 in *Discopyge tschudii* Haeckel, 1845, *P.* c.f. *lactuca* Van Beneden, 1850 in *Dipturus trachyderma* (Krefft & Stehmann, 1975); and *P. sinuosiceps* Williams, 1959 in *Hexanchus griseus* (Bonnaterre, 1788). However, from these four species, only *P. lactuca* has been confirmed as a valid species (Caira *et al.* 2020).

Phyllobothrium lactuca has been found in several elasmobranchs from different parts of the world, and its identification has been deficient (Ruhnke, 1996). Therefore, there is still no clarity about its morphological distinctions and geographical distribution. However, *Phyllobothrium* sp., from this study, has a shorter scolex length, larger suckers, and less folded bothridia than *P. lactuca*, according to the description given by Ruhnke (1996).

Recently, molecular markers have been used in studies about cestodes (Caira *et al.* 2020), and a great reorganization of genera within Phyllobothriidea was done. Consequently, morphological descriptions of genera have changed to organize the group. Therefore, we cannot confirm

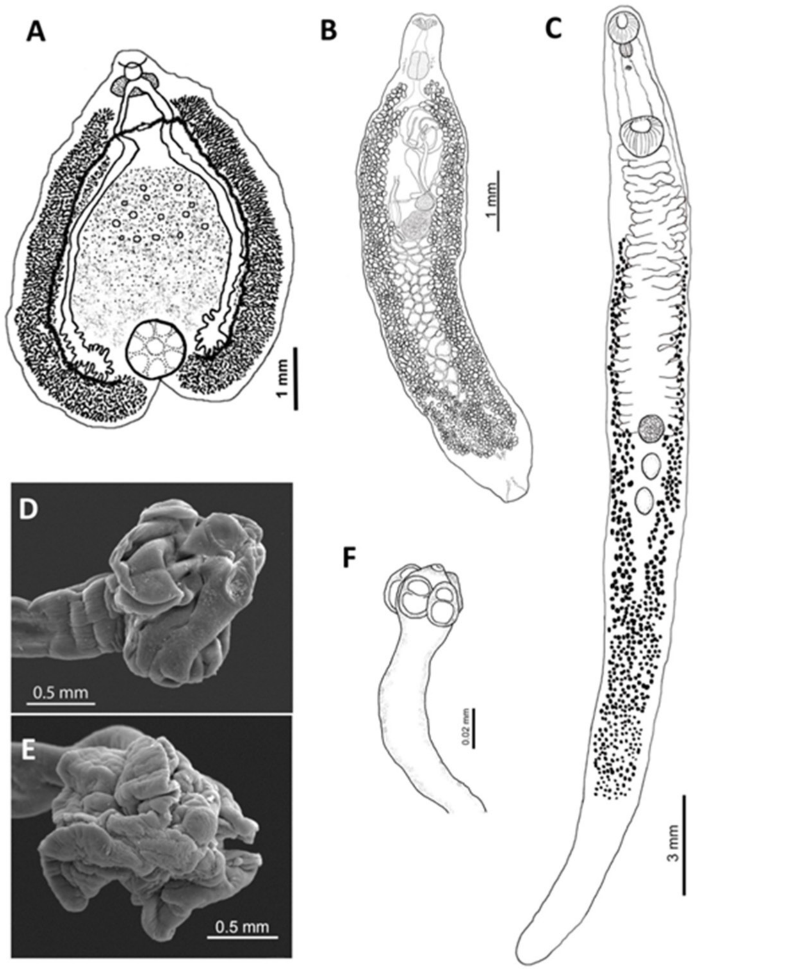

Fig. 2. Parasitic platyhelminthes. A) whole body of *Calicotyle* sp., B) whole body of *Asthenocotyle kaikourensis*; C) whole body of *Otodistomum veliporum*, D) lateral view, and E) apical view of the scolex of a Phyllobothriidea specimen. F) Whole body of a larval cestode.

the species found here, and we prefer to designate the specimens as Phyllobothriidea gen. sp.

Family: undetermined

Species: undetermined (larva) (Fig. 2F)

Only one specimen at the larval stage was found in the intestine of *S. bivius*. Body length at least 1.83 mm (specimen cut at the posterior part of the body) and 0.022 mm wide. Scolex with an apical sucker and four bothridia. Apical sucker, 0.069 mm of diameter. Bothridia, 0.239-0.267 length × 0.135-0.142 wide mm, with free posterior ends, divided into two loculi.

Remarks: In the past, this kind of larva would be assigned as Tetraphyllidea, which was a group reported in several fish, such as elasmobranchs and bony fish from Chile (Espínola-Novelo *et al.* 2018; Muñoz & Olmos, 2008). The identification of theses larval cestodes was based in the presence of four bothridia, however, Caira *et al.* (2014) demonstrated that species from the Tetraphyllidea group was not supported, instead several species of different genera and orders conform another group (Order: Onchoproteocephalidea). Therefore, for this study, it was not possible to identify this larval cestode.

Phylum: Nematoda

Class: Secernentea

Family: Physalopteridae

Two species found only in *S. bivius*. These nematode species showed similar cephalic characteristics; however, they differed in some reproductive morphology, indicated below.

Species: *Proleptus* c.f. *niedmanni* Torres and Grandjean, 1983 (Fig. 3 A-C)

One hundred-four specimens were found in the stomach of the host. Three males and three females were measured. Cephalic characteristics are similar between sexes and like *Heliconema*. Mouth opening elongate, with a pair of pseudolabia, each with a conical tooth. An amphid at each lateral side of the mouth. Four cephalic papillae, one pair dorsally and one pair ventrally situated. Cephalic cuticular collar. Female with ventral vulva, close to the anus. Male with nine caudal papillae. Left spicule 4 times longer than the right spicule.

Male body length 31.51-36.35 mm × 0.509-0.686 mm wide. Muscular esophagus 0.404-0.681 mm long. Glandular esophagus 2.58-4.48 mm long. Nerve ring at 0.410-0.685 mm from the anterior edge. Right spicule 0.462-0.502 mm long, a left spicule 1.82-2.14 mm long. Tail conical 0.862-1.071 mm long. Caudal papillae: three precloacal pairs, one paracloacal pair, and 5 postcloacal pairs.

Female body length 40.6-48.2 mm × 0.230-0.880 mm wide. Vulva at 1.05-1.15 mm from the posterior edge, and at 0.380-0.570 mm from the anus. Tail conical, 0.580-0.680 mm long. Oval eggs, 0.050-0.060 mm long × 0.030-0.040 mm width.

Remarks: Two species of *Proleptus* have been recorded in Chile, *P. carvajali* Fernández & Villalba, 1985 and *P. niedmanni*, both from elasmobranchs. Specifically, *P. niedmanni* was recorded in *Schroederichthys chilensis* from south-central Chile, and the morphometry mostly agrees with the specimens found in this study for *S. bivius*, although with differences in the esophagus length and caudal papillae. Esophagus lengths were shorter in our specimens possibly related to a smaller body length than *P. niedmanni* found in *S. chilensis* (31-36 mm *vs* 36-51 mm for males; 40-48 mm *vs* 39-61 mm for females). Torres & Grandjean (1983) also indicated six pairs of postcloacal papillae in male nematodes and noted that some papillae were very close to the anus. We considered a paracloacal pair of papillae that was in the cloacal zone, although this pair was slightly posterior to cloacal opening. Therefore, to count five or six pairs of postcloacal papillae depend on the author's criteria.

Species: *Heliconema* sp. (Fig. 3 D-F)

Ten specimens found in the stomach of the host. Two females and one male measured. Cephalic characteristics are similar between sexes. Mouth opening elongated, a pair of pseudolabia, each with a conical tooth. An amphid at each lateral side of the mouth. Four cephalic papillae, one pair dorsally and one pair ventrally situated. A cephalic cuticular collar. Female with vulva ventral situated in the third posterior portion of the body. Male with twelve caudal papillae. Left spicule 4 times longer than the right spicule.

Male body length 30.50 mm × 0.524 mm wide. Muscular esophagus 0.371 mm long. Glandular esophagus 3.39 mm long. Nerve ring to 0.514 mm from the anterior edge. Right spicule 0.696 mm long, left spicule 2.32 mm long. Tail conical, 0.712 mm long. Caudal papillae: four precloacal pairs, two paracloacal pairs, and six postcloacal pairs. A pair of phasmids.

Female body length 17.67-19.87 mm × 0.374-0.448 mm wide. Muscular esophagus 0.389-0.501 mm long. Glandular esophagus 2.00-2.18 mm long. Nerve ring at 0.342-0.810 mm from the anterior edge. Vulva not prominent, at 3.98-7.91 mm from the posterior edge. Tail conical, 0.498-0.526 mm long.

Remarks: *Heliconema psammobatidus* Threlfall and Carvajal, 1984 is the only species of the genus *Heliconema* recorded in *Sympterygia lima* (Poeppig, 1835) (=*Psammobatis lima*) in Central Chile (Threlfall & Carvajal, 1984). The *Heliconema* sp. from the present study differed to *H. psammobatidus* in the number of caudal papillae (10 pairs *vs* 12 pairs, respectively). Considering the key for species of *Heliconema* given by Akram (1996), *Heliconema* sp. also differed from the other ten species in the number of caudal papillae.

It is important to note that *Proleptus* and *Heliconema* specimens are similar in cephalic morphology and body sizes. The distinction between *Proleptus* c.f. *niedmanni* and *Heliconema* sp. are in the vulva position and number of caudal papillae.

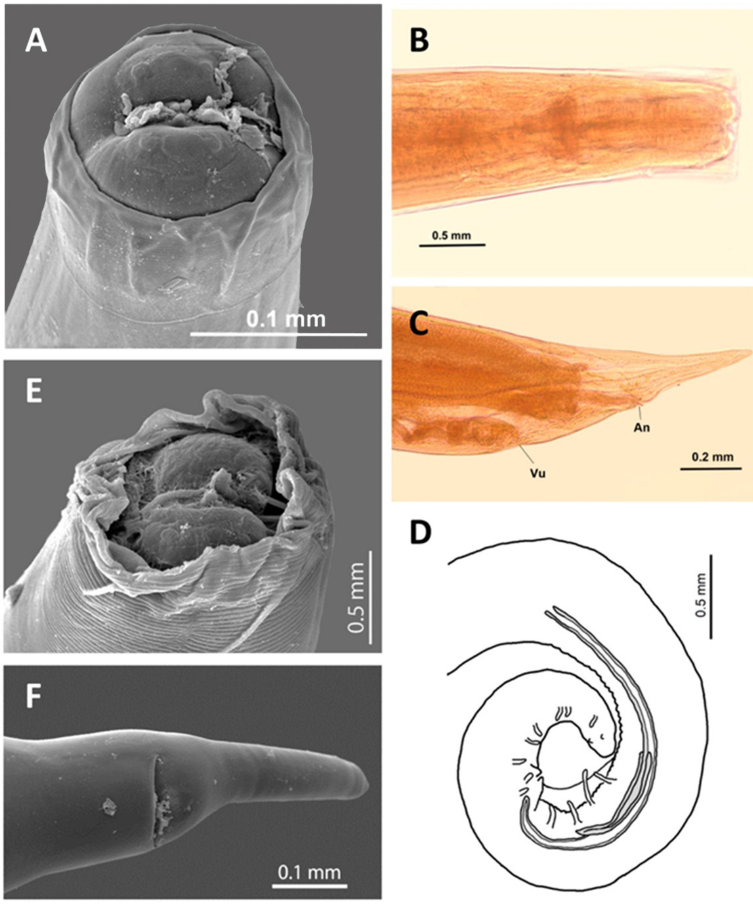

Fig. 3. Nematodes of Physalopteridae: A) SEM of cephalic apical view, B) anterior region of the body, and C) posterior region of a female (Vu: vulva, An: anus) of *Proleptus niedmanni*, D) SEM of apical cephalic view, E) SEM of a female tail and F) drawings of a male tail of *Heliconema* sp.

Family: Anisakidae

Species: *Pseudoterranova* sp. (Fig. 4 A-B)

Thirty-five specimens at larval stage were found encysted in the body cavity and stomach wall of *E. granulosus*, and three larval specimens were found in the body cavity of *S. bivius*.

Five specimens of *E. granulosus* were measured. Body length 18.85-28.94 mm × 0.415-0.487 mm wide. Nerve ring at 0.172-0.195 mm from the anterior edge. Muscular esophagus, 0.906-1.125 mm long. Ventricle, 0.635-0.748 mm long. Intestinal caecum, 1.09-1.14 mm long. Tail conical with rounded end, 0.100-0.125 mm long.

Species: *Anisakis* sp. (Fig. 4 C-D)

Twenty-nine specimens at larval stage were found encysted in the body cavity and stomach wall of *E. granulosus*, and only one larval specimen was found in the liver of *S. bivius*.

Five specimens of *E. granulosus* were measured. Body length 16.35-26.44 mm × 0.354-0.579 mm wide. Nerve ring at 0.170-0.180 mm from the anterior edge. Muscular esophagus, 1.66-2.25 mm long. Ventricle, 0.611-0.718 mm long. Tail conical, 0.104-0.114 mm long.

Undetermined Anisakids

Five specimens were found in the stomach wall and muscles of *E. granulosus*. These nematodes were dry and rigid, and it was impossible to identify the genus; however, according to some features of the tail or cephalic zone, they were determined as Anisakidae gen. sp., possibly correspond to the taxa here recorded.

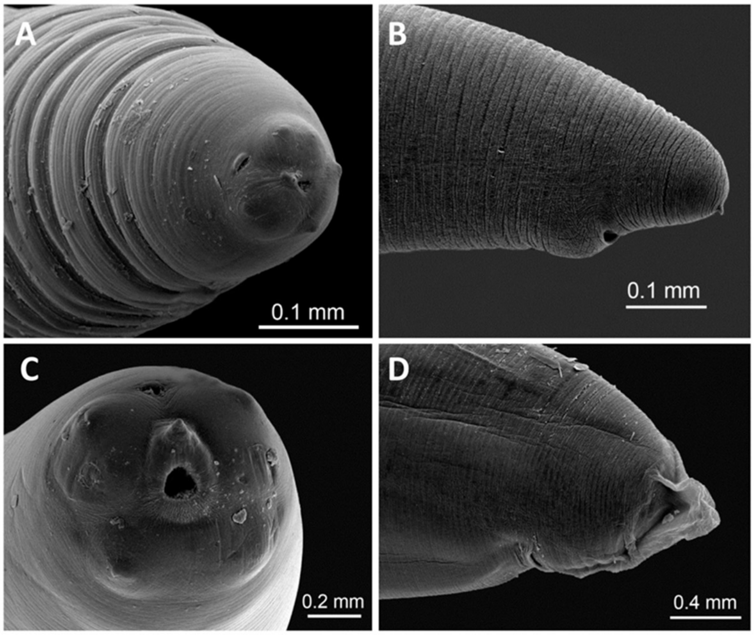

Fig. 4. Larval nematodes of Anisakidae: A) SEM of cephalic apical view and B) tail of *Pseudoterranova* sp.; C) SEM of cephalic apical view and D) tail of *Anisakis* sp.

Remarks: Anisakid nematodes are distributed worldwide and parasitize many kinds of marine animals. In fish, *Anisakis* are found at larval stages, with only a few morphological distinctions between species, making a precise identification difficult. In Chile, two larvae of *Anisakis* morphotypes have been recorded: *Anisakis simplex* sensu Davey, 1971 and *Anisakis physeteris* (Baylis, 1923). Also, most *Pseudoterranova* larvae have been identified as *P. decipiens* (Muñoz & Olmos, 2008). The identification of anisakids, implemented with a molecular approach, has revealed that species identified as "*Anisakis simplex*" and *A. physeteris* are each a species complex (Mattiucci & Nascetti, 2006; Mattiucci *et al.* 1997, 2014; McClelland, 2002). Moreover, the morphometric data of *Anisakis simplex* and *Pseudoterranova decipiens* (Krabbe, 1878) given by Hurst (1984) do not agree with the specimens of the present study. Therefore, identifications based on larval morphologies are here undetermined.

Parasitological descriptors

Pseudoterranova sp. was the most abundant parasite in *E. granulosus*, whereas *Tautochondria* sp. and *Proleptus* sp. were the most prevalent and abundant parasites in *S. bivius* (Table 1). Out of all the parasite taxa found in both shark species, only the two anisakid species, *Pseudoterranova* sp. and *Anisakis* sp., were shared by both the hosts, which correspond to 14.6% of similitude between all the parasite species found in the sharks. However, both anisakid species were more abundant and prevalent in *E. granulosus* than in *S. bivius* (Table 1).

The sample size of *E. granulosus* was sufficient to know the parasite diversity because there were little differences between the observed parasite richness (10 species) and those estimated by Chao 2, Jacknife 1 and Jacknife 2 (11, 13 and 12, respectively) (Fig. 5). However, the sample size of *S. bivius* was relatively small, because the observed richness was concordant with Chao 2 estimates, but differed in four and six species with Jacknife 1 and Jacknife 2, respectively (Fig. 5). Therefore, the correlation analyses were applied only for *E. granulosus*. Richness and abundances of parasite infracommunities were directly correlated in this shark species (n= 24, r_s= 0.48; $P < 0.02$), although, infracommunity richness of parasites did not correlate with host body length of *E. granulosus* (r_s= -0.15; $P > 0.20$) and neither did the parasitic abundance with the host body length (r_s= -0.12; $P > 0.50$).

DISCUSSION

In the present study, several parasite species were recorded in the limited sample sizes of *E. granulosus* and *S. bivius*. Copepods, cestodes, and nematodes were the most represented parasite groups in both shark species. Only a few species of digeneans and monogeneans were present, whereas acanthocephalans, isopods, and hirudineans were absent. Ten parasite taxa were found in 25 specimens of *E. granulosus*, but Espínola-Novelo *et al.* (2018) found 14 parasite species in 133 specimens of *E. granulosus*, which may be due to a larger sample size. For *S. bivius* several parasite species were found in just seven specimens. Considering that only two or three parasite species from the whole parasite community of both sharks were prevalent and abundant, it is possible that parasite communities were well represented in number of species for *E. granulosus* and *S. bivius*. However, it's likely that more parasites exist in the shark species collected from the Chilean austral zone.

E. granulosus from the Chilean austral zone had low parasite abundance, and prevalence below 50%. Similarly, low abundance and prevalence of parasites of the *Etmopterus* genus have been found in studies made in other localities (Dallarés *et al.* 2017; Espínola-Novelo *et al.* 2018; Isbert *et al.* 2015). The low parasitic abundance of *E. granulosus* may be determined by many factors, such as the physic-chemical characteristics of water affecting free stages of parasites, predation rates, or low abundance and prevalence of parasites in intermediate hosts as has been shown using different approaches to the population dynamics of parasites (Esch & Fernández, 1993). However, there are no direct evidence of parasite dynamic infection in *E. granulosus*.

Besides the fact that a small sample size of *S. bivius* was analysed in this study, a greater average of species richness and abundance of parasites was found compared to *E. granulosus*. There is no record of the parasite communities in the *Schroederichthys* species; however, there is evidence of the abundances of nematodes, *Proleptus niedmanni* (=*Proleptus acutus,* according to Fernández & Villalba, 1985), in the redspotted catshark *Schroederichthys chilensis*,

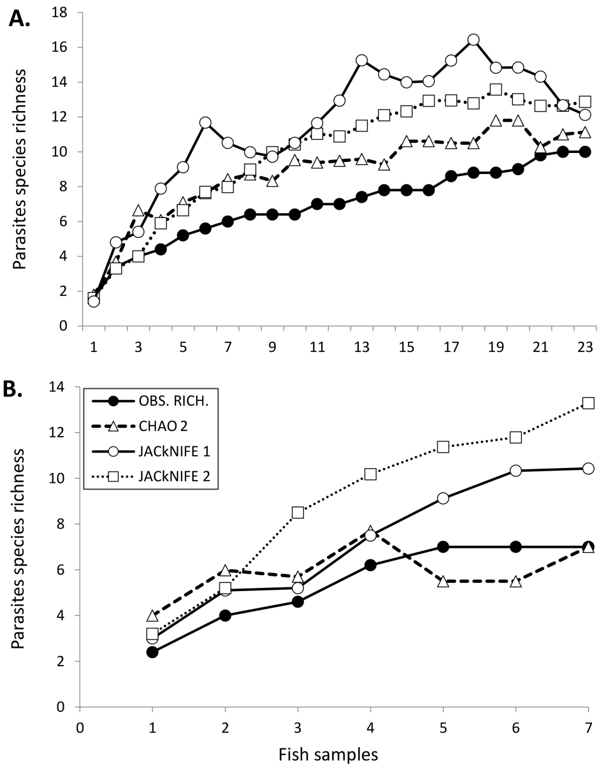

Fig. 5. Three estimates of parasite richness (Chao 2, Jacknife 1, Jacknife 2) and observed richness (Obs. Rich) over the host sample size for A) *Etmopterus granulosus*, and B) *Schroederichthys bivius*. Fig. 5.

with 14 nematodes/fish (George-Nascimento & Vergara, 1982). This nematode is transmitted by predation from Chilean crab *Cancer plebejus* to the redspotted catshark, *S. chilensis* (see George-Nascimento *et al.* 1994). *Schroederichthys bivius* is an opportunistic predator with a wide food spectrum, including polychaetes, anemones, cephalopods, and a wide diversity of fish and crustaceans (Reyes & Hüne, 2012). This shark has a switch in prey preference around 64 cm body length, consuming mostly decapod crustaceans. In the present study, *S. bivius* was smaller than 57 cm of body length, however, there may also be increase of parasite abundance associated with crab intake, similar to *S. chilensis* (George-Nascimento & Vergara, 1982).

Parasite-host specificity has a great influence on the composition and structure of parasite communities, thus host species with specific parasites would have great difference in the composition of parasites (Salgado-Maldonado *et al.* 2016). There are no studies focused on host-specificity in sharks; however, in the checklists of parasites (Muñoz & Olmos, 2007, 2008) and recent records of parasites in sharks from Chile (Espínola-Novelo *et al.* 2018) just a few species are shared among sharks. Thus, a copepod (*Neoalbionella* sp.) and a monogenean (*Calicotyle* sp.) have been previously recorded only in the *E. granulosus* from central and northern Chile (Espínola-Novelo *et al.* 2018; Rodríguez *et al.* 2010). Meanwhile, copepods (*Tautochondria* sp. and *L. bivia*) and nematodes (*Proleptus niedmanni* and *Heliconema* sp.) have been found only in *S. bivius* so far, whereas *P. niedmanni* was in the congeneric shark species, *S. chilensis* from the central Chile (George-Nascimento & Vergara, 1982; Torres & Grandjean, 1983). Therefore, more closely related host species (genus or family level) tend to have similar parasites (Poulin, 1998). Besides the fact that *E. granulosus* and *S. bivius* live in sympatry, they share only few parasites, thus their parasitological differences can be due to the distance of host taxonomic relatedness. Similar results were found in two sympatric sharks from northern Chile (Espínola-Novelo *et al.* 2018); *E. granulosus* (Squaliformes: Etmopteridae) and *Apristurus nasutus* (Carcharhiniformes: Scyliorhinidae) that altogether have 16 parasite taxa but shared only two species. Then again, the low similitude of parasite community composition may be due to low taxonomic relatedness between the sharks beside they live in the same habitat. In conclusion, *E. granulosus* and *S. bivius* have different evolutionary histories with several differences in their ecological characteristics, having as a result different parasite lineage and species composition.

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