

First record of parasites of *Dosidicus gigas* (d'Orbigny, 1835) (Cephalopoda: Ommastrephidae) from the Humboldt Current system off Chile

Primer registro de los parásitos de *Dosidicus gigas* (d'Orbigny, 1835) (Cephalopoda: Ommastrephidae)
en el sistema de corriente de Humboldt frente a Chile

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Resumen.- Se describió y cuantificó la fauna parasitaria de 124 calamares (*Dosidicus gigas*) capturados en aguas oceánicas y costeras en Chile Central (32°S - 40°S). El 97% de los calamares presentó al menos un taxón parásito. Se determinó la prevalencia y abundancia para cada especie de parásito. Cada taxón parasitario se describió en base a microscopía de luz e imágenes de microscopía electrónica de barrido. La fauna parasitaria estuvo compuesta por céstodos larvales: *Hepatoxylon trichiuri* (Abundancia: 2,6; Prevalencia: 70,2%), *Tentacularia coryphaenae* (Abundancia: 0,1; Prevalencia: 5,6%), plerocercos de Tetracyllidae (Abundancia: 9,1; Prevalencia: 83,1%), *Pelichnibothrium speciosum* y los nemátodos *Anisakis* Tipo I (Abundancia: 0,06; Prevalencia: 6,5%) y Tipo II (Abundancia: 0,52; Prevalencia: 17,7%). Se han descrito alrededor de 9 taxa parasitarios en *D. gigas*, todos encontrados en calamares menores a 40 cm de longitud del manto, y de los cuales cinco (plerocercos de Tetracyllidae, *Tentacularia coryphaenae*, *Pelichnibothrium speciosum* y *Anisakis* Tipo I y II), se registraron en este estudio, pero en calamares de mayor tamaño. El parásito *H. trichiuri* es un nuevo registro para *D. gigas* en estas latitudes del Pacífico Sur, comprendiendo el límite sur de su rango de distribución. El plerocercos de Tetracyllidae, por tener un órgano apical con la misma forma y tejido celular que una ventosa accesoria, podría pertenecer al género *Phyllobothrium*. Los *Anisakis* Tipo I y II, sólo se pueden distinguir por la forma de la cola y la presencia-ausencia del mucrón. Se sugiere la realización de análisis moleculares cuando hay carencia de caracteres morfológicos de diagnósticos en los estados larvales.

Palabras clave: Calamar de Humboldt, fauna parasitaria, Pacífico suroriental

Abstract.- The parasitic fauna of 124 jumbo squids (*Dosidicus gigas*) caught from oceanic and coastal waters off Central Chile (32°S-40°S) was described and quantified. The 97% of the squids harboured at least one parasitic taxon. Prevalence of infection and abundance were established for each parasite species. Each parasitic taxon was described based on light microscopy and scanning electron microscopy images. The parasitic fauna was composed of larval cestodes: *Hepatoxylon trichiuri* (Abundance: 2.6; Prevalence: 70.2%), *Tentacularia coryphaenae* (Abundance: 0.1; Prevalence: 5.6%), a Tetracyllidean plerocercoid (Abundance: 9.1; Prevalence: 83.1%), *Pelichnibothrium speciosum* and the nematodes *Anisakis* Type I (Abundance: 0.06; Prevalence: 6.5%) and *Anisakis* Type II (Abundance: 0.52; Prevalence: 17.7%). Around nine taxa have been reported in *D. gigas* smaller than 40 cm mantle length, five of them were found in this study (Tetracyllidean plerocercoid, *Tentacularia coryphaenae*, *Pelichnibothrium speciosum*, and *Anisakis* Type I and II), however, in bigger squids. The parasite *H. trichiuri* is a new record for *D. gigas* in these latitudes of the South Pacific, comprising the southern limit of its distribution range. The Tetracyllidean plerocercoid could correspond to *Phyllobothrium* based on the shape and tissues of its apical organ, which is very similar to the accessory suckers. The nematodes *Anisakis* Type I and II can only be distinguished by the tail shape and presence-absence of a mucron. It is suggested to carry out molecular analysis for diagnosis of larval stages when morphological characters are lacking.

Key words: Jumbo squid, helminth fauna, southeastern Pacific

Introduction

The jumbo squid, *Dosidicus gigas* (d'Orbigny, 1835), is the most abundant squid of the Pacific Ocean (Nigmatullin *et al.* 2001, Keyl *et al.* 2008), and the largest marine invertebrate of the Chilean fauna (Schmiede & Acuña 1992, Fernández & Vásquez 1995). The geographic distribution of *D. gigas* extends from approximately 40°N to 42°S in the eastern Pacific Ocean, and lives both in oceanic as well as neritic water, from surface waters down to depths of 1200 m (Nigmatullin *et al.* 2001, Keyl *et al.* 2008). For *D. gigas*, three groups (small, medium, large) have been distinguished based on the size at maturity (Nigmatullin *et al.* 2001). The helminth fauna of medium sized *D. gigas* (300-431 mm mantle length) from Nicaraguan, Equatorial and Peruvian waters (11°N-22°S), has been reported to be composed of larval stages of Didymozoidae, the cestoda *Pelichnibothrium speciosum*, *Phyllobothrium* sp., *Tentacularia coryphaenae*, and the nematodes *Anisakis simplex*, *A. physeteris*, *Porrocaecum* sp., *Contracaecum* sp., and *Spinitectus* sp. (Shukhgalter & Nigmatullin 2001). However, Nigmatullin *et al.* (2005¹) also studied helminth fauna in five specimens of large mantle length (545-995 mm) *D. gigas* (area 7-9°S and 83°30'-84°W), finding four helminth species: *Pelichnibothrium speciosum*, *Tentacularia coryphaenae*, *Anisakis physeteris*, *Porrocaecum* sp. (Nigmatullin *et al.* 2005¹). Towards the South Pacific, only *Bothriotaenia* sp. has been registered for *D. gigas* from Chile (36°35'S, Talcahuano) by Wilhelm (1954); there is no recent record of this parasite in these latitudes. This squid is considered as a paratenic host for the larval parasites that infect it, and the definitive hosts are fish, sharks and marine mammals (Shukhgalter & Nigmatullin 2001).

The existence of unstable anatomical and meristic characters (*e.g.* in coccidians), cryptic species (*e.g.* in anisakid nematodes) or morphotypes (*e.g.* in cestodes) present particular difficulties for the identification because of the plasticity of body structures in most of the 200 endoparasitic species reported in cephalopods (Hochberg 1990, Pascual *et al.* 2007). In fact, most of these parasites have not been taxonomically identified to species level, with high rates of synonymy (close to 70%) for numerous nominal parasite species or larval stages identified by light microscopy (Pascual *et al.* 2007).

The use of scanning electron microscopy (SEM) has allowed the study of morphoanatomic and morphometric characteristics of the surface structures of many parasitic taxa, including some larvae of nematode and cestode species from cephalopods (Abollo *et al.* 1998, Gestal *et al.* 1998a, b, Pascual *et al.* 1999).

The aim of this study was to report a quantitative characterization and to describe the morphology of the helminth fauna of *D. gigas* from oceanic and coastal waters off central-southern Chile. Moreover, the results in this preliminary study of parasites in *D. gigas* from central and southern Pacific Ocean waters provided an opportunity to compare its helminth fauna with those previously reported from other latitudes in the Pacific Ocean. The differences in the helminth fauna may indicate a potential use for stock identification.

Material and methods

From July 2003 to February 2004, 51 samples of *D. gigas* were collected from the by-catch obtained by purse-seine fishing for jack mackerel, *Trachurus murphyi* in oceanic waters off central-southern Chile (32°S-40°S) at depths between 10 and 70 m. In addition, 73 samples were obtained from trawlers fishing for Chilean hake, *Merluccius gayi*, and Patagonian grenadier, *Macruronus magellanicus*, on the continental shelf of central-southern Chile (34°S-40°S) near bottom at depths between 120 and 405 m (see Ibáñez *et al.* 2008, Fig. 1). The mantle length (ML, cm) of each squid was measured and was 61.1 cm in average (range = 30-92 cm, SD = 16.5 cm). The mantle cavity, muscle fibres, digestive tract, gonads, gills, digestive gland and other viscera were examined under a stereoscopic microscope, searching for endoparasites.

The methodology used for obtaining the parasites followed Pardo-Gandarillas *et al.* (2004). The nomenclature of Nematoda follows Yamaguti (1961), Hochberg (1990) and Shukhgalter & Nigmatullin (2001), and for Cestoda it follows Williams (1968), Stunkard (1977), Hochberg (1990), Schmidt (1986), Scholtz *et al.* (1998), Shukhgalter & Nigmatullin (2001) and Palm (2004). Demographic features were expressed in terms of prevalence and abundance (total number of parasitic individuals for a parasitic taxon / total number examined hosts; Margolis *et al.* 1982).

All parasites obtained were fixed in 10% formalin. Some nematodes were cleared in a mixture of glycerine, lactic acid, phenol and distilled water (lactophenol). Morphometric measures of 10-20 individuals of each parasite species were made, following Shukhgalter & Nigmatullin (2001) and Palm (2004). The internal

¹Nigmatullin CM, AS Shchetinnikov & OA Shukhgalter. 2005. Food and parasite relations and role of the dominant ommastrephid squids in oceanic ecosystems of southeast Pacific in 1980-1985. Third International Symposium on Pacific squids. Lima, Peru, 28-30 November, 2005.

anatomy of each parasite species was drawn under a stereoscopic or light microscope. This material was deposited in the collection of molluscs parasites of Museo Nacional de Historia Natural de Chile (MNHNCL).

For SEM analyses the parasites were fixed in either 10% formalin in seawater (SW) or 2% glutaraldehyde in SW with 0.025M sodium cacodylate (Turner & Boyle 1975). They were dehydrated in an ethanol series, critical point dried, and ion sputtered with gold.

Additionally, six large squids (80-88 cm ML) Coquimbo (29°53'S, 71°36'W) were examined in December 2008. In these specimens histology of the rectum was performed for cysts with merocercoids to be observed. Plerocercoids of Tetraphyllidean larvae were collected from the caecum, and along with small pieces of rectum, fixed in Davidson's fluid for 24 h, transferred to 70% ethanol, and further processed for histology (Shaw & Battle 1957). Five micrometer thick sections were cut from the rectum pieces, and from the plerocercoids longitudinal and transverse 6 µm thick sections of the scolex were cut. Photographs were taken from a Zeiss Axiostar Plus microscope with a Cannon Powershot A 620 camera.

Results

Of the 124 specimens of *D. gigas* examined, 96.7% were infected with at least one of the six taxa identified in this study.

Cestoda

Order: Trypanorhyncha Diesing, 1863

Family: Sphyricephalidae Pintner, 1913

Genus: *Hepatoxylon* Bosc, 1811

Hepatoxylon trichiuri (Holten, 1802): (Abundance: 2.6; Prevalence: 70.2%) (Figs. 1a, 2)

Description: Measurements based on 20 specimens. Size range 13-27 mm. Scolex large and muscular (length = 4-8 mm, maximum width = 4-5 mm, Fig. 1a). Two bothria large oval to elongated (length = 2-4 mm) separated by narrow partition (Figs. 2a,b). Four tentacles short, globular or truncated (length = 0.6-0.78 mm, maximum width = 0.57-0.71 mm) (Fig. 2a). Metabasal armature homeoacanthous homeomorphous; hooks in 12-14 rows slender uncinata with anterior extension of the base

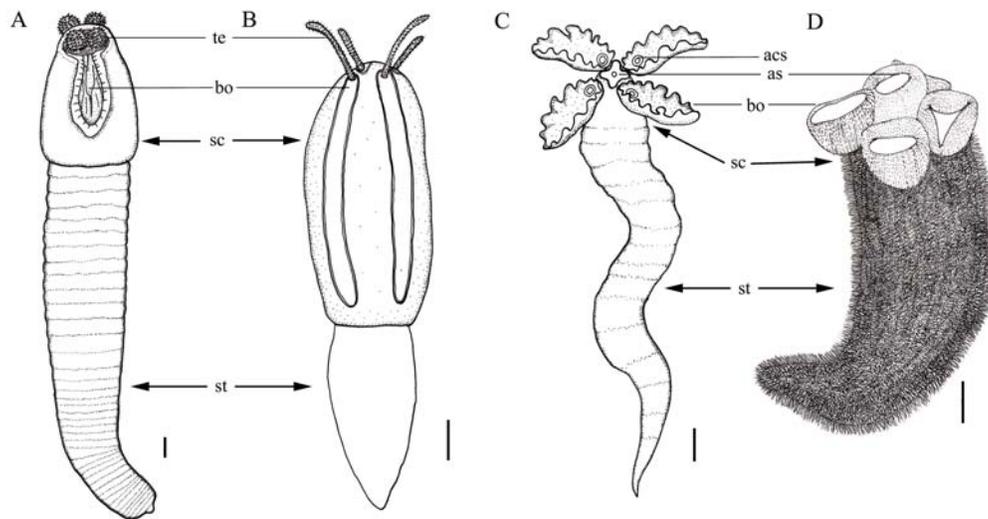


Figure 1

Cestode parasites of *D. gigas*. A) *Hepatoxylon trichiuri*, B) *Tentacularia coryphaenae*, C) Tetraphyllidea plerocercoid D) *Pelichnibothrium speciosum*. Scale bar 1mm, except D 0.1 mm. sc = scolex, bo = bothria, te = tentacle, st = strobila, as = apical sucker, acs = accessory sucker

Parásitos céstodos de *D. gigas*. A) *Hepatoxylon trichiuri*, B) *Tentacularia coryphaenae*, C) Plerocercoides Tetraphyllidea D) *Pelichnibothrium speciosum*. Barra 1 mm, excepto D 0,1 mm. sc = scolex, bo = botria, te = tentáculo, st = estróbila, as = ventosa apical, acs = ventosa accesoria

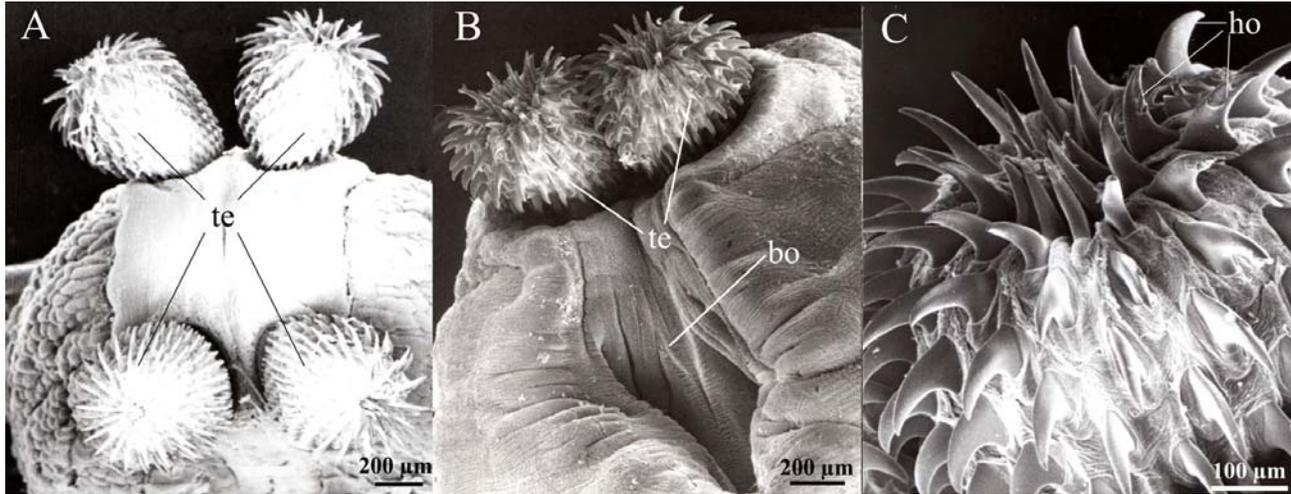


Figure 2

Hepatoxylon trichiuri from *D. gigas*. Scanning electron microscopy (SEM) of: **A) tentacles, B) tentacles and bothria, C) tentacle armature. bo = bothria, te = tentacle, ho = hook**

Hepatoxylon trichiuri de *D. gigas*. Microscopía electrónica de barrido (MEB) de: **A) tentáculos, B) tentáculos y botria, C) armadura tentacular. bo = botria, te = tentáculo, ho = gancho**

(length = 0.09-0.12 mm, basal = 0.06-0.08 mm) (Fig. 2c). Hosts: Adults parasites of elasmobranchs (Schmidt 1986, George-Nascimento 1987, Palm 2004). Plerocercoids in a wide variety of fishes in Chile (George-Nascimento 1987). Distribution: Cosmopolitan (Schmidt 1986). Site: Mantle cavity, gonads and stomach. Deposition number of material: 10 specimens, MNHNCL 100001.

Remarks: The shape of the hooks (uncinate) and the tentacles armature are identical to specimens described by Palm (2004). Moreover, tentacles had 12-14 hook rows similar to those (12-15) described by Palm (2004).

Family: Tentaculariidae Poche, 1926

Tentacularia coryphaenae Bosc, 1797: (Abundance: 0.1; Prevalence: 5.6 %) (Figs. 1b, 3)

Description: Measurements based on 20 specimens. Size range 8-15 mm. Scolex elongated and craspedote (length = 5-10 mm, maximum width = 1-3 mm), (Fig. 1b). Four tentacles short, slender, armature homeoacanthous homeomorphous with uncinat hooks (Figs. 3a,b) (length = 0.030-0.033 mm, basal = 0.020-0.026 mm) and tridentate basal hooks (length = 0.015-0.026 mm, basal = 0.008-0.010 mm) (Fig. 3c). Four large and narrow bothria separated (length = 4.5-9 mm), with hook-like microtriches along the bothrial margins present (Fig. 2d). Hosts: Larvae parasites of marine fishes and cephalopods (George-

Nascimento 1987, Hochberg 1990, Palm 2004). Adult parasites of elasmobranchs (George-Nascimento 1987, Hochberg 1990, Palm 2004). Distribution: Cosmopolitan. Site: Mantle cavity and gonads. Deposition number of material: 10 specimens, MNHNCL 100002.

Remarks: The body measurements of *T. coryphaenae* were similar to specimens described by Shukhgalter & Nigmatullin (2001). The length and basal lengths of metabasal and basal hooks were within the range of specimens measured by Shukhgalter & Nigmatullin (2001). The shape of hook and tentacle armature was identical to specimens described by Palm (2000, 2004).

Order: Tetracystida Carus, 1863

Plerocercoid larvae Tetracystida: (Abundance: 9.1; Prevalence: 83.1%) (Figs. 1c, 4, 5)

Description: Measurements based on 20 specimens. Size range 6-19 mm. Scolex short (length = 1-3 mm, maximum width = 1-4 mm), with apical sucker (Figs. 1c, 4a,b,c, 5a). Four bothria large and sessile (length = 0.5-2 mm) (Fig. 1c), with margins folded and curled (Figs. 4a). One accessory sucker on each bothria (Fig. 1c, 4b). The apical region of the scolex has a dome-shaped structure, and its internal tissue does not differ histologically from the rest of the scolex, except for the tissue surrounding the tip of similar to an accessory sucker composed by fibers of radial muscle and delimited by a thin muscular membrane (Fig.

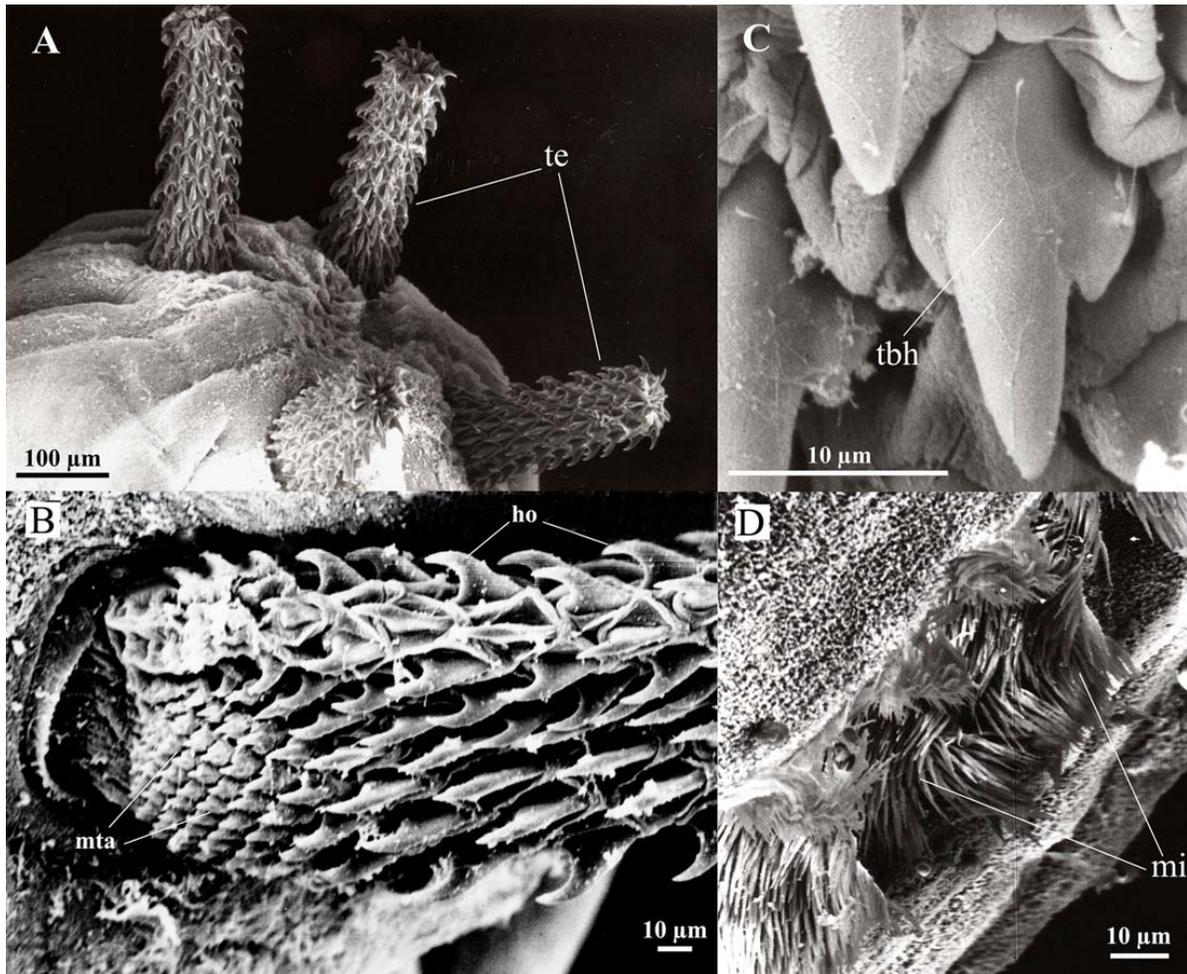


Figure 3

Tentacularia coryphaenae from *D. gigas*. SEM of: A) tentacles and bothria, B) tentacular armature, C) basal tentacular armature showing tridigitate basal hooks, D) hook-like microtriches along the bothrial margins. te = tentacle, mi = microtriches, mta = metabasal tentacle armature, tbh = tridentated basal hook

Tentacularia coryphaenae de *D. gigas*. MEB de: A) tentáculos y botrias, B) armadura tentacular, C) armadura de la base del tentáculo mostrando ganchos basales tridigitados, D) Microtrichos en forma de ganchos a lo largo del margen botrial. te = tentáculo, mi = microtrichos, mta = armadura tentacular metabasal, tbh = gancho basal tridentado

5b). Hosts: Adults parasites of elasmobranches (Williams 1968, Hochberg 1990). Distribution: Pacific, Atlantic and Indian Oceans. Site: Stomach, caecum and intestine. Deposition number of material: 10 specimens, MNHNCL 100003.

Remarks: the presence of an apical sucker-like structure, accessory sucker on each bothria and the bothrial shape (folded and curled) resembles *Phyllobothrium timidum* Linton, 1922 described by Stunkard (1977). Morphometric measurements of larvae are similar to specimens described by Stunkard (1977) and Shukhgalter & Nigmatullin (2001),

what makes us believe this is a related species. However, molecular analysis suggests that previous reports of plerocercoids of *Phyllobothrium* spp. in squid may have been in error (Brickle *et al.* 2001).

Family: Phyllobothriidae Braun, 1900

Pelichnibothrium speciosum Monticelli, 1889 (Figs. 1d, 6)

Description: Measurements based on 10 cysts and 10 specimens. Size of cysts was 0.57-0.19 × 0.10-0.14 mm. Larvae of small size (length = 0.12-0.15 mm width = 0.057-



Figure 4

Tetraphyllidea plerocercoid from *D. gigas*. SEM of: A) bothria and apical sucker, B) bothrial folds, C) detail of apical sucker. bo = bothria, as = apical sucker, acs = accessory sucker

Plerocercoides Tetraphyllidea de *D. gigas*. MEB de: A) botria y ventosa apical, B) pliegues botriales C) detalle de ventosa apical. bo = botria, as = ventosa apical, acs = ventosa accesoria

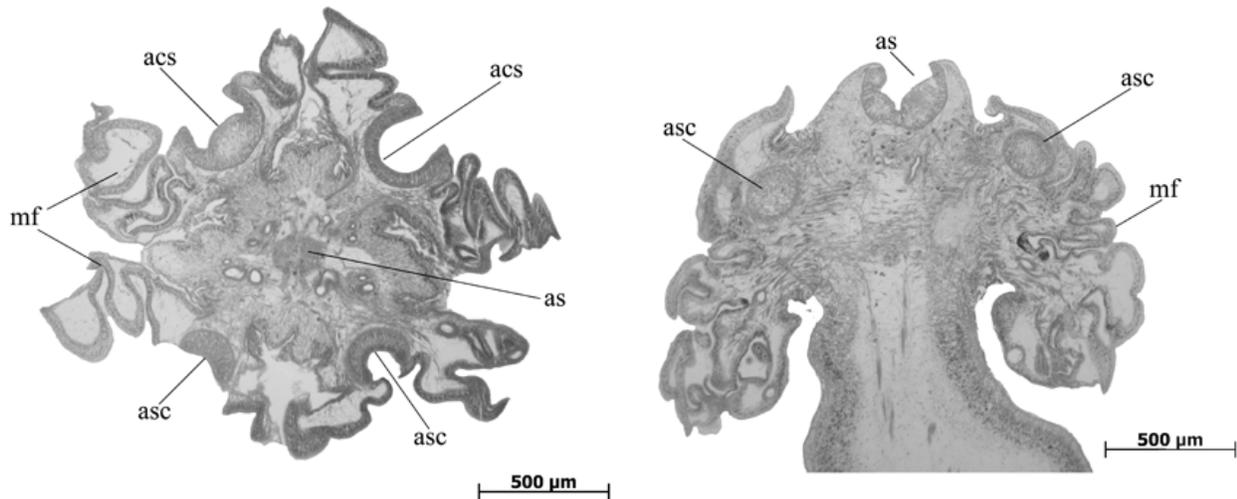


Figure 5

Histological sections of a Tetraphyllidea plerocercoid from *D. gigas*. A) Transverse section, B) Detail of apical region in longitudinal section. as = apical sucker, acs = accessory sucker, mf = margin fold. Stain: haematoxylin and eosin (H & E)

Corte histológico de plerocercoides Tetraphyllidea de *D. gigas*. A) Corte transversal, B) Detalle de la región apical en corte longitudinal. as = ventosa apical, acs = ventosa accesoria, mf = pliegue marginal. Tinción: hematoxilina y eosina (H & E)

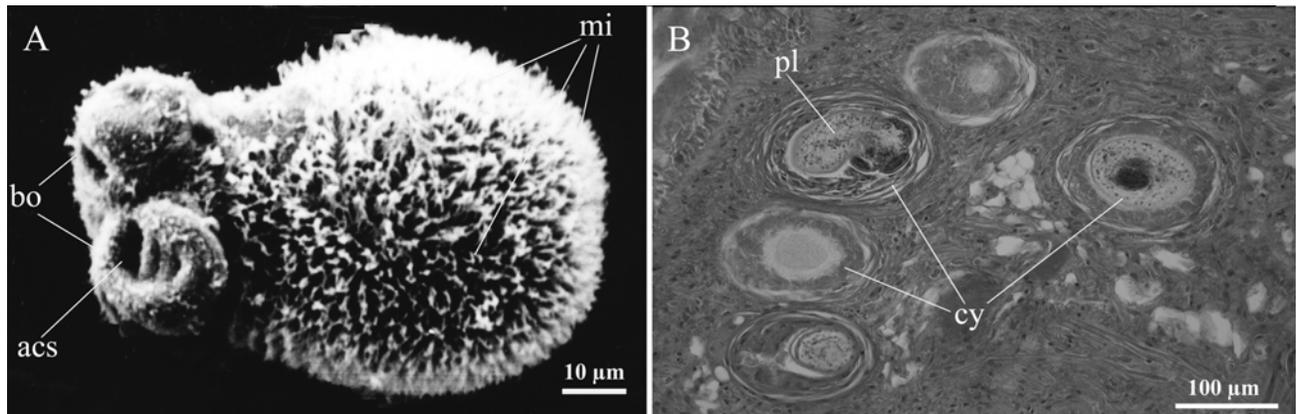


Figure 6

Pelichnibothrium speciosum from *D. gigas*. SEM of: A) larva, B) Histological section of rectum tissue of *D. gigas* showing cysts. Stain: haematoxylin and eosin (H & E). bo = bothria, acs = accessory sucker, mi = microtriches, pl = plerocercoid, cy = cysts

Pelichnibothrium speciosum de *D. gigas*. MEB de: A) larva, B) preparación histológica de tejido del recto de *D. gigas* mostrando los quistes. Tinción: hematoxilina y eosina (H & E). bo = botria, acs = ventosa accesoria, mi = microtricos, pl = plerocercoide, cy = quistes

0.085 mm. Scolex short not clearly differentiated, with apical sucker (diameter = 0.028 – 0.040 mm) (Fig. 1d). Four bothria sessile with free posterior ends (length = 0.037–0.040 mm) (Fig. 6a), with an accessory sucker (Fig. 6a). Strobila short and covered with microtriches (Fig. 1d, 6a). In the same host individual different stages of development of this parasite species were found (Fig. 6b). Hosts: Larvae parasites of marine fishes and squids, adult parasites of *Prionace glauca* (Hochberg 1990, Scholtz *et al.* 1998). Distribution: Pacific, Atlantic and Indian Oceans. Site: intestine. Deposition number of material: rectum tissue with cysts, MNHNCL 100004.

Remarks: the presence of an apical sucker, an accessory sucker on each bothria and the body covered with microtriches resembles *Pelichnibothrium speciosum* described by Scholtz *et al.* (1998).

Nematoda

Family: Anisakidae Skrjabin & Karokhin, 1945

Anisakis Type II: (Abundance: 0.52; Prevalence: 17.7%) (Figs. 7a, 8a,b).

Description: Measurements based on 10 specimens. Size range 27–33 mm, maximum body widths 0.63–0.74 mm. Length from anterior end of body to nerve ring 0.35–0.43 mm. Oesophagus length 1.82–2.89 mm, maximum width 0.16–0.30 mm. Size of ventriculus 0.53–0.65 × 0.33–0.55 mm. Postanal tail round and without mucron or spine

(length 0.17–0.32 mm) (Figs. 7a, 8b). Body surface totally striated (Fig. 7a). Mouth triangular anteroventrally (Fig. 8a). Boring tooth located ventral to the mouth. Excretory pore transverse slit situated between the ventrolateral lips (Fig. 8a). Postanal tail round, without terminal mucron. Hosts: Larvae parasites of marine fishes and cephalopods (George-Nascimento 1987, Hochberg 1990). Adult parasites of marine mammals (Hochberg 1990). Distribution: Cosmopolitan. Site: Gonads, stomach, caecum and intestine. Deposition number of material: 10 specimens, MNHNCL 100005.

Remarks: The ventriculus is relatively shorter and wider than in *Anisakis* Type I, with a ventriculus-intestinal junction characterized by shape slightly bilobed. The body measurement of *Anisakis* Type II was very similar to specimens of *A. physeteris* described by Shukhgalter & Nigmatullin (2001). The absence of a tail mucron was the principal character that differentiated it from *Anisakis* Type I.

Anisakis Type I: (Abundance: 0.06; Prevalence: 6.5%) (Fig. 7b, 8c,d)

Description: Measurements based on 10 specimens. Size range 25–35 mm, maximum body widths 0.62–0.75 mm. Length from anterior end of body to nerve ring 0.79–0.86 mm. Oesophagus length 1.79–2.74 mm, maximum width 0.18–0.25 mm. Size of ventriculus 0.65–0.79 × 0.21–0.39 mm. Rounded tail (length = 0.20–0.30 mm) with a mucron (length

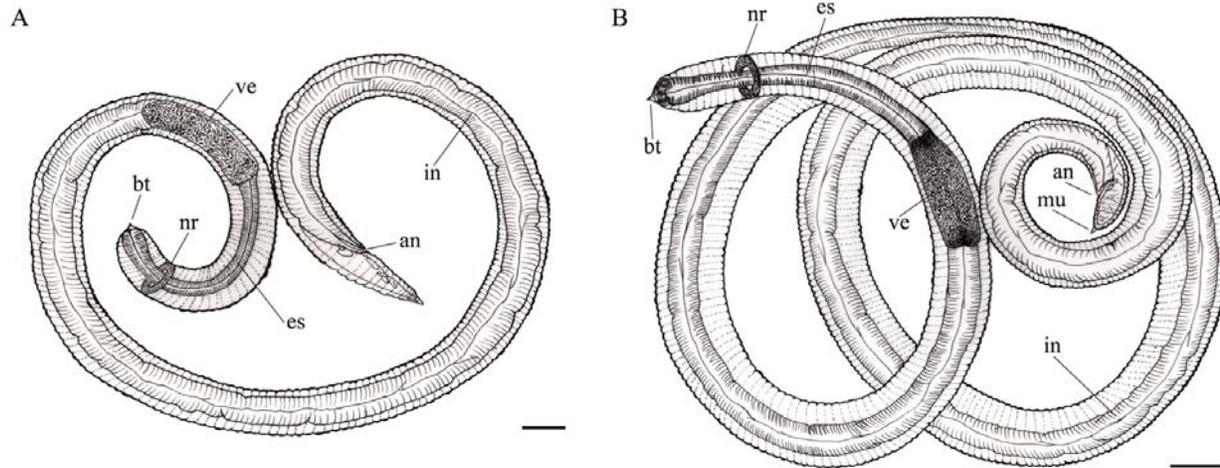


Figure 7

Nematode parasites of *D. gigas*. A) *Anisakis* Type II, scale bar = 0.7 mm. B) *Anisakis* Type I, scale bar = 0.6 mm. bt = boring tooth, nr = nerve ring, es = esophagus, ve = ventriculus, in = intestine, an = anus, mu = mucron

Nemátodos parásitos de *D. gigas*. A) *Anisakis* Tipo II, barra = 0,7 mm. B) *Anisakis* Tipo I, barra = 0,6 mm. bt = diente perforador, nr = anillo nervioso, es = esófago, ve = ventrículo, in = intestino, an = ano, mu = mucrón

= 0.02-0.05 mm) (Fig. 7b, 8d). Body surface totally striated (Fig. 7b). Mouth rounded with dorsal and ventrolateral lips with papilla (Fig. 8c). Tooth located ventral to the mouth projecting anteroventrally. Excretory pore transverse slit situated between the ventrolateral lips, opens from a single excretory duct. Rectum opens at anus, with rectal glands. Postanal tail round, with a terminal mucron or spine. Hosts: Larvae parasites of marine fishes and cephalopods (George-Nascimento 1987, Hochberg 1990). Adult parasites of marine mammals (Hochberg 1990, Abollo *et al.* 1998). Distribution: Cosmopolitan. Site: Gonads, stomach, caecum and intestine. Deposition number of material: 1 specimen, MNHNCL 100006.

Remarks: The ventriculus is rather long and narrow with respect to *Anisakis* Type II, with a rounded posterior margin. The body measurements of *Anisakis* Type I was very similar to specimens of *A. simplex* described by Shukhgalter & Nigmatullin (2001). The presence of a tail mucron was the principal character that differentiated it from *Anisakis* Type II.

Discussion

The present study is the first record of *Dosidicus gigas* parasites from south-central Chilean waters. We found six parasitic taxa all represented by larval stages, being *Hepatoxylon trichiuri* the first record for this host. Around nine taxa have been reported in *D. gigas* individuals

smaller than 40 cm of ML (Shukhgalter & Nigmatullin 2001), and five of them were found in this study (*Pelichnibothrium speciosum*, plerocercoids Tetrathyllidea, *Tentacularia coryphaenae* and *Anisakis* Type I and II), but in squids of larger body size. The parasite composition of *D. gigas* is very similar to those described for other ommastrephid squids (Naidenova *et al.* 1985, Bower *et al.* 1990, Bower & Margolis 1991, Nigmatullin & Shukhgalter 2001, Nigmatullin *et al.* 2005¹) indicating a closeness in terms of ecological niches (Shukhgalter & Nigmatullin 2001).

The genus *Hepatoxylon* is well known and includes two species, *H. trichiuri* and *H. megacephalum* (Palm 2004). The genus is distributed worldwide from the tropical to the boreal region, occurring as adults in pelagic sharks, with the plerocercoids infesting teleosts and sharks (Schmidt 1986, Palm 2004). Larval stage of this parasite has been reported in other four squid species, *Architeuthis dux*, *Illex argentinus*, *Sthenoteuthis pteropus* and *Todarodes angolensis*, but in the Atlantic Ocean (Hochberg 1990, Palm 2004). However, it has not been reported in Pacific Ocean squids. Therefore, this would be the first reported of *H. trichiuri* infecting squids in the Pacific Ocean.

The genus *Tentacularia* is well known and comprises only a single species, *T. coryphaenae* (Palm 2004). This genus is distributed worldwide in tropical and subtropical

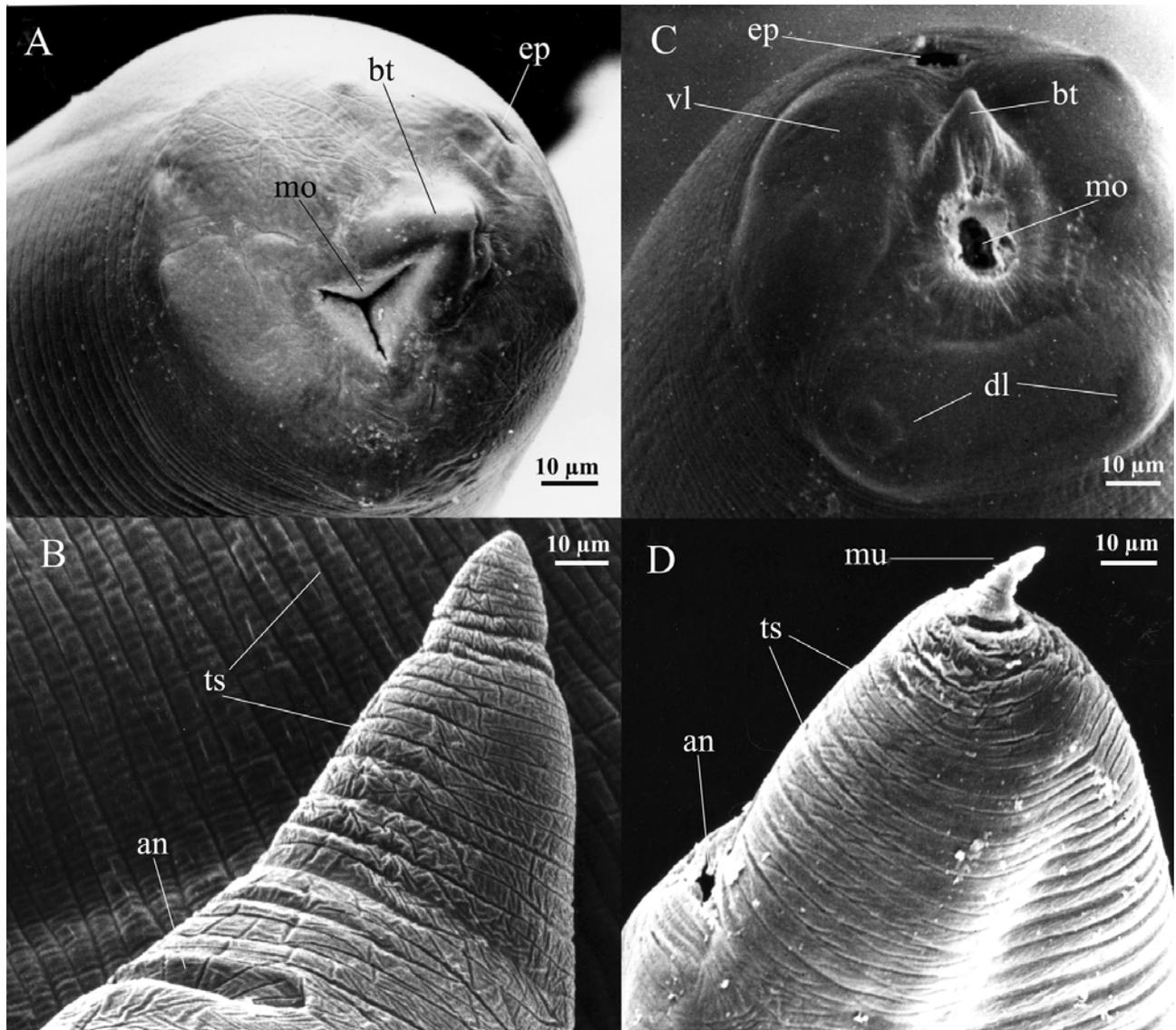


Figure 8

Anisakis from *D. gigas*. SEM of: A) head with mouth triangular and four papilla of *A. Type II*, B) postanal rounded tail of *A. Type II* C) head showing mouth rounded with dorsal and ventrolateral lips with papilla of *A. Type I*, D) postanal rounded tail with terminal mucron of *A. Type I* bt = boring tooth, an = anus, mu = mucron, ep = excretory pore, mo = mouth, vl = ventral lip, dl = dorsal lip, ts = transverse striation

Anisakis de *D. gigas*. MEB de: A) cabeza con boca triangular de *A. Tipo II*, B) cola postanal redondeada de *A. Tipo II*, C) cabeza mostrando la boca redondeada con labios dorsal y ventrolaterales con papilas de *A. Tipo I*, D) cola postanal redondeada, con un mucrón terminal de *A. Tipo I* bt = diente perforador, an = ano, mu = mucrón, ep = poro excretor, mo = boca, vl = labio ventral, dl = labio dorsal, ts = estriación transversal

seas, inhabiting migratory scombrids and larger pelagic predatory fish as second intermediate hosts and carcharhinid sharks as final hosts (George-Nascimento 1987, Hochberg 1990, Palm 2004). Palm (2000) studied *T. coryphaenae* from fish hosts with SEM, recording the tentacular armature and microtriches along the bothrial margins. A similar larval cestode with the same taxonomic characteristics was recorded in this study from *D. gigas*, and was identified as *T. coryphaenae*. This taxon has also been registered from 19 other squid species (Hochberg 1990, Palm 2004).

The Tetraphyllidean plerocercoid was difficult to identify to the genus or species level, since only larval specimens were available, but it is very similar to *Phyllobothrium* genus. Following the approach of Caira *et al.* (1999) in the description of larval cestodes, in stained specimens it is possible to distinguish a structure similar to an apical organ, with a discrete boundary between this structure and the tissue of the scolex proper. Furthermore, the apical organ has the same shape and tissue than the accessory suckers, so it could be a *Phyllobothrium* plerocercoid. However, molecular analysis is necessary to confirm this. These larvae are present in many species of cephalopods and fishes and the adults are found in several sharks and rays (Williams 1968, Hochberg 1990).

Pelichnibothrium speciosum is the only species of the genus (Scholtz *et al.* 1998). Larvae of this species parasitize 12 cephalopod species and some fishes (Hochberg 1990), and adults of *P. speciosum* seem to be specific parasites of *Prionace glauca* (Scholtz *et al.* 1998). The geographical distribution of *P. speciosum* is fairly extensive and probably cosmopolitan, as records include the North Atlantic Ocean, inclusive of the Mediterranean Sea, the Indian Ocean and the Pacific Ocean (Scholtz *et al.* 1998, Shukhgalter & Nigmatullin 2001).

A low prevalence and abundance of *Anisakis* Type I rather than *Anisakis* Type II was found in this study, a similar situation has been reported from Peru (Shukhgalter & Nigmatullin 2001). Larval *Anisakis* species have been differentiated morphologically by the shape and size of the ventriculus and the presence-absence of tail mucron (see Hochberg 1990, Shukhgalter & Nigmatullin 2001). *Anisakis* Type I and II showed differences in the ventriculus size and the ventriculus-intestinal junction were characterized by the shape rounded and slightly bilobed in both *Anisakis* respectively. Furthermore, the shape of the mouth and the tail with the presence-absence of tail mucron too distinguished between *Anisakis* Type I and II. However,

the molecular analysis indicates that the genetic markers are very useful to identify larval stages of *Anisakis* spp., in the cases that the morphological characters are not sufficient at the genus level or species (Mattiucci *et al.* 2005, 2008) or when sibling species exist like the morphotypes Type I that involve at least three biological species (*Anisakis simplex*, *A. pegreffii* and *A. typical*), and the morphotype Type II (*A. physeteris*, *A. brevispiculata* and *A. paggiae*), in the North Atlantic (Mattiucci *et al.* 2008). In fact the molecular analysis of *Anisakis* Type I and II infecting the fishes *Merluccius gayi* and *Hoplostethus atlanticus*, caught off the Chilean coast, correspond to *Anisakis pegreffii* and *A. paggiae*, respectively (Mattiucci *et al.* 2008²). These records make us think that *Anisakis* spp. infecting *D. gigas* from Chile could be one of these species.

Some metazoan parasites have been used to monitor populations of different cephalopod species from the North Pacific, North Atlantic and the Mediterranean Seas (Bower & Margolis 1991, Pascual & Hochberg 1996). *Hepatoxylon trichiuri* appears to be absent from *D. gigas* from Central and Eastern Pacific (Bower *et al.* 1990, Shukhgalter & Nigmatullin 2001). However, this parasite species is common in Chilean waters, suggesting that it could be a good indicator (biological tag) of a relatively low migration of *D. gigas* from Perú to Chile. Keyl *et al.* (2008) suggest that the Nazca Ridge possibly acts as a barrier for smaller *D. gigas*, preventing them from migrating to cooler water masses in the south in years of low abundance. However, the genetic data show a great genetic flow between these areas (Sandoval-Castellanos *et al.* 2007), and the parasite could help to reveal the migration paths in the Humboldt Current System.

Comparing with literature, differences in composition of parasitic fauna can be the result of geographic variations in relation to trophic habits. Nigmatullin *et al.* (2001, 2005¹) suggest that these parasites are transmitted by planktonic crustaceans and small fishes like myctophids from the diet of Ommastrephid squids. In Chile *D. gigas* feed on myctophids, predatory fishes, euphausiids and cephalopods (Ibáñez *et al.* 2008), which are responsible for the parasitic fauna in these latitudes.

The occurrence of parasites in many species of cephalopods is common, and these molluscs play an

²Mattiucci S, M Paoletti, S Damiano, SC Webb, M George-Nascimento, B Berland, A Garcia & G Nascetti. 2008. New genetic and ecological data on *Anisakis* spp. (Nematoda: Anisakidae), with evidence for new sibling species. XXV Congress of the Società Italiana di Parassitologia, Pisa 18-21 giugno, 2008. Parassitologia 50(Suppl. 1): 20. [Abstract]

important role as a trophic bridge for parasite flow in marine ecosystems (Abollo *et al.* 1998). We suggest that *D. gigas* might be a parathenic host for cestodes and nematodes in Chilean waters, because this squid is prey of sperm whales (Clarke & Paliza 2000), sharks (Reyes 2005, Pardo-Gandarillas *et al.* 2007), swordfish (Ibáñez *et al.* 2004, Castillo *et al.* 2007) and sea lions (George-Nascimento *et al.* 1985), predators that are the definitive host for the parasites recorded in this study.

For confirmation of the taxonomic status level of larval stages of the parasites we suggest the use of molecular tools.

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