



Profilicollis chasmagnathi (Acanthocephala) parasitizing freshwater fishes: paratenicity and an exception to the phylogenetic conservatism of the genus?

E. Levy¹ · M.A. Rossin¹ · P.E. Braicovich¹ · J.T. Timi¹

Received: 2 April 2020 / Accepted: 21 July 2020
© Springer-Verlag GmbH Germany, part of Springer Nature 2020

Abstract

Polymorphid acanthocephalans are parasites of marine mammals, waterfowl and ichthyophagous birds. Among these, the genus *Profilicollis* is known to use exclusively decapods as intermediate hosts. Here, we report the first record of living cystacanths of *Profilicollis* parasitizing the body cavity of a fish host, *Oligosarcus jenynsii*, inhabiting the freshwater section of an estuarial system, Mar Chiquita coastal lagoon, in south-east Buenos Aires Province, Argentina. In this environment, cystacanths of *Profilicollis chasmagnathi* have been previously recorded infecting decapod crabs and as transient accidental infections in the gut of some carcinophagous fishes. In the present study, larvae from the crab *Neohelice granulata*, from the intestine of the estuarine fish *Odontesthes argentinensis* and from the body cavity of *O. jenynsii* were morphologically and genetically compared, confirming their identity as *P. chasmagnathi*, a species characteristic of estuaries and marine coasts along Argentina, Uruguay and Chile. These findings can be interpreted as a possible case of incipient paratenicity for *Profilicollis*, and a colonization event of freshwater habitats, probably promoted by the highly variable conditions, typical of ecotonal environments. In addition, cystacanths of the genus *Polymorphus* were also found in *O. jenynsii*, representing the first record of this genus in *Oligosarcus* from Argentina.

Key words *Profilicollis chasmagnathi* · phylogenetic conservatism · paratenicity · freshwater colonization

Introduction

Acanthocephalans of the family Polymorphidae Meyer, 1931 are parasites of marine mammals, waterfowl and fish-eating birds (Schmidt and Huggins 1973). At present, fifteen polymorphid genera are recognized worldwide (Amin 2013; Presswell et al. 2020), whose life cycles typically include crustaceans as intermediate hosts and, in some cases, fish as paratenic hosts (Schmidt 1985). Among them, *Profilicollis* Meyer, 1931 was considered as a subgenus of *Polymorphus*

Lühe, 1911 for many decades, until Nickol et al. (1999) reinstated its generic status based on ecological evidences. These differences are mainly related to life cycle characteristics; indeed, whereas members of *Polymorphus* use amphipods as intermediate hosts, those of *Profilicollis* use decapods (Nickol et al. 1999, 2002). Amphipods were the ancestral intermediate hosts, while the association with decapods represents episodes of secondary colonization as shown by phylogenetic studies that support the monophyly and the validity of the genus *Profilicollis* (García-Varela and Pérez-Ponce de León 2008; García-Varela et al. 2013).

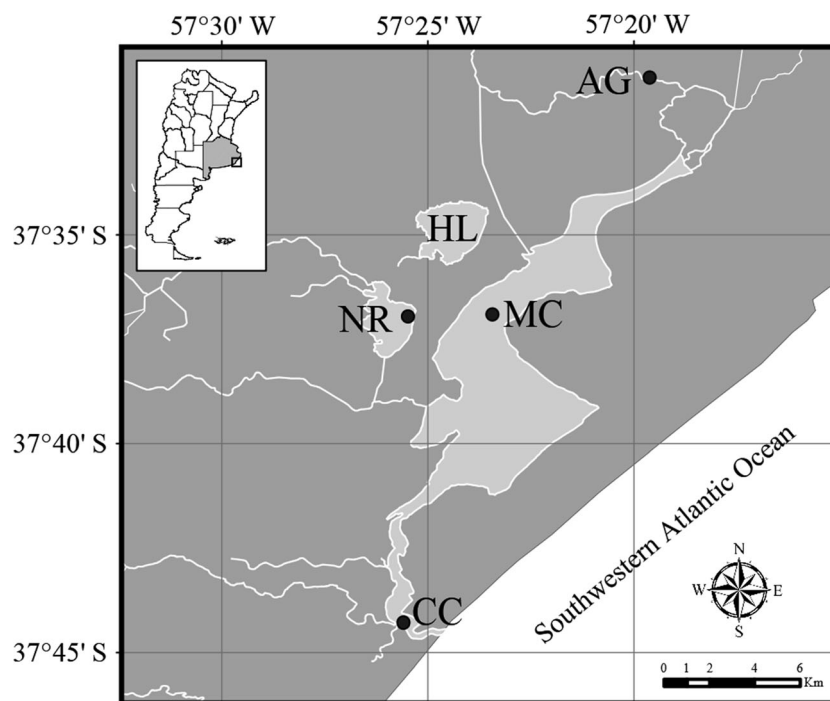
An additional difference between *Polymorphus* and *Profilicollis* seems to be the use of fish as paratenic hosts by the former, a feature shared by other polymorphid genera, i.e. *Andracantha* Schmidt, 1975, *Bolbosoma* Porta, 1908 and *Corynosoma* Lühe, 1911 (Aznar et al. 2006). Indeed, cystacanths of *Polymorphus* have been reported parasitizing the internal organs of several freshwater fish in America (Amin et al. 1995; Santos et al. 2008; García-Prieto et al. 2010; Alcántar-Escalera et al. 2013; Rauque et al. 2018). On the other hand, cystacanths of *Profilicollis* have occasionally

Section Editor: Shokoofeh Shamsi

✉ M.A. Rossin
mrossin@mdp.edu.ar

¹ Laboratorio de Ictioparasitología, Instituto de Investigaciones Marinas y Costeras (IIMyC), Facultad de Ciencias Exactas y Naturales, Universidad Nacional de Mar del Plata- Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), (7600) Mar del Plata, 3350 Funes, Argentina

Fig. 1 Map of the study area showing Mar Chiquita coastal lagoon and sampling sites (black dots), its main tributary streams, and Nahuel Rucá and Hinojales lakes, in Buenos Aires Province, Argentina. Locality References: AG: Arroyo Grande stream, HL: Hinojales lake NR: Nahuel Rucá lake, MC: Mar Chiquita coastal lagoon, CC: estuarine area near Cangrejo creek discharge



been found in the stomach and intestinal contents of fishes (Alarcos and Etchegoin 2010).

Proflicollis chasmagnathi (Holcman-Spector, Mañé-Garzón & Dei-Cas, 1977) is the only representative of the genus so far reported at adult stage along the southwestern Atlantic coast (Lorenti et al. 2018). This species has been recorded at the estuaries of Buenos Aires Province and Patagonian coasts, infecting the gut of several bird species (Martorelli 1989; Vizcaíno 1989; Diaz et al. 2011; La Sala et al. 2013; Lorenti et al. 2018). Cystacanths of *this species* are common parasites of different crab species inhabiting estuarine and rocky intertidal marine habitats in Uruguay and Argentina (Holcman-Spector et al. 1977; Martorelli 1989; Alda et al. 2011; La Sala et al. 2012; Méndez Casariego et al. 2016; Rodríguez et al. 2017). These cystacanths occur at high prevalence in two varunid crab species, *Cyrtograpsus angulatus* (Dana, 1851) and *Neohelice granulata* (Dana, 1851) in soft bottom intertidal areas and salt marshes of the Mar Chiquita coastal lagoon, Buenos Aires Province, Argentina (Martorelli 1989; Méndez Casariego et al. 2016). These crabs have also been found in the stomach and intestine, along with cystacanths of *P. chasmagnathi*, of several marine fish species that use the estuary as feeding grounds (Alarcos and Etchegoin 2010).

During parasitological studies on estuarine and freshwater fishes from the Mar Chiquita basin, cystacanths referable to *Proflicollis* were found in the intestine of the silverside *Odontesthes argentinensis* (Valenciennes, 1835) (Atherinopsidae) from estuarine areas. Similar larvae were

also found in the mesenteries of the dientudo *Oligosarcus jenynsii* (Günther, 1864) (Characidae) from a freshwater tributary of the lagoon, along with larvae of the genus *Polymorphus* in the same microhabitat. This was an unexpected finding, because members of *Proflicollis* have never been recorded infecting the internal organs of fishes.

To the best of our knowledge, there is a single previous record of polymorphid acanthocephalans in the genus *Oligosarcus*, namely of cystacanths of *Polymorphus* sp. in *O. hepsetus* (Cuvier, 1829) from Brazil (Abdallah et al. 2004). The closely related genera *Proflicollis* and *Polymorphus* can be very difficult to distinguish based only on their morphology because they display some degree of overlapping in diagnostic features (Amin 1992). Furthermore, species within *Proflicollis* are very similar to each other (Rodríguez and D'Elía 2016; Rodríguez et al. 2016). Consequently, the aim of this research was to identify those larval acanthocephalans found parasitizing *O. jenynsii* to get insight on the potential departures from the typical life cycle (two-host and marine-estuarine) of *Proflicollis*, which could be driven by the variability in the ecological conditions imposed by an ecotonal freshwater-estuarine environment.

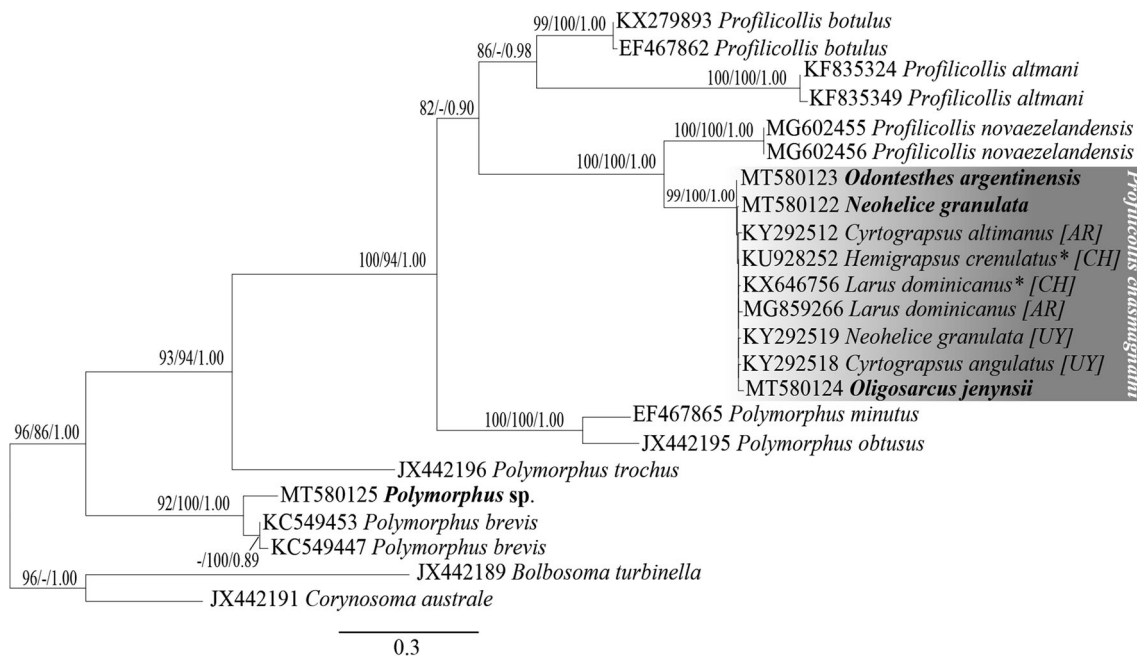


Fig. 2 Maximum likelihood tree inferred by mtDNA *cox1* showing the taxonomic position of *Profilicollis chasmagnathi* and *Polymorphus* sp. from three host species (*Neohelice granulata*, *Oligosarcus jenynsii* and *Odontesthes argentinensis*). Nodal supports are indicated for ML, MP (1,000 replicates, only bootstrap values greater than 70% are shown)

and BI (only posterior probabilities greater than 0.7 are shown). Names in bold correspond to sequences obtained in the present study. Asterisks indicate sequences deposited as *P. antarcticus* that were posteriorly identified as *P. chasmagnathi* (KU928252, Rodríguez and D'Elia 2016; KX646756, Rodríguez et al. 2016)

Materials and Methods

Study area

Fish samples were obtained at three different locations of the Mar Chiquita basin. This is a coastal lagoon in south-east Buenos Aires Province, Argentina. It is an elongated water body, parallel to the coastline, separated from the sea by a barrier of dunes and only connected to it by an inlet that seasonally varies both in width and in position (Isla 1997). Mar Chiquita is characterized by a marked salinity gradient, with values ranging from 29.15 (at the inlet area) to 2.8 (at its innermost section) (Marcovecchio et al. 2019). These authors identified three different areas, an external area functioning as a coastal marine system, an intermediate estuarine system and

an inland water system, whose extents vary inter-annually depending on the rainfall. For example, the extent of the marine section varied between 12 and 71% of the total surface across rainy and dry years (Marcovecchio et al. 2019). The limit between these sections is also variable daily and seasonally depending on the amplitude of the tides, the meteorological conditions and the volume of fresh water present in the lagoon (Reta et al., 2001). The lagoon, with a total area of ~60 km² receives waters from numerous streams (Marcovecchio et al. 2019) being also in connection with two Pampean shallow lakes, Nahuel Rucá and Hinojales. One of its main tributaries is Arroyo Grande stream, to the north of the lagoon. This system is therefore a vast ecotonal region of great biodiversity that combines freshwater environments, wetlands and grasslands with marshes, coastal dunes and beaches (Fig 1).

Table 1 Prevalence and mean abundance, 95% confidence intervals in parenthesis, of *Profilicollis chasmagnathi* and *Polymorphus* sp. parasitizing *Oligosarcus jenynsii*, *Odontesthes argentinensis* and *Neohelice granulata* from Mar Chiquita basin, Buenos Aires Province,

Argentina. Locality References: AG: Arroyo Grande, CC: estuarine area near Cangrejo creek discharge, MC: Mar Chiquita lagoon, NR: Nahuel Rucá lake

	<i>Profilicollis chasmagnathi</i>					<i>Polymorphus</i> sp.		
	Locality	N	Infected	Prevalence (%)	Mean Abundance	Infected	Prevalence (%)	Mean Abundance
<i>Oligosarcus jenynsii</i>	AG	30	7	23.3 (11; 41)	0.43 (0.2; 0.9)	1	3.3 (0.2; 16.3)	0.07 (0; 0.2)
<i>Oligosarcus jenynsii</i>	NR	214	0	0	0	27	12.6 (8.6; 17.6)	0.18 (0.1; 0.3)
<i>Odontesthes argentinensis</i>	MC	40	11	27.5 (16; 44)	0.45 (0.2; 0.8)	0	0	0
<i>Neohelice granulata</i>	CC	3	3	100 (37; 100)	8.33 (6.0; 10.7)	0	0	0

Host samples

Fish were captured, by means of gillnets, at different locations during parasitological surveys in the lagoon. A total of 30 specimens of *O. jenynsii* were obtained at Arroyo Grande (AG: 37°31'14"S, 57°19'37" W; April 2018) and 40 *O. argentinensis* were caught inside the lagoon body (MC: 37°36'54"S, 57°23'26"W; April 2017–August 2018). Additionally, a sample of three decapod crabs, *N. granulata*, collected by hand in an estuarine area near the freshwater discharge of Cangrejo Creek (CC: 37°44'14"S, 57°26'18"W; March 2019), was used to obtain cystacanths from the natural intermediate host. Also data from a sample of 214 *O. jenynsii*, caught at Nahuel Rucá lake (NR: 37°36'57"S, 57°25'29"W; July 2008–October 2012) was used for comparative purposes (Fig. 1).

Parasitological studies and morphology

All hosts were frozen at -20°C until examination. After thawing at room temperature, they were necropsied and the polymorphid acanthocephalans were collected. Prevalence and mean abundance (sensu Bush et al. 1997) were calculated for each parasite species in each host sample, with 95% bootstrap confidence intervals, following Rózsa et al. (2000), using Quantitative Parasitology software (QPweb) (Reiczigel et al. 2019). The microhabitat and state of preservation of each cystacanth was recorded, considering larvae as viable when entire and encysted, or as dead when partially reabsorbed (darkened and degraded).

Complete individuals were kept in distilled water for several hours in order to allow the eversion of the proboscis prior to fixation; they were later identified at generic level following Amin (1992) and Presswell et al. (2020). A single *Proflicollis* sp. from each host species and one *Polymorphus* sp. found parasitizing *O. jenynsii* were reserved for genetic

identification (fixed and preserved in 96% ethanol). The remaining fully extended specimens were fixed in 4% formalin to later perform morphological studies under a stereoscopic microscope. Measurements included total length, proboscis length and width, neck length and width, somatic spines area length and width and hind trunk length and width. In addition, the number of hook rows and hooks per row was recorded. These variables were used in the taxonomical identification of the cystacanths, to the lowest level possible.

DNA extraction, PCR amplification, sequencing and sequence analysis

DNA extractions were carried out using the DNeasy Blood and Tissue® kit (QIAGEN, Hilden, Germany) on whole specimens. A region of the mitochondrial DNA was amplified by polymerase chain reaction (PCR). Mitochondrial cytochrome c oxidase subunit 1 (mtDNA *cox1*) was amplified using the universal DNA primers LCO1490 (forward) and HCO2198 (reverse) described by Folmer et al. (1994). All PCR reactions were set up in 25 µl reactions using 5 µl of DNA (≥10 ng) as the template, 0.5 µl (0.5 mM) of each primer and 12.5 µl (2X) HotStarTaq Master Mix (QIAGEN). The PCR was carried out using the following conditions: initial step for enzyme activation and denaturation at 95 °C for 15 min, followed by 35 cycles of amplification at 94 °C for 30 s, 50 °C for 30 s and 72 °C for 1:45 min, followed by 10 min of post amplification at 72 °C. Each PCR product was purified using QIAquick spin columns (QIAquick Gel Extraction Kit, QIAGEN). Sequencing was performed using ABI 3730XLs automated sequencer (Applied Biosystems, Macrogen, South Korea).

Sequences were edited and assembled in Proseq 3.5 (Filatov 2002). For identification, consensus sequences were compared against the NCBI database using the BLAST algorithm (Altschul et al. 1990). Curated contig sequences were

Table 2 Measurements of *Proflicollis chasmagnathi* parasitizing *Oligosarcus jenynsii*, *Odontesthes argentinensis* and *Neohelice granulata*. Measurements are given in micrometres (µm)

	<i>Oligosarcus jenynsii</i> (n=8)		<i>Odontesthes argentinensis</i> (n=4)		<i>Neohelice granulata</i> (n=6)	
	Mean	Range	Mean	Range	Mean	Range
Total Length	5179.4	4665–5473	5882.9	5429–6177	5878.7	5109–6536
Proboscis Length	624.1	535–756	668.8	619–743	696.6	605–738
Proboscis Width	472.1	398–527	512.9	469–613	526.1	501–551
Neck Length	1463.6	1303–1800	1633.3	1462–17516	1530.0	1274–1775
Neck Width	370.4	296–499	312.9	264–350	359.2	334–404
Somatic Spines Area Length	1299.5	973–1830	1581.6	1511–1705	1702.2	1486–1904
Somatic Spines Area Width	633.5	582–685	646.3	538–721	804.1	734–886
Hind Trunk Length	1606.9	1552–1680	1999.2	1791–2158	1950.0	1743–2148
Hind Trunk Width	846.3	615–995	994.3	958–1049	1062.9	1023–1122

deposited in GenBank (Accession numbers: MT580122, MT580123, MT580124, MT580125, Fig. 2)

The obtained fragments from the mtDNA *cox1* gene were aligned with sequences from other members of the genera *Proflicollis* and *Polymorphus*, retrieved from GenBank. Sequences from *Bolbosoma turbinella* (Diesing, 1851) and *Corynosoma australe* Johnston, 1937 were also aligned and used as outgroup. The alignment was performed by ClustalW (Thompson et al. 1994) as implemented in MEGA 7.0 software package (Kumar et al. 2016), using default parameters. The reading frame for the mtDNA *cox1* sequences was determined by translating the sequences, specifying the appropriate gene code (invmDNA) and by starting at different positions in the alignment and inspecting for stop codons.

The estimation of intra and interspecific genetic divergence among specimens was conducted in MEGA7.0 software package (Kumar et al. 2016), using the Tamura-Nei model (Tamura and Nei 1993).

Hypothesis on the specific identity of the cystacanths found in different host species were tested using evolutionary three inference methods, based on analysis of character-state data, as recommended by Nadler and Pérez-Ponce de León (2011), which has the advantage of revealing the particular changes in states supporting individual species. For that purpose, three different inference methods, namely maximum parsimony (MP), maximum likelihood (ML) and Bayesian inference (BI), were used to construct trees in order to visualize relationships between the sequences obtained in the present study and those from cystacanths and adults retrieved from Genbank. MP analyses were performed using PAUP* 4.0b10 (Swofford 2001), using a heuristic search with tree-bisection-reconnection (TBR), branch swapping and random addition of sequence. All characters were treated as unordered. ML analyses were performed using PhyML 3.1 (Guindon and Gascuel 2003).

Reliabilities of phylogenetic relationships were evaluated using nonparametric bootstrap analysis (Felsenstein 1985) with 1,000 replicates for MP and ML trees. Bootstrap values exceeding 70 were considered well supported (Hillis and Bull 1993). Bayesian inference was performed with MrBayes 3.1.1 (Ronquist and Huelsenbeck 2003). The Bayesian posterior probability analysis was performed with the MCMC algorithm where the number of chains was 4, the temperature of heated chains was 0.2 with 1,000,000 generations while the sub-sampling frequency was 100, with a burn-in fraction of 0.25. JModelTest (Posada 2008) was run to determine the best-fit model for the obtained data set, as implemented in the Akaike information criterion (AIC) (Posada and Buckley 2004). The best-fit model GTR + I + G was used for BI analysis and ML.

Results

Larval acanthocephalans, referable to *Proflicollis*, were found encysted in the intestinal mesenteries of *O. jenynsii* caught at AG. Similar cystacanths were found free in the intestinal contents of *O. argentinensis* from MC and in the body cavity of *N. granulata* (the natural host) from CC. A relatively high prevalence and low mean abundance of *Proflicollis* were recorded in both fish species (Table 1). In addition, a specimen of *O. jenynsii* from AG had two cystacanths referable to *Polymorphus* encysted in its intestinal mesenteries. In comparison, those *O. jenynsii* from NR were parasitized only by *Polymorphus* sp., at lower prevalence and mean abundance than in AG (Table 1).

The cystacanths found in *O. argentinensis* were in most cases associated with remnants of the crab *C. angulatus*, present in the intestinal contents. All of them had their proboscis invaginated and were therefore considered as viable. Some *O. argentinensis* presented encapsulated lesions in their intestinal wall, with dark, degraded contents that, in some cases, contained remains of proboscis that resembled those of *Proflicollis* sp. These lesions could indicate previous attachment sites; however, they were not quantified as *Proflicollis* sp., since their identity could not be asserted. On the other hand, all cystacanths found on *O. jenynsii* were encysted in the mesenteries and evidenced a strong reaction by the host, represented by brownish encapsulations. Despite this, 46% of them were identified as viable.

The *Proflicollis* sp. collected from the three host species (*O. argentinensis*, *O. jenynsii* and *N. granulata*) were morphologically similar to each other, with a constant number of hook rows in the proboscis (18) and of hooks in each row (8), although larvae from *O. jenynsii* showed slightly smaller body measurements than those from crabs and *O. argentinensis* (Table 2). Morphological and morphometric data allowed determining them as *P. chasmagnathi* according to Amin, 1992 and Vizcaíno, 1989.

Further confirmation was accomplished through the genetic studies, which demonstrated that all specimens studied were in effect *P. chasmagnathi*. The mtDNA *cox1* sequences (637 bp) from the cystacanths obtained from *N. granulata* crabs, *O. jenynsii* and *O. argentinensis* matched >99% with the mtDNA *cox1* sequences of *P. chasmagnathi* deposited in GenBank. The genetic divergences between the *P. chasmagnathi* from the present study and those from GenBank were in average 1.0% (range: 0.9% - 1%). When compared with sequences from congeners available in GenBank (*P. botulus* (Van Cleave, 1916), *P. altmani* (Perry, 1942) and *P. novaezelandensis* Brockerhoff & Smales, 2002), the interspecific divergences ranged from 16 to 30%.

Regarding those larvae morphologically determined as *Polymorphus* sp., the closest match of a mtDNA *cox1* sequence from the cystacanth found in *O. jenynsii* (631 bp)

was 93.8% to a sequence of *Polymorphus* deposited in GenBank. The genetic divergence among species of *Polymorphus* retrieved from GenBank varied between 7.3, with *P. brevis* (Van Cleave, 1916), to 42%, with *P. minutus* (Zeder, 1800).

The results from all genetic analyses are summarized in a single tree with bootstrap and posterior probability values, as shown in Fig. 2. The ML analysis resulted in a single tree with -log likelihood: 4382.4844. The MP analysis revealed that 333 characters were constant; 293 were parsimony-informative, and 27 variable characters were parsimony-uninformative. For BI analysis, the average standard deviation of split frequencies was 3.29×10^{-3} , after 1×10^6 generations. Trees based on ML, MP and BI analysis yielded similar topologies, as shown in the ML consensus tree (Fig. 2).

In summary, the mtDNA *cox1* analysis allowed determining that the *Profilicollis* cystacanths sequenced in this study were in effect *P. chasmagnathi*, while the specific status of the *Polymorphus* sp. found in *O. jenynsii* could not be further asserted, although its sequence was included as a sister taxon to *P. brevis* in a well-supported clade.

Discussion

An integrative approach, combining morphological and genetic evidences allowed confirming that all cystacanths of the genus *Profilicollis* found in *O. argentinensis* from MC, *O. jenynsii* from AG and *N. granulata* from CC, are conspecific and belong to *P. chasmagnathi*. Those cystacanths of *Polymorphus* sp. found in *O. jenynsii* could not be morphologically determined beyond the genus level due to their larval stage, since the diagnostic characters at species level often rely on the structure and morphology of the reproductive system in both sexes (Presswell et al. 2020).

The genetic analysis showed that the relationships among *Profilicollis* and *Polymorphus* species, recovered in this study, are congruent with those from previous studies (García-Varela et al. 2013; Huston et al. 2020). Indeed, phylogenetic relationships among species of *Polymorphus* were poorly resolved, indicating that, as currently constituted, *Polymorphus* is not monophyletic (García-Varela et al. 2013), with the present material being placed genetically closer to *P. brevis* among the compared congeners. On the other hand, sequences of *Profilicollis* obtained in this study resulted in a well-supported clade with all sequences available of *P. chasmagnathi* (cystacanths and adults) from South American hosts. The low genetic divergence of *P. chasmagnathi* across host species was congruent with those values from previous studies in the region (Rodríguez et al. 2017; Lorenti et al. 2018). A diagnostic feature of the genus *Profilicollis* is the use of decapod crustaceans as intermediate hosts (Nickol et al. 1999, 2002) and up to the present, the lack

of paratenic hosts, being directly transmitted to aquatic birds (its definitive hosts) through the consumption of crustaceans (Lorenti et al. 2018). *Profilicollis chasmagnathi* has been reported in several species of estuarine-dependent fishes (*O. argentinensis*, *Paralichthys orbignyanus* (Valenciennes, 1839), *Micropogonias furnieri* (Desmarest, 1823) and *Pogonias cromis* (Linnaeus, 1766)) from Mar Chiquita (Alarcos and Etchegoin 2010). The authors found these acanthocephalans in the gut of fishes, consequently, such infections should be considered as accidental and transient, being the result of predation on the decapod crabs inhabiting the lagoon. In fact, although crabs harbouring *Profilicollis* larvae are often predated by teleosts and elasmobranchs, it is considered that these fish do not play any role in the life cycle of the parasite (Oliva et al. 2008). In the present study, the presence of tissue lesions containing acanthocephalan hooks in the mucosa of some specimens of *O. argentinensis*, but not of attached worms, supports the idea of the transient nature of such infections in this hosts. This is supported by the morphometric similitude between worms from crabs and silversides, indicating that larvae were released from the crab's body during digestion.

On the other hand, the presence of *P. chasmagnathi* in the mesenteries of *O. jenynsii* represents a long-term or permanent infection, with the fish acting as paratenic host, a symbiotic relationship that had not been reported yet for this genus. In the present study, crab remains were found neither in intestinal nor in stomach contents of any dientudo; however, the presence of cystacanths of *P. chasmagnathi* would indicate the previous consumption of infected crabs. *Oligosarcus jenynsii* is a generalist carnivorous and a freshwater species, tending to piscivory at larger sizes (Nunes and Hartz 2006), but freshwater decapods (shrimps and crabs) are frequently reported preys in other localities (Rodrigues et al. 2012). It is therefore possible that it feeds on estuarine crabs when this fish, visits myxo-oligohaline areas, at the north of the lagoon, mostly during winter (González-Castro et al. 2009). It is possible too that the infections occurred in freshwater areas, thanks to the marked euryhalinity of the crabs *N. granulata* and *C. angulatus*, commonly parasitized by *P. chasmagnathi*, especially of the latter, which was abundant in Arroyo Grande (pers. obs.).

The absence of *P. chasmagnathi* in a large sample of *O. jenynsii* from Nahuel Rucá is therefore, explained by the rarity of both crab species in that lake. Indeed, only few specimens of *C. angulatus* were sporadically observed during fish sampling, all of them of a size too large to be prey of *O. jenynsii* (pers. obs.), supporting a crab-fish transmission in Arroyo Grande where small crabs can be eaten by the fish.

The generalized life-cycle pattern of acanthocephalans invariably includes an arthropod intermediate host (Kennedy 2006). In the case of polymorphids, these are crustaceans (García-Varela et al. 2013), which are infected by consuming

the parasite's eggs. After penetrating the gut wall of the crustacean host, the acanthor larvae develops into an acanthella and then into a cystacanth stage (Reish 1950; Rayski and Garden 1961). The acquisition of paratenic hosts, albeit facultative ones, is rare among acanthocephalans (Kennedy 2006); however, it enables them to ascend trophic levels and so move through food chains favouring transmission (Kennedy 2006) by bridging the trophic gap between intermediate and definitive hosts (Aznar et al. 2006). Beyond this evolutionary advantage, sometimes it is difficult to assess whether a host with acanthocephalans in the body cavity is actually a paratenic or an accidental host, in whose body cavity acanthocephalans may also occur (Kennedy 2006). A possible mechanism of the extra-intestinal infection by larval acanthocephalans in fish has been proposed for other genera; in fact, it is not uncommon to find partially excysted parasites in the body cavity and its organs of suitable definitive hosts (Kennedy 2006). According to this author, the age of cystacanths can play a role in the efficiency and site of infection. As examples, De Giusti (1949) and Nickol (1985) found that immature larvae could not effectively attach to their definitive fish hosts and would pass through the intestine wall to encyst (still as a cystacanth) in an extra-intestinal site, while older larvae are able to attach to the intestine and normally progress with their life cycle. This could be the case for those cystacanths found in dientudos, although they are not the definitive hosts, and could explain the smaller size of cystacanths infecting them relative to those found in crabs. Nevertheless, the fact that this fish could be a suboptimal host affecting the parasite's development, as also indicated by the finding of dead worms, cannot be discarded.

Host capture, the colonization of new hosts by parasites (Holmes and Price 1980), seems to be an extended phenomenon in polymorphids, not only at the level of definitive (Kennedy 2006) and intermediate hosts (García-Varela et al. 2013), but also of paratenic hosts. Indeed, birds and mammals have captured *Polymorphus* species from fish, while marine mammals, may have captured *Corynosoma* and *Bolbosoma* species from birds (Kennedy 2006). Similarly, the acquisition of decapods as intermediate hosts for some genera (*Profilicollis*, *Arhytmorhynchus* Lühe, 1911, *Ibirhynchus* García-Varela, Pérez-Ponce de León, Aznar & Nadler, 2011 and *Hexaglandula* Petrochenko, 1950) and of euphausiids for *Bolbosoma*, represent episodes of secondary colonization from amphipods, the ancestral intermediate hosts (García-Varela et al. 2013). Therefore, the incorporation of a paratenic host in the life cycle of *Profilicollis*, even in an apparently incipient stage, should not be surprising, since it has been frequent in related genera. The transmission of *P. chasmagnathi* cystacanths to a fish paratenic host implies their ability to penetrate the fish's intestine and re-encyst in the mesenteries. This process is the same followed by the phylogenetically closely related genus *Polymorphus* (Alcántar-

Escalera et al. 2013; Huston et al. 2020; Presswell et al. 2020), as well as by other polymorphids, such as *Andracantha*, *Bolbosoma* and *Corynosoma* (Aznar et al. 2006; García-Varela et al. 2013). On the other hand, the disability of *P. chasmagnathi* to parasitize the internal organs or tissues of other fish species, in whose guts is frequently found (Alarcos and Etchegoin 2010; present study), could represent a kind of host specificity and requires further research, although differential host defence mechanisms could be involved. The finding of dark lesions in the intestines of some silversides containing proboscis remains could be indicative of such defences. For example, the polymorphid *Corynosoma strumosum* (Rudolphi, 1802) may or may not respond secreting a protective thick layer of glycocalyx on its tegument (Skorobrekova and Nikishin 2017) depending on the nature of the encapsulation defence mounted by the different species of paratenic fish hosts (varying from fibroblastic to leukocytal). In consequence, *C. strumosum* shows variable degrees of adaptation to disparate hosts (Skorobrekova and Nikishin 2017; Nikishin and Skorobrekova 2019).

Beyond the possible role of *O. jenynsii* as host of *P. chasmagnathi* could be considered as accidental or as an incipient paratenicity phenomenon, a high proportion of these larvae found were alive at the time of capture of these hosts. This implies that they could be viable for the infection of ichthyophagous birds, which would potentially enable *P. chasmagnathi* to widen its host range. Such facts are relevant, especially considering the low specificity of this parasite for its definitive hosts. Indeed, *P. chasmagnathi* has been reported in six families of birds, belonging to five orders (Martorelli 1989; Vizcaino 1989; Torres et al. 1993; Diaz et al. 2011; La Sala et al. 2013; Lorenti et al. 2018), including podicipedids and phalacrocoracids, which are primarily piscivorous (Pettracci et al. 2009; Josens et al. 2010) and abundant in Mar Chiquita basin (Favero et al. 2001; Ferrero and Iribarne 2001).

The capture of a fish paratenic host challenges the phylogenetic conservatism of the genus *Profilicollis*. However, host specificity cannot be considered a fixed trait, because in spite of being phylogenetically constrained to a large extent, it is strongly influenced by local environmental conditions (Mouillot et al. 2006), which cause considerable variation in realized host specificity (Wells and Clark 2019). Indeed, it has been proposed that estuaries and coastal-brackish lagoons are environments physically variable enough to select generalist genotypes of fish, in order they can adjust their morphology, physiology and behaviour to a wide range of conditions (Bamber and Henderson 1988). This selected plasticity would pre-adapt estuarine populations to invade, colonize and radiate into vacant niches in freshwater (Bamber and Henderson 1988), an eco-evolutionary mechanism that could explain a new host-parasite system, such as *P. chasmagnathi*-*O. jenynsii*.

The present findings represent the first record of the incorporation of a paratenic host in the life cycle of a member of the genus *Proflicollis*, and consequently an exception to the phylogenetic conservatism characteristic of this genus. Regarding the ecology of *Proflicollis* it would also imply a possible mechanism for the colonization of both a freshwater host and the freshwater environment, by an acanthocephalan genus 'exclusive' of marine and brackish habitats. This transition is probably promoted by the highly variable environmental conditions, typical of ecotonal environments between marine and freshwater realms, such as Mar Chiquita coastal lagoon.

Acknowledgements The authors would like to thank Lic. M. Graziano (Laboratorio de Biología Molecular, Instituto de Investigaciones Marinas y Costeras (IIMyC), UNMdP-CONICET) for her help and advice during the molecular studies, Dr. M. González-Castro (IIMyC) for his help in the capture and determination of the fish in Mar Chiquita and Arroyo Grande, and Mr. P. Urrutia for allowing the samplings to be carried out in his property (Nahuel Rucá lake). Finally, Mr. J. Levy, Mr. S. Cruz and Mrs. L. Fito for their help in the collection of samples.

Funding This study was financed by the Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET) (PIP No. 112-201501-00973), the Fondo para la Investigación Científica y Tecnológica (PICT No. 2013) and the Universidad Nacional de Mar del Plata (EXA 1016/20).

Compliance with ethical standards All applicable institutional, national and international guidelines for the care and use of animals were followed.

Conflicts of interest/Competing interests The authors declare no conflict of interests.

References

- Abdallah VD, Azevedo RK, Luque JL (2004) Metazoários Parasitos dos lambaris *Astyanax bimaculatus* (Linnaeus, 1758), *A. paraguayensis* Eigenmann, 1908 e *Oligosarcus hepsetus* (Cuvier, 1829) (Osteichthyes: Characidae), do Rio Guandu, Estado do Rio de Janeiro, Brasil. *Rev Bras Parasitol Vet* 13:57–63
- Arcos AJ, Etchegoin JA (2010) Parasite assemblages of estuarine-dependent marine fishes from Mar Chiquita coastal lagoon (Buenos Aires Province, Argentina). *Parasitol Res* 107:1083–1091. <https://doi.org/10.1007/s00436-010-1974-z>
- Alcántar-Escalera FJ, García-Varela M, Vázquez Domínguez E, Pérez-Ponce de León G (2013) Using DNA barcoding to link cystacanths and adults of the acanthocephalan *Polymorphus brevis* in central Mexico. *Mol Ecol Resour* 13:1116–1124. <https://doi.org/10.1111/1755-0998.12090>
- Alda P, La Sala L, Marcotegui P, Martorelli SR (2011) Parasites and epibionts of grapsid crabs in Bahía Blanca estuary, Argentina. *Crustaceana* 84:559–571
- Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ (1990) Basic local alignment search tool. *J Mol Biol* 215:403–410. [https://doi.org/10.1016/S0022-2836\(05\)80360-2](https://doi.org/10.1016/S0022-2836(05)80360-2)
- Amin OM (1992) Review of the genus *Polymorphus* Luhe, 1911 (Acanthocephala: Polymorphidae), with the synonymization of *Hexaglandula* Petrochenko, 1950, and *Subcorynosoma* Hoklova, 1967, and a key to the species. *Qatar Univ Sci J* 12:115–123
- Amin OM (2013) Classification of the Acanthocephala. *Folia Parasitol* 60:273–305. <https://doi.org/10.14411/fp.2013.031>
- Amin OM, Heckmann RA, Mesa R, Mesa E (1995) Description and host relationships of cystacanths of *Polymorphus spindlatus* (Acanthocephala: Polymorphidae) from their paratenic fish hosts in Perú. *J Helminthol Soc Wash* 62:249–253
- Aznar FJ, Pérez-Ponce de León G, Raga JA (2006) Status of *Corynosoma* (Acanthocephala: Polymorphidae) based on anatomical, ecological and phylogenetic evidence, with the erection of *Pseudocorynosoma* n. gen. *J Parasitol* 92:548–564. <https://doi.org/10.1645/GE-715R.1>
- Bamber RN, Henderson PA (1988) Pre-adaptive plasticity in atherinids and the estuarine seat of teleost evolution. *J Fish Biol* 33:17–23. <https://doi.org/10.1111/j.1095-8649.1988.tb05554.x>
- Bush AO, Lafferty KD, Lotz JM, Shostak AW (1997) Parasitology meets ecology on its own terms: Margolis et al. revisited. *J Parasitol* 83: 575–583. <https://doi.org/10.2307/3284227>
- De Giusti DL (1949) The life cycle of *Leptorhynchoides thecatus* (Linton), an acanthocephalan of fish. *J Parasitol* 35:437–460. <https://doi.org/10.2307/3273647>
- Díaz JI, Cremonte F, Navone GT (2011) Helminths of the kelp gull, *Larus dominicanus*, from the northern Patagonian coast. *Parasitol Res* 109:1555–1562. <https://doi.org/10.1007/s00436-011-2396-2>
- Favero M, Bachmann S, Copello S, Mariano-Jelicich R, Silva MP, Ghys M, Khatchikian C, Mauco L (2001) Aves marinas del sudeste bonaerense. In: Iribarne O (ed) Reserva de Biósfera Mar Chiquita: Características Físicas, Biológicas y Ecológicas. Editorial Martín, Mar del Plata, Argentina, pp 251–267
- Felsenstein J (1985) Confidence Limits on Phylogenies: An Approach Using the Bootstrap. *Evolution* 39:783–791. <https://doi.org/10.1111/j.1558-5646.1985.tb00420.x>
- Ferrero L, Iribarne O (2001) Avifauna de Mar Chiquita. Síntesis del trabajo de Mariano Manuel Martínez. In: Iribarne O (ed) Reserva de Biósfera Mar Chiquita: Características Físicas, Biológicas y Ecológicas. Editorial Martín, Mar del Plata, Argentina, pp 227–250
- Filatov DA (2002) ProSeq: A software for preparation and evolutionary analysis of DNA sequence data sets. *Mol Ecol Notes* 2:621–624. <https://doi.org/10.1046/j.1471-8286.2002.00313.x>
- Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Mol Mar Biol Biotechnol* 3: 294–299
- García-Prieto L, García-Varela M, Mendoza-Garfías B, Pérez-Ponce de León G (2010) Checklist of the Acanthocephala in wildlife vertebrates of Mexico, vol 1419. *Zootaxa*, pp 1–50. <https://doi.org/10.11646/zootaxa.2419.1.1>
- García-Varela M, Pérez-Ponce de León G (2008) Validating the systematic position of *Proflicollis* Meyer, 1931 and *Hexaglandula* Petrochenko, 1950 (Acanthocephala: Polymorphidae) using cytochrome c oxidase (cox 1). *J Parasitol* 94:212–217. <https://doi.org/10.1645/GE-1257.1>
- García-Varela M, Pérez-Ponce de León G, Aznar FJ, Nadler SA (2013) Phylogenetic relationship among genera of Polymorphidae (Acanthocephala), inferred from nuclear and mitochondrial gene sequences. *Mol Phylogenet Evol* 68:176–184. <https://doi.org/10.1016/j.ympev.2013.03.029>
- González-Castro M, Díaz de Astarloa JM, Cousseau MB, Figueroa DE, Delpiani SM, Bruno DO, Guzzoni JM, Blasina GE, Deli Antoni MY (2009) Fish composition in a south-western Atlantic temperate coastal lagoon: spatial-temporal variation and relationships with environmental variables. *J Mar Biol Assoc UK* 89:593–604. <https://doi.org/10.1017/S0025315409003002>
- Guindon S, Gascuel O (2003) A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. *Syst Biol* 52: 696–704. <https://doi.org/10.1080/10635150390235520>

- Hillis DM, Bull JJ (1993) An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. *Syst Biol* 42:182–192. <https://doi.org/10.1093/sysbio/42.2.182>
- Holman-Spector B, Mañé-Garzón F, Dei-Cas E (1977) Una larva cystacantha (Acanthocephala) de la cavidad general de *Chasmagnathus granulata* Dana, 1851. *Rev Biol Uruguay* 5:67–76
- Holmes JC, Price PW (1980) Parasite communities: the roles of phylogeny and ecology. *Syst Zool* 29:203–213. <https://doi.org/10.2307/2412650>
- Huston DC, Cribb TH, Smales LR (2020) Molecular characterisation of acanthocephalans from Australian marine teleosts: proposal of a new family, synonymy of another and transfer of taxa between orders. *Syst Parasitol* 97:1–23. <https://doi.org/10.1007/s11230-019-09896-2>
- Isla FI (1997) Seasonal behaviour of Mar Chiquita tidal inlet in relation to adjacent beaches, Argentina. *J Coast Res* 13:1221–1232
- Josens ML, Bó MS, Favero M (2010) Foraging ecology of the Great Grebe *Podiceps major* in Mar Chiquita Lagoon (Buenos Aires, Argentina). *Ardeola* 57:133–141
- Kennedy CR (2006) Ecology of the Acanthocephala. Cambridge University Press, Cambridge, UK
- Kumar S, Stecher G, Tamura K (2016) MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Mol Biol Evol* 33:1870–1874. <https://doi.org/10.1093/molbev/msw054>
- La Sala LF, Perez AM, Martorelli SR (2012) Epidemiology of acanthocephalan infections in crabs from the Bahía Blanca Estuary, Argentina. *J Helminthol* 86:446–452. <https://doi.org/10.1017/S0022149X11000678>
- La Sala LF, Perez AM, Smits JE, Martorelli SR (2013) Pathology of enteric infections induced by the acanthocephalan *Profilicollis chasmagnathi* in Olog's gull, *Larus atlanticus*, from Argentina. *J Helminthol* 87:17–23. <https://doi.org/10.1017/S0022149X11000721>
- Lorenti E, Rodríguez SM, Cremonte F, D'Elía G, Díaz JI (2018) Life cycle of the parasite *Profilicollis chasmagnathi* (Acanthocephala) on the Patagonian coast of Argentina based on morphological and molecular data. *J Parasitol* 104:479–485. <https://doi.org/10.1645/17-134>
- Marcovecchio JE, De Marco SG, Magani F, Spetter CV, Beltrame MO, Cionchi JL (2019) Hydraulic stopper effect as a regulator of inorganic nutrients distribution in Mar Chiquita coastal lagoon (Argentina). *Ecohydrol Hydrobiol* 19: 629–641. <https://doi.org/10.1016/j.ecohyd.2019.04.005>
- Martorelli SR (1989) El rol de *Cyrtograpsus angulatus* (Crustacea; Brachyura) en los ciclos de vida de *Microphallus szidati* (Digenea; Microphallidae) y *Falsificollis chasmagnathi* (Acanthocephala: Filicollidae). Algunos aspectos de su ecología parasitaria. *Mem Inst Oswaldo Cruz* 84:567–574. <https://doi.org/10.1590/S0074-02761989000400016>
- Méndez Casariego A, Merlo M, Etchegoin J (2016) Spatial variability of larval parasites harboured by two crab species in an estuarine environment in Argentina. *J Mar Biol Assoc UK* 96:633–637. <https://doi.org/10.1017/S0025315415000594>
- Mouillot D, Krasnov BR, Shenbrot GI, Gaston KJ, Poulin R (2006) Conservatism of host specificity in parasites. *Ecography* 29:596–602. <https://doi.org/10.1111/j.0906-7590.2006.04507.x>
- Nadler SA, Pérez-Ponce de León G (2011) Integrating molecular and morphological approaches for characterizing parasite cryptic species: implications for parasitology. *Parasitology* 138:1688–1709. <https://doi.org/10.1017/S003118201000168X>
- Nickol BB (1985) Epizootiology. In: Crompton DWT, Nickol BB (eds) *Biology of the Acanthocephala*. Cambridge University Press, Cambridge UK, pp 307–346
- Nickol BB, Crompton DWT, Searle DW (1999) Reintroduction of *Profilicollis* Meyer, 1931, as a genus in Acanthocephala: Significance of the intermediate host. *J Parasitol* 85:716–718. <https://doi.org/10.2307/3285748>
- Nickol BB, Heard RW, Smith NF (2002) Acanthocephalans from crabs in the southeastern US, with the first intermediate hosts known for *Arhythmorhynchus frassoni* and *Hexaglandula corynosoma*. *J Parasitol* 88:79–83. [https://doi.org/10.1645/0022-3395\(2002\)088\[0079:AFCITS\]2.0.CO;2](https://doi.org/10.1645/0022-3395(2002)088[0079:AFCITS]2.0.CO;2)
- Nikishin VP, Skorobrekova EM (2019) Two Strategies of acanthocephalan interrelations with paratenic hosts. *Biol Bull Russ Acad Sci* 46: 814–822. <https://doi.org/10.1134/S1062359019080090>
- Nunes DM, Hartz SM (2006) Feeding dynamics and ecomorphology of *Oligosarcus jenynsii* (Günther, 1864) and *Oligosarcus robustus* (Menezes, 1969) in the lagoa Fortaleza, Southern Brazil. *Braz J Biol* 66:121–132. <https://doi.org/10.1590/S1519-69842006000100016>
- Oliva ME, Barrios I, Thatje S, Laudien J (2008) Changes in prevalence and intensity of infection of *Profilicollis altmani* (Perry, 1942) cystacanth (Acanthocephala) parasitizing the mole crab *Emerita analoga* (Stimpson, 1857): an El Niño cascade effect? *Helgol Mar Res* 62:S57–S62. <https://doi.org/10.1007/s10152-007-0082-7>
- Petracci PF, Cereghetti J, Martín J, Obed S (2009) Dieta del Biguá (*Phalacrocorax olivaceus*) durante la primavera en el estuario de Bahía Blanca, Buenos Aires, Argentina. *El Homero* 24:73–78
- Posada D (2008) jModelTest: phylogenetic model averaging. *Mol Biol Evol* 25:1253–1256. <https://doi.org/10.1093/molbev/msn083>
- Posada D, Buckley TR (2004) Model selection and model averaging in phylogenetics: advantages of akaike information criterion and bayesian approaches over Likelihood Ratio tests. *Syst Biol* 53: 793–808. <https://doi.org/10.1080/10635150490522304>
- Presswell B, Bennett JD, Smales LR (2020) Morphological and molecular characterisation of a new genus and species of acanthocephalan, *Tenuisoma tarapungi* n.g., n. sp. (Acanthocephala: Polymorphidae) infecting red-billed gulls in New Zealand, with a key to the genera of the Polymorphidae Meyer, 1931. *Syst Parasitol* 97:25–39. <https://doi.org/10.1007/s11230-019-09898-0>
- Rauque C, Viozzi G, Flores V, Vega R, Waicheim A, Salgado-Maldonado G (2018) Helminth parasites of alien freshwater fishes in Patagonia (Argentina). *Int J Parasitol* 7:369–379. <https://doi.org/10.1016/j.ijppaw.2018.09.008>
- Rayski C, Garden EA (1961) Life-cycle of an acanthocephalan parasite of the eider duck. *Nature* 192:185–186. <https://doi.org/10.1038/192185a0>
- Reiczigel J, Marozzi M, Fabian I, Rozsa L (2019) Biostatistics for parasitologists – a primer to Quantitative Parasitology. *Trends Parasitol* 35:277–281. <https://doi.org/10.1016/j.pt.2019.01.003>
- Reish DJ (1950) Preliminary note on the life cycle of the acanthocephalan, *Polymorphus kenti* Van Cleave, 1947. *J Parasitol* 36:496. <https://doi.org/10.2307/3273182>
- Reta R, Martos P, Perillo G, Piccolo M, Ferrante A (2001) Características hidrográficas del estuario de la laguna Mar Chiquita. In: Iribarne O (ed) *Reserva de Biósfera Mar Chiquita: Características Físicas, Biológicas y Ecológicas*. Editorial Martín, Mar del Plata, Argentina, pp 31–52
- Rodrigues LR, Fontoura NF, da Motta Marques D (2012) Feeding dynamics of *Oligosarcus jenynsii* (Günther, 1864) in a subtropical coastal lake assessed by gut-content analysis and stable isotopes. *IJPAES* 2:126–134
- Rodríguez SM, D'Elía G (2016) Pan-American marine coastal distribution of *Profilicollis altmani* based on morphometric and phylogenetic analysis of cystacanth from the mole crab *Emerita brasiliensis*. *J Helminthol* 91:371–375. <https://doi.org/10.1017/S0022149X16000237>
- Rodríguez SM, D'Elía G, Valdivia N (2016) The phylogeny and life cycle of two species of *Profilicollis* (Acanthocephala: Polymorphidae) in marine hosts off Pacific coast of Chile. *J*

- Helminthol 91:589–596. <https://doi.org/10.1017/S0022149X16000638>
- Rodríguez SM, Díaz JI, D'Elía G (2017) Morphological and molecular evidence on the existence of a single estuarine acanthocephalan species of the genus *Profilicollis* along the Atlantic and Pacific coasts of southern South America. *Syst Parasitol* 94:527–533. <https://doi.org/10.1007/s11230-017-9716-6>
- Ronquist F, Huelsenbeck JP (2003) MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics* 19:1572–1574. <https://doi.org/10.1093/bioinformatics/btg180>
- Rózsa L, Reiczigel J, Majoros G (2000) Quantifying parasites in samples of hosts. *J Parasitol* 86:228–232. [https://doi.org/10.1645/0022-3395\(2000\)086\[0228:QPISOH\]2.0.CO;2](https://doi.org/10.1645/0022-3395(2000)086[0228:QPISOH]2.0.CO;2)
- Santos CP, Gibson DI, Tavares LER, Luque JL (2008) Checklist of the Acanthocephala associated with the fishes of Brazil. *Zootaxa* 1938: 1–22. <https://doi.org/10.5281/zenodo.184999>
- Schmidt GD (1985) Development and life cycles. In: Crompton DWT, Nickol BB (eds) *Biology of the Acanthocephala*. Cambridge University Press, Cambridge, UK, pp 273–305
- Schmidt G, Huggins E (1973) Acanthocephala of South American Fishes. Part 2. Palaeacanthocephala. *J Parasitol* 59(5):836–838. <https://doi.org/10.2307/3278419>
- Skorobrekhova EM, Nikishin VP (2017) The morphological peculiarities of the acanthocephalan *Corynosoma strumosum* (Rudolphi, 1802) (Polymorphidae) in paratenic hosts, the eelpout *Zoarces elongatus* (Kner, 1868) (Zoaridae) and the halibut *Hippoglossus stenolepis* (Schmidt, 1904) (Pleuronectidae). *Russ J Mar Biol* 43:49–56. <https://doi.org/10.1134/S1063074017010126>
- Swofford DL (2001) PAUP*: phylogenetic analysis using parsimony (and other methods) 4. 0:b5
- Tamura K, Nei M (1993) Estimation of the number of nucleotide substitutions in the control region of mitochondrial DNA in humans and chimpanzees. *Mol Biol Evol* 10:512–526. <https://doi.org/10.1093/oxfordjournals.molbev.a040023>
- Thompson JD, Higgins DG, Gibson TJ (1994) CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res* 22:4673–4680. <https://doi.org/10.1093/nar/22.22.4673>
- Torres P, Schlatter R, Montefusco A, Gesche W, Ruiz E, Contreras A (1993) Helminth parasites of piscivorous birds from lakes in the south of Chile. *Mem Inst Oswaldo Cruz* 88:341–343
- Vizcaino SI (1989) Acanthocephalan parasites of argentine birds I. Morphological complements to the knowledge of *Polymorphus (Profilicollis) chasmagnathi* comb. nov. (Polymorphidae). *Stud Neotrop Fauna E* 24:189–192. <https://doi.org/10.1080/01650528909360790>
- Wells K, Clark NJ (2019) Host specificity in variable environments. *Trends Parasitol* 29:203–213. <https://doi.org/10.1016/j.pt.2019.04.001>

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.