Research Articles



First report of marine Gastrotricha from Chilean beaches

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ABSTRACT. A recent study of the meiofauna from several intertidal sites along the coast of Chile yielded several gastrotrich specimens. The number of specimens was too small to assess the ecology of the individual species; however, as there are no published records of Gastrotricha in Chile until recently, it is important to document the presence of this group in order to stimulate further research. We report specimens of the genera *Lepidodasys, Cephalodasys, Macrodasys, Tetranchyroderma, Xenotrichula* and several morphotypes of *Turbanella*. Additionally, we provide some initial data on the population density of *Lepidodasys* sp. at Coihuin (Puerto Montt) and an observation of them feeding on nematodes.

Keywords: Gastrotricha; Lepidodasyidae; Macrodasyidae; Turbanellidae; Thaumastodermatidae; benthic; biodiversity; Chile

INTRODUCTION

Meiofauna are microscopic invertebrates (mostly less than 1 mm in length) that inhabit sediments and secondary substrates (e.g., macroalgae and sessile invertebrates) in both marine and freshwater environments (Giere, 2009). Meiofauna is typically present in high abundances (100-1,000's of individuals per 50 mL sample) and also encompass a very high diversity of organisms from a large number of phyla. Global knowledge about meiofauna is very much biased towards Europe and North America (see, e.g., Balsamo et al., 2008 for freshwater species). The majority of the information on Chilean meiofauna comes from taxonomic papers, starting with the reports of the Lund Expedition to Chile (Brattström & Dahl, 1951). The best-known taxa of Chilean meiofauna are the most abundant, the nematodes and the harpacticoid copepods. The nematodes have been described by Wieser (1953, 1954, 1956, 1959), Lorenzen (1973, 1974, 1975) and Clasing (1980, 1983, 1986). The harpacticoid copepods

The Gastrotricha is one of the less well studied meiofaunal phyla in general, and in Chile there are currently no published reports on their occurrence, diversity or ecology (Lee *et al.*, 2008; Sielfeld, 2008). This is unusual as they are ubiquitous, and frequently abundant in marine sediments along the length of the Chilean coast.

The Gastrotricha currently includes about 841 described species (Kieneke & Schmidt-Rhaesa, 2015; statistics in WoRMS 2018) in two subtaxa: Macrodasyida and Chaetonotida. All gastrotrichs have ventral cilia for locomotion, the characteristic from which their name is derived (from the Greek *gaster*

have been described by Mielke (1985, 1986, 1989, 1992), George (1996, 1998, 2005) and George & Schminke (1999). There have also been some papers on the ecology of Chilean meiofauna, typically at the level of principal groups rather than species, including Neira *et al.* (2001), Rodríguez *et al.* (2001), Sellanes *et al.* (2003), Lee *et al.* (2006) and Lee & Riveros (2012).

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"stomach" and thrix "hairy"). The body is covered with a generally thin cuticle, which in several species can form spines, scales or other solid structures. The body is typically slender, and the head region may be offset from the rest of the body; the head may bear appendages such as palps and/or tentacles. There is a terminal or subterminal mouth of variable width, leading to a short buccal cavity and then to a strong muscular pharynx with a y-shaped (Chaetonotida) or inverted y-shaped lumen (Macrodasyida). Contraction of the radial musculature of the pharyngeal myoepithelium opens the lumen and creates suction for the ingestion of food (usually bacteria, algae or detritus). In most species of Macrodasyida, there are pharyngeal pores at the base of the pharynx through which excess water can be eliminated. Food is passed into a straight intestine that ends in a subterminal anus.

One of the most important sets of taxonomic characters are the adhesive tubes, abundance and distribution of which are extremely important for species determination. The adhesive tubes contain the apical openings of the ducts of two glands. The first gland produces an adhesive and the second a releaser substance in a system known as the duo-gland system. These glands are used by gastrotrichs for position maintenance, allowing them to attach to sediment particles when necessary and then release when they need to move on. In species of Macrodasyida and Neodasys, the adhesive tubes can be present in several regions of the body. Anteriorly, around the ventral mouth margin (TbA), extending along the length of the body in lateral, ventrolateral, dorsolateral and/or dorsal series (abbreviated as follows: TbL, TbVL, TbDL, and TbD, respectively); as a ventral patch toward the posterior end (TbV), or on the posterior end, either inserting directly into the posterior margin or along a tail, or inserting on foot like extensions (TbP). In the Chaetonotida, except for the genus Neodasys, the adhesive tubes are present only on the posterior end and often form a v-shaped furca (see, e.g., Remane, 1936; Ruppert, 1991; Kienecke & Schmidt-Rhaesa, 2015 for overviews on the morphology).

First records of Gastrotricha from Chilean marine and brackish water intertidal sediments from around Coquimbo, Puerto Montt and Punta Arenas, Chile, are reported. Individuals belonging to at least six genera were observed; however, identification to the species level was difficult as optimal equipment was not always available for observations. Nevertheless, it was considered worth documenting the specimens because they are the first reports of gastrotrichs from Chilean waters, hoping that these observations will stimulate further research on this taxon in Chile.

MATERIALS AND METHODS

Qualitative samples were collected at eight sites in three regions of Chile: Coquimbo (~30°S), Puerto Montt (~41°S) and the Magellan Strait (53°S). Additional quantitative samples were taken at Coihuin, Los Lagos only (see below). All samples, unless otherwise stated, were taken in the mid to low intertidal zone. The eight sites were as follows:

Laredo (52°58'8.4"S, 70°49'33.6"W). A small beach within the Centro de Investigación en Acuicultura of the Universidad de Magallanes, located to the northeast of Punta Arenas on the Magellan Strait. The beach consists of medium-fine sand which supports a rich and diverse meiofaunal community, characterized by the presence of abundant interstitial polychaetes of the genus *Polygordius*. The sample was collected on September 29, 2017.

Huinay (42°22'30"S, 72°25'22.8"W). A large muddy and stony estuarine tidal flat located next to the Fundación San Ignacio del Huinay field station at the southern end of the Comau fjord. The samples were collected on October the 7 and 8, 2017.

Coihuin (41°30'0"S, 72°53'2.4"W). An extensive coarse sand tidal flat located just to the east of Puerto Montt. The tidal flats support an abundant and diverse meiofauna community; additionally, an abundant microphytobenthos (MPB) was present. The samples were collected from the mid intertidal on October 1, 2017.

Canal Tenglo (41°29'42"S, 72°59'9.6"W). A small muddy tidal flat located below the Centro i~mar (Universidad Los Lagos), Puerto Montt. This site also supports a rich and diverse meiobenthos and MPB but is also subject to industrial effluent from the adjacent canning plant, shipyard, and marinas. The samples were collected on October 5, 2017.

Estaquilla (41°23'38.4"S, 73°51'32.4"W). An exposed sandy beach with coarse sand located on the Pacific coast near Puerto Montt. This site supports an interstitial fauna dominated by large predatory turbellarians of the family Otoplanidae. Samples were collected from the mid to low intertidal on October 12, 2017.

Totoralillo (30°4'22.8"S, 71°22'33.6"W). An exposed sandy beach to the south of Coquimbo. The bay contains a small island connected to the beach by a sandy isthmus. The beach to the south of the isthmus is composed of medium-fine sand, to the north, the sediments are much coarser and include significant biogenic material. The samples were collected on October 23, 2017.

El Faro (29°54'21.6"S, 71°16'30"W). This exposed sandy beach is composed of medium to fine sand and runs north from Coquimbo to Teatinos (approximately 16 km). Samples were collected from the mid to low intertidal, at La Serena in front of the lighthouse "El Faro" on October 25, 2017.

Teatinos (29°49'30"S, 71°17'31.2"W). This site is at the extreme northern end of the same beach as El Faro. Wave exposure on this beach increases as one move northwards. Samples were collected from the mid to low intertidal on October 26, 2017.

At each site and sampling date, qualitative samples were collected by taking a small volume, approximately 500 mL of sediment with a small shovel, and placing it in a plastic bag for transport to the laboratory. An exception was Coihuin, where five replicate 50 mL quantitative meiofaunal samples were collected every month using a syringe modified to form a piston corer, fixed in 5% formalin and mounted in glycerol (see Lee & Riveros (2012) for the detailed methodology). Where possible, extractions were made on the day of collection; however, in some cases, the samples were stored overnight in a refrigerator (4°C) or cold room (10°C). The sediment was mixed in a large container (~ 2 L) with an excess of isotonic magnesium chloride (MgCl₂ 75g L^{-1}) and left for 10 to 15 min. The fauna was then extracted from the sediment using the decantation method. The sealed jar was agitated for approximately 30 s, and then the supernatant was poured through a 63 or 45 µm sieve. The extracted fauna was then washed from the sieve into a petri dish with seawater and examined under a stereo microscope. Live individual gastrotrichs were picked from the petri dish using a mouth pipette, consisting of a flexible tube attached to a Pasteur pipette that had been heated and drawn to a fine capillary. The specimens were then mounted on standard microscope slides with a coverslip, observed with a compound microscope (Bikon Eclipse E600 in Punta Arenas, Olympus BX43 in Puerto Montt, Nikon in Huinay and Zeiss Primo Star in Coquimbo), and images and videos taken using either a Pentax. Canon or Sony camera depending on the laboratory in which the observations were made. The types of stereo microscopes and compound microscopes varied from location to location.

RESULTS

Nine putative species belonging to five families of Macrodasyida and one family of Chaetonotida were present along with eight sites. One site (Cohuin beach) was very rich in diversity and abundance of gastrotrichs, whereas the other localities revealed only single species (Table 1). For terminology, we use, in accordance to other publications mentioning undetermined gastrotrichs, the following form: the genus name and sp. is followed by the country (Chile) and a letter (A, B, C, D) to distinguish different putative species.

Lepidodasys sp. Chile A

Species of the genus *Lepidodasys* (Fig. 1a) have scales that appear to be spindle-shaped with a central keel and are not imbricated (Fig. 1b). A monthly sampling at Coihuin indicates that the abundance of *Lepidodasys* averages 59 individuals per 50 mL of sediment, with a maximum abundance of 221 individuals in March 2014 and a minimum of 9 individuals in May 2014. *Lepidodasys* was always present in the samples. In the samples from March 2015, one individual was fixed while consuming a nematode of the genus *Microlaimus* (Figs. 1c-d). The individual was being consumed tail first; this behavior has also been observed in predatory nematodes and may be due to the presence of the anteriorly directed setae of the prey.

Cephalodasys sp. Chile A and B, *Macrodasys* sp. Chile A

Specimens from two sites are assigned to the genus Cephalodasys. The specimens from El Faro have a clear separation of the head region from the remainder of the body (Fig. 1e). TbA cannot be observed with certainty. In the documented specimen, there is an uneven number (7) of TbP, with the outermost adhesive tubes distinctly separated from the central tubes (Fig. 1e). In the specimen from Coihuin, the separation of the head from the body is less clearly defined. The TbA are difficult to observe. The specimen has 14 TbP (Fig. 1f) compared to the seven on the El Faro specimen, which taken with the other morphological variation, may suggest that the species present at Coihuin is distinct from that at El Faro. There is an undetermined number of TbL in specimens from both sites. Each of the specimens of Macrodasys has a different number of TbP and degree of tapering of the posterior end (Figs. 1h-i).

The exact number of adhesive tubes cannot be determined, with some individuals having a moderate number and others having significantly more. The differences between the two specimens from Coihuin can be because one being a juvenile specimen. The pharyngeal pores in the specimens from Canal Tenglo are at 65% of the pharynx length. The buccal cavity in all the specimens contains a distal ring of small teeth or flap-like structures (Fig. 1g). TbA could not be observed in any of the specimens.

The specimens belonging to the genera *Tetranchyroderma* (Figs. 2a-b) and *Xenotrichula* (Fig. 2c) show no specific characters beyond those that assigned them to the respective genera. The *Turbanella* specimens are described in more detail here because distinctly different morphs could be differentiated.

Table 1. Biodiversity of gastrotrichs at the eight sampling stations. Numbers indicate relative abundance (1: single specimen, 2: few specimens, 3: many specimens). LA: Laredo, HU: Huinay, CO: Cohuin, CT: Canal Tenglo, ES: Estaquilla, TO: Totoralillo, FA: Faro (La Serena), TE: Punta Teatinos. A, B, C, D: different putative species.

Family	Species	LA	HU	CO	СТ	ES	ТО	FA	TE
Order Macrodasyida									
Lepidodasyidae	Lepidodasys sp. Chile A	-	-	X3	-	-	-	-	-
Cephalodasyidae	Cephalodasys sp. Chile A	-	-	X1	-	-	-	-	-
	Cephalodasys sp. Chile B	-	-	-	-	-	-	X3	-
Macrodasyidae	Macrodasys sp. Chile A	-	-	X2	X2	-	-	-	-
Thaumastodermatidae	Tetranchyroderma sp. Chile A	-	-	X1	-	-	-	-	-
Turbanellidae	Turbanella sp. Chile A, northern species	-	-	-	-	-	X2	-	X2
Turbanellidae	Turbanella sp. Chile B, southern species	-	X2	-	-	-	-	-	-
Turbanellidae	<i>Turbanella</i> sp. Chile C	X1	-	-	-	-	-	-	-
Turbanellidae	<i>Turbanella</i> sp. Chile D	-	-	-	-	X1	-	-	-
Order Chaetonotida									
Xenotrichulidae	Xenotrichula sp. Chile A	-	-	X1	-	-	-	-	-

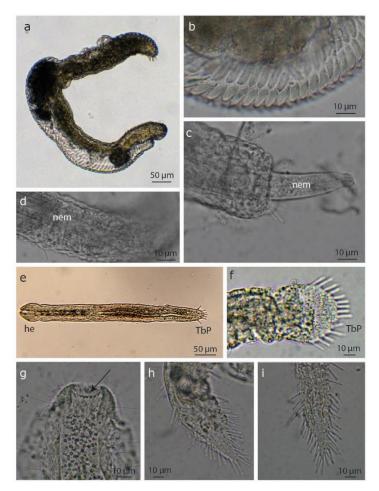


Figure 1. *Lepidodasys.* a) Specimen from Coihuin with "inflated" regions, b) section where the scales are best visible, c) ingestion of a nematode (*Microlaimus* sp.) (nem) sucked in by the muscular pharynx, d) nematode (nem) seen within the *Lepidodasys* specimen; *Cephalodasys* e) specimen from El Faro, entire animal showing the clear separation of the head region (he) and six TbP, f) specimen from Coihuin, posterior end with 14 posterior adhesive tubes (TbP); *Macrodasys* g) specimens from Canal Tenglo, anterior end of showing the tooth-like structures (arrows), h) specimen from Canal Tenglo, the posterior ends showing the the number of TbP, i) specimen from Coihuin, the posterior ends showing the the number of TbP. All images from live specimens except c) and d) from fixed specimens.

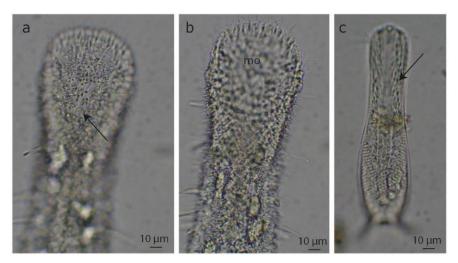


Figure 2. a-b) Two images taken at different focal planes of the same specimen of *Tetranchyoderma* sp. from Coihuin, showing the anterior end with a wide mouth (mo) and some tetrancers (arrow in a), c) *Xenotrichula* sp. from Coihuin showing the cirri (arrow) on the anterior end of the animal.

Turbanella sp.

Records: Laredo on September 29, 2017, one specimen; Huinay on October 7 and 8, 2017, four specimens; Estaquilla on October 12, 2017, one specimen; Totoralillo on October 23, 2017, three specimens; Teatinos on October 26, 2017, two specimens.

Turbanella sp. Chile A (southern morph)

Unless otherwise stated, the four specimens from Huinay all conform to the diagnostic characteristics of the genus *Turbanella* (Todaro & Hummon, 2008).

The length of the specimens is around 1 mm. The head is rounded, slightly offset from the remainder of the body and without lateral extensions (Figs. 3d-e). The mouth is comparatively broad (Fig. 3d). Eyes are present (Figs. 3d-e). There are six (two specimens) or seven (two specimens) TbA on a fleshy base (Fig. 3c). The lengths of the TbA decrease from the lateral to the midline position, with the innermost adhesive tube being the same or shorter than the adjacent adhesive tube (Fig. 3c). There are between 11 and 12 TbL and six and seven TbP per side (Figs. 3b,f,g). The lengths of the TbP decrease from the lateral to the midline position. A medial caudal cone is present (Table 2).

Turbanella sp. Chile B (northern morph)

Unless otherwise stated, the five specimens from Totoralillo and Teatinos all conformed to the diagnostic characteristics of the genus *Turbanella*. The specimens are between 450 and 600 μ m in length. The head is slightly separated from the remainder of the body and is roundish and slender, without lateral extensions (Figs.

4a,b,e,g). The mouth is comparatively small (Fig. 4b). Eyes are present (Figs. 4b,d,g). The number of TbA could be observed in only two specimens; one has seven and the other six or seven adhesive tubes. The tubes increase in length from the lateral to the midline position. The exact number of TbL could not be determined but is around 20 adhesive tubes. The specimens have different numbers of TbP, and the tubes are asymmetrical in four of the five specimens. The specimens from Teatinos had 6/7 (left/right) and 5/6 adhesive tubes (Fig. 4c), the specimens from Totoralillo have 7/8 and 6/7 adhesive tubes (Fig. 4f) and the remaining specimen from Totoralillo had nine adhesive tubes on each side. The length of the adhesive tubes decreases from the lateral to the midline position, with the two outermost tubes distinctly more prolonged than the remaining tubes. In the specimens from Teatinos, the two outermost adhesive tubes were spread to form a "V" shape (Fig. 4c). A caudal cone is present (Figs. 4c,f; Table 2).

The individual specimens from Laredo and Estaquilla were insufficient to observe all the characters necessary to make a reliable identification. The characters that could be observed are summarised in Table 3.

DISCUSSION

Lepidodasys sp.

To date, nine species of *Lepidodasys* have been described around the world. About half of the species have scales with a keel (Hochberg & Atherton, 2011; Lee & Chang, 2011). The nine species have been found in different oceans, but there are very few reports from

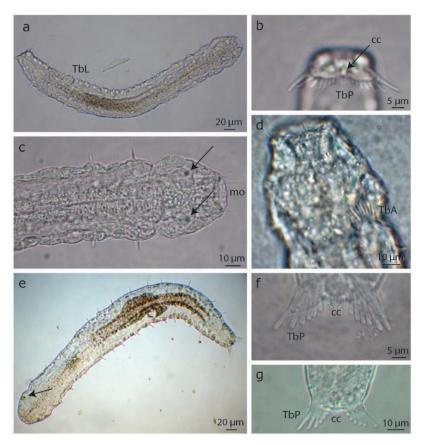


Figure 3. *Turbanella*, Huinay specimens - southern morph. a) Image showing the habitus of specimen 1 where the posterior end is doubled over, also showing the lateral adhesive tubes (TbL), b) enlarged image of the posterior adhesive tubes (TbP) and the caudal cone (cc) of specimen 1, c) anterior end of specimen 1, note the eyes (arrows) and mouth (mo), d) anterior adhesive tubes (TbA) of specimen 2, e) image showing the habitus of specimen 3, note the eyes (arrow), f) the TbP of specimen 3, g) the TbP of specimen 4.

Table 2. Comparison of characters of the southern andthe northern morph of Chilean *Turbanella*.

Characters	Southern morph	Northern morph		
Length	1,000 µm	450-600 μm		
Mouth opening	broad	narrow		
Head constriction	present	present		
Eyes	present	present		
TbA	6-7	6-7		
TbL	11-12	around 20		
TbP	6-7	5-9		
Caudal cone	present	present		

the Pacific. Two species were described from Japan, and an undetermined species was reported from Hawaii (Lee & Chang, 2011). The Coihuin species is the first record from the south-eastern Pacific. The reported case of feeding on a nematode is the first published record of *Lepidodasys* feeding.

Cephalodasys sp.

Thirteen species of the genus *Cephalodasys* have been described (Kieneke *et al.*, 2015), with only one species from the Pacific (*C. pacificus* from the Galapagos, Schmidt, 1974).

Macrodasys sp.

Macrodasys is a diverse genus, with 36 species currently described (Kieneke & Schmidt-Rhaesa, 2015). Only two of these species have been recorded from the Pacific: *M. cunctatus* from Puget Sound (USA Pacific coast) by Wieser (1957) and *M. pacificus* from the Galapagos by Schmidt (1974).

Tetranchyroderma sp.

The single specimen found belongs to the diverse genus *Tetranchyroderma*, of which 82 species are documented (Kieneke & Schmidt-Rhaesa, 2015). Species are generally distinguished by the morphology of the head, distribution of adhesive tubes and the structure of

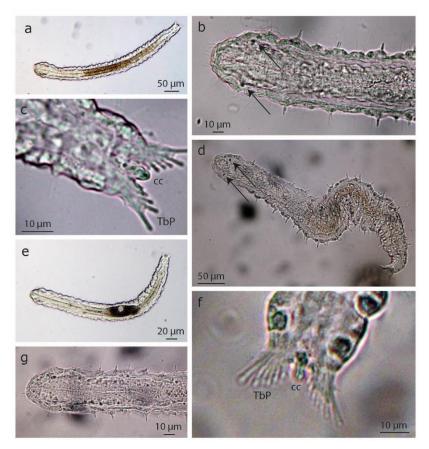


Figure 4. *Turbanella*, Teatinos, and Totoralillo - northern morph. a) Entire specimen 1 from Teatinos, b) anterior ends, note the eyes (arrows in b and d) of specimen 2 from Teatinos, c) posterior ends with posterior adhesive tubes (TbP) and caudal cone (cc) of specimen 2 from Teatinos, d) entire specimen 1 from Totoralillo, e) entire specimen 2 from Totoralillo, f) posterior ends with posterior adhesive tubes (TbP) and caudal cone (cc) of specimen 3 from Totoralillo, g) anterior ends, note the eyes (arrows in b and d) of specimen 2 from Totoralillo.

	Turbanella (Estaquilla)	Turbanella (Laredo)	
Total length in μm	520	undetermined	
Head shape	Slightly separated from the remainder of the body; narrowing towards the anterior end; no lateral extensions (Fig. 5a)	Slightly separated from the remainder of the body; narrowing towards the anterior end; two lateral extensions present (Fig. 5c-d)	
TbA	Probably 5, equal in length	5 on one side and 6 on the other, the innermost tube is shorter than the others	
TbL	9	Not observed	
ТЪР	4 on one side and 5 on the other. Outermost tubes longer than the others and slightly spread (Fig. 5B)	4 on one side and 5 on the other, but one tube is probably missing as there is a gap (Fig. 5e)	
Caudal cone	Present (Fig. 5b)	Absent (Fig. 5e)	

Table 3. A summary of the visible characteristics of the *Turbanella* specimens collected at Laredo and Estaquilla.

the cuticle (as spined scales in the form of triances, tetrancres, pentancres, feathered ancres, etc.; see Todaro, 2002).

Turbanella sp.

The characteristics for the genus are as follows (see Kieneke & Schmidt-Rhaesa, 2015): The head region is

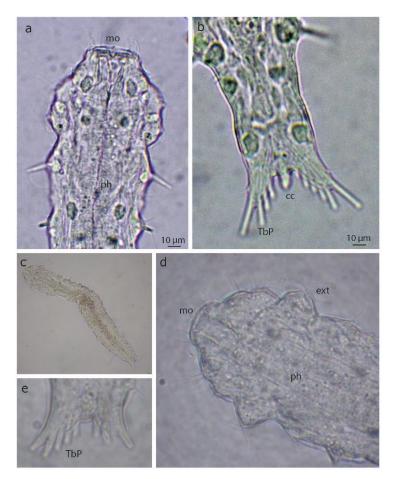


Figure 5. *Turbanella* from Laredo and Estaquilla. a) The anterior end with mouth opening (mo) and pharynx (ph) of the specimen from Estaquilla. Note the separation of the head from the remainder of the body, b) the posterior end of the specimen from Estaquilla showing the posterior adhesive tubes (TbP) and caudal cone (cc), c) the entire specimen from Laredo, d) the anterior end of the Laredo specimen showing the extensions (ext) of the head, e) the posterior end of the Laredo specimen, note that the caudal cone is absent. Scale for c-e not available.

distinctly delimited in most species and may have lateral extensions. TbA is on fleshy feet and is handlike arranged, TbL is abundant and may occur in different positions from ventral to dorsal. TbP is on caudal feet, often with an unpaired median cone between them.

Southern morph

The morphology has some resemblance to two species, *Turbanella corderoi* Dioni, 1960 from Montevideo, Uruguay and *Turbanella brusci* Hochberg, 2002 from the Pacific coast of Australia (North Stradbroke Island, Queensland). In *T. corderoi*, the head is slightly set off against the remaining body. Eyes are figured, but not described. There are 6 TbA on a fleshy base, 15-20 TbL, and 6-7 TbP. (Dioni, 1960). A distinct caudal cone is not present. The Huinay specimens differ from *T*.

corderoi in the number of TbL and the presence of a caudal cone.

The length and the shape of the head region of the Chilean specimens are the same as in *T. brusci*, but eyes were not described for this species (Hochberg, 2002). Hochberg (2002) gives a wide range for the number of adhesive tubes, and the numbers observed on the Huinay specimens fall within these ranges. T. brusci has between 6 and 12 TbA, 10 to 25 TbL and 6 to 9 TbP; and a caudal cone is present. The greatest difference between the Chilean and Australian specimens seems to be the number of TbA. The range of 6 to 12 is explained by the juveniles having fewer TbA than the adults. Therefore, it seems possible that T. brusci has a larger number of TbA than the specimens at Huinay. Nevertheless, the Australian and Chilean populations are very similar to each other and could represent closely related species.

Northern morph

The morphology is similar to *Turbanella hyalina* (Schultze, 1853), as described by Kieneke & Nikoukar (2017); however, there are clear differences. T. hyalina is a species described from northern European waters (North and Baltic Seas). T. hyalina is described as being slightly larger (591-864 um) than Chilean specimens. though the head shape is similar (Kieneke & Nikoukar, 2017). The description of T. hyalina identifies 7-12 TbA, up to 57 TbL (TbDL + TbL + TbVL) and 6-11 TbP. The TbP are often present in asymmetrical numbers. The length and patterns of the TbA and TbP in the European populations correspond to those observed in the Chilean specimens. In addition to the size differences, there are also slight differences in the number of adhesive tubes between the Chilean specimens and T. hyalina. Furthermore, pigmented eyes are not present in T. hyalina. Taken together, this suggests that the Chilean species is not T. hyalina, though it is probably a closely related species.

Xenotrichula sp.

The genus currently consists of 15 described species (Kieneke & Schmidt-Rhaesa, 2015). Within the Pacific, some unidentified specimens have been recorded from the Galapagos (Schmidt, 1974).

CONCLUSIONS

This preliminary investigation of the gastrotrich fauna of the coast of Chile has revealed at least nine working species of Macrodasyida and one species of Chaetonotida. This low diversity is undoubtedly due to the small number of samples examined, but also likely to be a result of the technical demands of taxonomic identification, which generally requires high-resolution DIC microscopy. Samples collected in other meiofauna projects (unpublished) indicate that the Gastrotricha are abundant and diverse along the length of the Chilean coast and that it is only the lack of taxonomic expertise that has prevented a better understanding of the group. We hope that this first look at the group will stimulate further investigations of the Gastrotricha in Chile.

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