





RESEARCH ARTICLE

Ypsilothuria bitentaculata bitentaculata (Echinodermata: Holothuroidea) from the southwestern Atlantic, with comments on its morphology

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ABSTRACT. Ypsilothuria bitentaculata bitentaculata (Ludwig, 1893), previously known from several localities in the Pacific Ocean, is recorded herein for the first time from the southwestern Atlantic Ocean based on eight specimens caught off the coast of southeastern Brazil, between 505–511 m deep. Several morphological details are added to the description of Y. b. bitentaculata, including photographs of specimens and calcareous ring plates, as well as scanning electron microscope images of the ossicles from the body wall, oral and anal siphons and introvert. Additionally, Y. b. bitentaculata is compared to its congeners.

KEY WORDS. Sea cucumber, Dendrochirotida, Ypsilothuriidae, Brazil, deep-sea, REVIZEE.

INTRODUCTION

Historically, the southwestern Atlantic deep-water benthic fauna has been poorly sampled and poorly understood (Manning et al. 1989, Tavares 1999). In recent years, however, there has been a proliferation of monitoring oceanographic cruises carried out by oil and gas companies as well as survey expeditions conducted by research institutions and the deep-sea fishing industry. As a result, knowledge of Brazil's deep-water benthic species, although very fragmentary, has been gradually increased.

Likewise, only recently more progress has been made on the discovery of deep-sea holothuroids (e.g. Moura et al. 2010, Campos et al. 2010, Moura et al. 2015).

Among the recent efforts undertaken to explore Brazil's deep-sea benthic habitats are a number of deep-water oceanographic investigations conducted in southeastern Brazil as part of the REVIZEE Program (Living Resources in the Exclusive Economic Zone) (Amaral and Rossi-Wongtschowski 2004). The REVIZEE cruises in southeastern Brazil yielded eight specimens of *Ypsilothuria bitentaculata bitentaculata* (Ludwig, 1893) (Ypsilothuriidae), a species new to the southwestern Atlantic fauna.

As currently understood, the genus *Ypsilothuria* comprises two species: *Y. bitentaculata* (Ludwig, 1893) and *Y. talismani* Perrier, 1886. These two species were questionably divided into five subspecies (Heding 1942, Paulay and Hansson 2013): *Y. b. bitentaculata* (Ludwig, 1893) [Pacific, South Africa and southwestern Atlantic – present record], *Y. b. attenuata* E. Perrier, 1886 [Davis

strait to Senegal], *Y. b. virginiensis* Heding, 1942 [West Indies] and *Y. talismani talismani* Perrier, 1886 [Atlantic coasts of France, Spain, Morocco, Senegal and also the Caribbean Sea], and *Y. talismani elegans* Heding, 1942 [West Indies]. The recognition of two subspecies by Heding (1942) within *Ypsilothuria bitentaculata* Ludwig, 1893 (namely *Y. bitentaculata attenuata* E. Perrier, 1886 and *Y. bitentaculata virginiensis* Heding, 1942) actually compulsorily creates three subspecies (ICZN 1999: Art.47.1), with the nominotypical subspecies *Ypsilothuria bitentaculata bitentaculata* Ludwig, 1893 being the third one (regardless of the name of the nominotypical subspecies not appearing in Heding 1942).

Here we present a detailed morphological description of the poorly known *Y. b. bitentaculata*, including color photographs of full specimens and of the calcareous ring plates, as well as scanning electron microscope images of the ossicles from the body wall, oral and anal siphons and introvert. Additionally, *Y. b. bitentaculata* is compared to its congeners.

MATERIAL AND METHODS

Specimens of *Ypsilothuria bitentaculata bitentaculata* were collected from infaunal assemblages with the aid of a Van Veen grab and a rectangular dredge, sieved onboard through a 1 mm mesh. Measurements were obtained from ethanol fixed specimens and correspond to the maximum lengths of the smallest and the largest specimens, respectively. Holothuroid morphological techniques and terminology followed Rowe and



Doty (1977) and Samyn et al. (2006); the taxonomic classification followed Miller et al. (2017). Ossicles were sampled from different body regions (i.e. introvert, tentacles, body wall, oral and anal regions) using household bleach, washed five times in distilled water and then, five times in absolute ethanol. A number of ossicle samples were mounted on slides with Entellan Merck for permanent storage and then examined and measured under optical microscope. Other ossicle samples were dried and mounted on metal stubs with double-sided tape, coated with gold and observed with a LEO 440 Scanning Electron Microscope (SEM).

Abbreviations and acronyms: specimen (spm); Museu de Zoologia, Universidade de São Paulo (MZUSP); Muséum national d'Histoire naturelle, Paris (MNHN); National Museum of Natural History, Smithsonian Institution, Washington D.C. (USNM).

TAXONOMY

Ypsilothuriidae Heding, 1942 Ypsilothuria E. Perrier, 1886

Ypsilothuria bitentaculata bitentaculata (Ludwig, 1893)

Figs 1-19

Sphaerothuria bitentaculata Ludwig, 1893:141, pl. XII, figs 16–17, pl. XIV, figs 5–14.

Sphaerothuria talismani – Deichmann 1930: 154, pl. 9, fig. 3 [not Ypsilothuria talismani Perrier, 1886]

Ypsilothuria bitentaculata – Panning 1949: 455, Pawson 1965: 6–7, text fig. 1, figs 1–5; Thandar 1999: 373–375, fig. 3 d–f; Massin and Hendrix 2011: 422, fig. 7; Mecho et al. 2014: 294, 295, fig. 13.

Diagnosis. Body strongly U-shapped. Tentacles crown of eight digitiform tentacles, the two lateral ones longest. Calcareous ring simple, radial and interradial plates deeply notched, without posterior projections. Plates of body wall forming a test, arched rods in tentacles. Knobbed plates in introvert.

Description. Body strongly U-shaped (Fig. 1), length 5–10mm. Tentacles crown of eight digitiform tentacles, the two lateral ones longest (Fig. 2). Oral and anal siphons short, opposite to each other, oral siphon wider than anal (Fig. 3). Body wall thorny due to presence of intricate tables, visible with naked eye (Fig. 4). Calcareous ring simple. Radial and interradial plates with curved base, without projections on the posterior surface, bifurcated anteriorly, the radial slightly higher than the interradial (Figs 5, 18).

Body wall tables with circular disc perforated, irregular in outline. Each disc consisting of a laminar stereom with holes ranging between 20 and 50 μ m all over the disc plate (plate size between 500 and 1400 μ m; Figs 6–7). High spire strong (400–500 μ m; Fig. 8), placed near edge of disc. Tentacles with arched rods (80–150 μ m, Fig. 9), with one central perforation, one central apophysis and perforated edge (2–4 holes). Introvert

with knobbed perforated plates, elongated, irregular in outline (160–180 μ m, Figs 10, 11). Plates of base of oral siphon with undeveloped spires (400–800 μ m, Fig. 15). Oral and anal siphons with similar rods, anal rods smaller, strongly arched (40–70 μ m, Figs 12–14). Anal siphon with irregularly perforated plates (300–600 μ m, Fig. 16).

Material examined. MZUSP 1306, Brazil, REVIZEE, off coast of São Paulo, (25°43'S, 45°09'W), 13/Jan./1998, 511m, 6 spms. Ibidem, (24°20'S, 43°46'W), 9/Jan./1998, 505m, 2 spms.

Comparative material examined. MNHN-IE-2005 6332, Slides of the type [body wall tables] Pacific, (6°35′S, 81°44 W), 24/Feb./1891, 782m. MNHN 3693, Pacific, (34°6′S, 119°8′W), 1301m: 20 mm long [all originally labeled *S. bitentaculata* Ludwig, 1893]. USNM 15689, United States, northwestern Atlantic, Virginia, RV Albatross, stn 2723, (36°46′N, 73°09′W), 23/Oct./1886, 3082m: 10 spms 3–10 mm long [originally labeled *Y. bitentaculata* (Ludwig, 1893)].

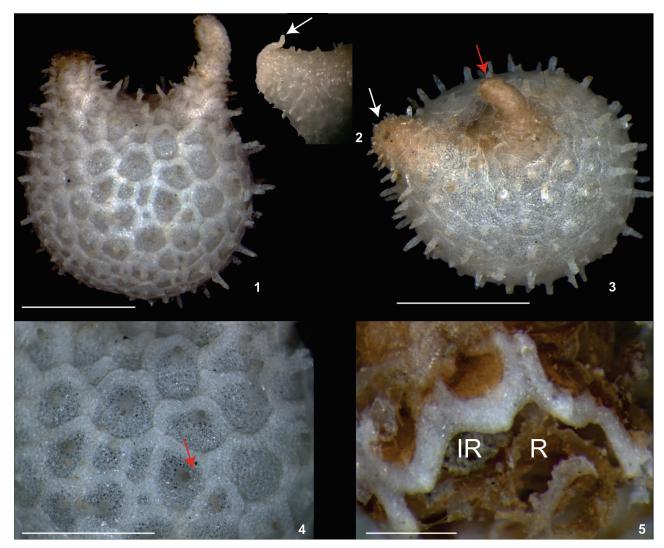
Distribution. *Ypsilothuria b. bitentaculata* has been regarded as a Pacific species (Oshima 1915, Pawson 1965, Gage et al. 1985). This is the first record of *Y. b. bitentaculata* from the southwestern Atlantic. According to Thandar (1999) *Y. b. bitentaculata* (as *Y. bitentaculata*) occurs in green mud sediments and between 225 and 4440 m (Cherbonnier and Feral 1978). Off the coast of São Paulo, *Y. b. bitentaculata* was found in silty sand bottoms around 500 m deep, within the previously known bathymetric distribution of the species. In southwestern Brazil *Y. bitentaculata bitentaculata* is so far lower bathyal in distribution possibly owing to the influence of the colder waters from the South Atlantic Central Water (SACW) over the southwestern Brazilian shelf. It is not uncommon for deep-water species to occur in depths shallower than usual owing to colder upwelling waters (Kowalewski et al. 2002).

Remarks. Heding (1942: 26) split *Ypsilothuria talismani* E. Perrier, 1886, *Y. bitentaculata* (Ludwig, 1893) and *Y. attenuata* E. Perrier, 1886 into two subspecies complexes, in which he included two additional subspecies: *Y. talismani talismani* E. Perrier, 1886, *Y. talismani elegans* Heding, 1942, *Y. bitentaculata bitentaculata* (Ludwig, 1893), *Y. bitentaculata attenuata* E. Perrier, 1886, and *Y. bitentaculata virginiensis* Heding, 1942.

Some authors have suggested that the morphological differences between the subspecies of *Ypsilothuria* are due to ontogeny only (Gage et al. 1985, Billet 1988). The shape of the calcareous ring, for example, changes with growth and the notches in the calcareous ring may not always be present, especially in juveniles. However, in the four specimens dissected for this study (maximal length: 6.0 mm; 7.9 mm; 8.2 mm and 10.0 mm), the shape of the calcareous ring was constant and the notches in the calcareous ring were always present.

Recent observations are not consistent with Billet's (1998) claim that the average size of the dermal plates varies with the size of the specimens. Specimens from the Indian Ocean with about 20 mm had two-spired dermal plates ranging between 1500 and 3000 µm (Samyn and Van den Spiegel 2016), whereas





Figures 1–5. *Ypsilothuria bitentaculata bitentaculata* (MZUSP 1306): (1) lateral view of the body; (2) detail of one of the two elongated lateral tentacles detail of the tentacle (white arrow); (3) dorsal view of a specimen preserved in ethanol (note oral and anal siphons, white and red arrows, respectively); (4) detail of the ossicles from the body wall (the red arrow indicates the spire); (5) calcareous ring. R, radial plate. IR, interradial plate. Scale bars: 1-3 = 5 mm, 4 = 1 mm, 5 = 0.5 mm.

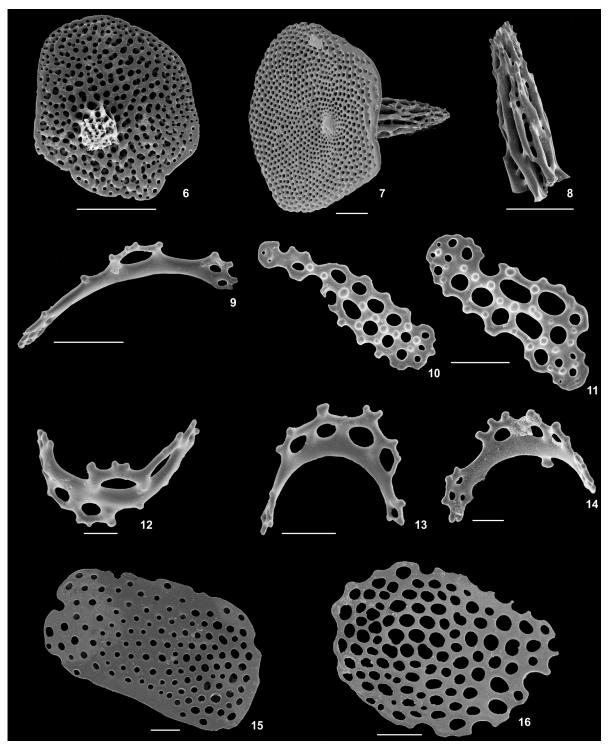
in the southwestern Atlantic a half-sized specimen (10 mm) had single-spired dermal plates ranging from 500 to 1400 μ m. Contrary, specimens from South Africa, although more than twice as large as the southwestern Atlantic specimens (25 mm), had single-spired, unexpectedly small dermal plates from 700 to 1270 μ m (Thandar 1999).

Several authors (e.g. Ludwig 1893, Deichmann 1930, Heding 1942, Pawson 1965, Massin 1996) have referred to the shape of the plates at the base of the tentacles to differentiate between *Y. t. talismani* and *Y. b. bitentaculata*. However, as noted by Thandar (1999) and in agreement with our results, the knobbed plates are in fact present in the introvert (not in the

tentacles). The plates of the introvert cannot be found neither in the type specimen of Y. bitentaculata (MNHN 6332) – since all that remains of it is a slide with the body wall plates –, nor in the in the specimen MNHN 3693 which is incomplete. In the specimen MNHN 3693 (20 mm) the size of the anal plates ranged from 400 to 600 μ m, whereas in the Brazilian specimens (5–10mm) the anal plates ranged from 300–600 μ m. The northwestern Atlantic (USNM 15689; 3–10 mm) and the southwestern Atlantic specimens from Brazil agree well with each other.

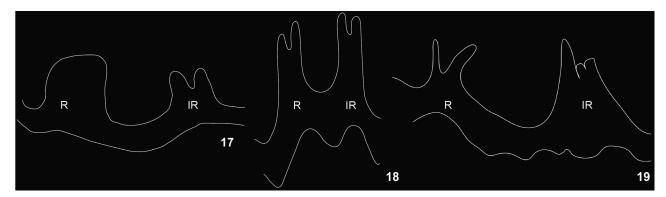
Ypsilothuria t. talismani actually differs from *Y. b. bitentaculata* in the morphology of the lateral interradial plates of the calcareous ring, whose anterior ends are entire in *Y. t. talismani*





Figures 6–16. SEM photomicrographs of the ossicles from *Ypsilothuria bitentaculata bitentaculata* (MZUSP 1306). (6–7) tables from body wall in dorsal and lateral views, respectively; (8) detail of the spire; (9) rod from tentacles; (10–11) knobbed plates from introvert (12–14); arched rods from anal and oral siphon. (15–16) plates from oral and anal siphons, respectively. Scale bars: $6 = 500 \, \mu m$, $7-8 = 200 \, \mu m$, $9-11 = 50 \, \mu m$, $12-16 = 20 \, \mu m$.





Figures 17–19. Outlines of the calcareous rings: (17) Ypsilothuria bitentaculata attenuata (after Heding, 1942); (18) Ypsilothuria bitentaculata bitentaculata (MZUSP 1306); (19) Ypsilothuria bitentaculata virginiensis (after Heding, 1942). Note in 18 radials and interradials narrow and the anterior end of both radials and interradials deeply notched. R, radial plate. IR, interradial plate.

and deeply notched in *Y. b. bitentaculata*. Additionally, the tentacle rods are straight and have few perforations at the ends in *Y. t. talismani* (viz. Heding 1942), whereas the rods are arched and have one central perforation and one central apophysis in *Y. b. bitentaculata*. Of noteworthy is that Billett's (1988: fig. 16B) illustration shows curved tentacle rods in *Y. talismani* similar to *Y. bitentaculata bitentaculata* (Fig. 9).

Ypsilothuria b. bitentaculata can be separated from Y. b. attenuata sensu Heding (1942) from the Northern Atlantic in having: (i) the calcareous ring with narrow radial and interradial plates (versus broad radial and interradial plates in Y. b. attenuata), (ii) the anterior end of the radial and interradial plates deeply notched in the radial and interradial plates (Figs 5, 17, 18) (versus notched only in the interradial plates in Y. b. attenuata) and (iii) arched tentacle rods (versus irregular tentacle rods in Y. b. attenuata).

However, the differences between *Y. b. bitentaculata* and *Y. b. attenuata* sensu Billett (1988) are subtler. In *Y. b. attenuata* from South Africa both the radial and interradial plates of the calcareous ring are narrow and notched just as in the southwestern Atlantic specimens of *Y. b. bitentaculata* (Figs 5, 18). In the southwestern Atlantic, however, the size of plate holes range between 20 to 50 µm, compared to 20 to 30 µm in South Africa. Additionally, in *Y. b. bitentaculata* the tentacles rods are provided with one central perforation and one central apophysis (Fig. 9), whilst in the South African *Y. b. attenuata* the tentacle rods are perforated near the end only and lack the central apophysis. In that respect, Billet's and Heding's *Y. b. attenuata* are similar to each other (Heding 1942: fig. 30: 1, 2; Billet 1988: fig. 20B).

Ypsilothuria b. bitentaculata and Y. b. virginiensis share similar shape of the calcareous ring (Figs 18, 19), but can be distinguished by slight differences in the morphology of the tentacle rods, which are arched and provided with one central perforation and one central apophysis in Y. b. bitentaculata versus arched and perforated only at the ends in Y. b. virginiensis.

The specimens from the southwestern Atlantic (São Paulo, Brazil) are herein assigned to *Y. b. bitentaculata* on the account of

the morphology of their tentacle rods and the size of the holes on their body wall plates.

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