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# FIXATION AND DESCRIPTION OF A NEOTYPE FOR *POLYCYCLUS RENIERII* LAMARCK, 1815 (TUNICATA, ASCIDIACEA, STYELIDAE, BOTRYLLINAE)[1]

**Riassunto.** *Designazione e descrizione di un neotipo per* Polycyclus renierii *Lamarck, 1815 (Tunicata, Ascidiacea, Styelidae, Botryllinae).*
Nel 1793 S.A. Renier descrisse una colonia, pescata al largo di Chioggia, che egli ritenne essere *Botryllus stellatus* (jr. syn. di *B. schlosseri*). Nel 1815 la specie di Renier fu riconosciuta da Lamarck essere una nuova specie (*Polycyclus renierii*) per la quale fu creato il nuovo genere *Polycyclus,* caratterizzato da un notevole spessore della colonia. In seguito genere e specie furono considerati sinonimo di *B. schlosseri*. Sulla base di nuovo materiale raccolto nella stessa zona della colonia di Renier, è stato possibile procedere ad una ridescrizione della specie che viene considerata diversa dal *B. schlosseri*, e, in assenza di materiale tipico, viene proposta la fissazione di un neotipo che viene depositato presso il Museo di Storia Naturale di Venezia.

**Summary.** The name *Polycyclus renierii* was created in 1815 by Lamarck for a species previously described by S.A. Renier (1793), based on a colony collected in the North Adriatic off Chioggia. The species was subsequently synonymised with *Botryllus schlosseri*. Based on abundant materials collected in the same geographical area, the species is recognised here as valid and a new description is given. As no type-material is available, a neotype is designated here – the specimen being deposited in the collections of the Museum of Natural History of Venice.

**Keywords:** taxonomy, ascidians, *Botryllus renierii*, *Polycyclus*, neotype, Adriatic, Mediterranean.

## Introduction

In 1793 Stefano Andrea Renier, professor of natural history at the University of Padova, described an interesting Botryllid species collected on the sandy bottom off the Venetian coasts, which he believed to be *Botryllus stellatus* Gaertner, 1774 (<*Alcyonium schlosseri* Pallas, 1766) (figs. 1a and c). In 1815, Lamarck recognized Renier's species as different from *B. schlosseri* and created for it the new genus and species *Polycyclus renierii.* The peculiar feature of the new genus was the thickness of the colony: a few centimetres in *Polycyclus* versus a few millimetres in *Botryllus*. The new genus was not recognized by either Savigny (1816) or Giard (1872) but was accepted by Della Valle (1877), Drasche (1883), Herdman (1886) and Lahille (1890) who, however, treated *Polycyclus* as a sub-genus of *Botryllus*. In the following century, the genus was recognized by Salfi (1932) whereas Harant & Vernières (1933) first considered it as a junior synonym of *Botryllus*, with *P. renierii* as a junior synonym of *B. schlosseri*. Finally, both genus and species disappeared from the literature.

In the last few years, I had the opportunity to study some colonies collected off Chioggia (southern Venetian coast), the same locality where the colonies studied by Renier were collected. Comparison with *B. schlosseri* showed that the two species are not the same (table 1). Therefore, a new description of the species is given and, as the type material for *Polycy-*

[1] This work is dedicated to the memory of the late Professor Charley Lambert, who passed away on 1.VI.2011.

*clus renierii* cannot be traced, and very likely was never preserved, a neotype, deposited at the Museum of Natural History of Venice, is proposed here.

***Botryllus renierii* (Lamarck, 1815)**

Citations and synonyms
*Botryllus stellatus*: Renier, 1793: 256.
*Polycyclus renierii* Lamarck, 1815: 340.
*Polycyclus elongatus* Delle Chiaie, 1841: 19 (synonymised by Della Valle, 1877).
*Botryllus baeri* Grube, 1864: 64 (synonymised by Della Valle, 1877).
*Polycyclus renierii*: Della Valle, 1877: 22; Lahille, 1890: 320; Salfi, 1932: 339.
*Botryllus (Polycyclus) renierii*: Drasche von, 1883: 13.
*Botryllus (Polycyclus) cyaneus* Drasche von, 1883: 14 (synonymised by Salfi, 1932).
*Botryllus (Polycyclus) violaceus* Drasche von, 1883: 14 (synonymised by Salfi, 1932).
*Polycyclus vallii* Lahille, 1890: 322 (synonymised by Salfi, 1932).
**Not** *Botryllus polycyclus* Savigny: 1816: 202 (probably *B. schlosseri*).

Type locality. The original type locality (off the Venetian coast, Northern Adriatic, Italy) is restricted here as follows, with the fixation of a neotype: 13 miles off Chioggia, 45°20'15''N, 12°43'30''E, sandy bottom, 23 m.

Material examined.

Neotype. MSNVE-22045: half of a globular colony of about 9 cm in diameter, approaching the change of generation: testis and fully developed larvae in filtering zooid, testis and eggs in buds, colour in life not recorded; 8.V.1996; 13 miles off Chioggia, 45°20'15"N, 12°43'30"E, sandy bottom, depth 23 m.

Further material examined. MSNVE-22046: half of a globular colony of about 9 ? 8 ? 4 cm, shortly after the change of generation (the regressing previous filtering zooids are still recognizable), colour in life not recorded, testis and eggs in filtering zooids and in buds; 25.V.1993; 8 miles off Lignano, sandy bottom, depth 15 m. MSNVE-22047: half of a globular colony of about 10 ? 9 ? 5 cm, dark brown, with oral siphons lighter, in life, large testis in filtering zooids, testis and eggs in buds; 21.V.1993; 8 miles off Lignano, sandy bottom, depth 15 m. MSNVE-22048: globular colony of about 4 cm in diameter, violet in life, gonads absent in filtering zooids, testis in buds; 25.II.1994; 8 miles off Lignano, sandy bottom, depth 15 m. Several colonies from the same area, as well as three colonies from Banyuls-sur-Mer (France) collected by Françoise Lafargue, were examined too: a piece of colony, 28.IV.1975, Rederis, sandy bottom, depth about 20 m; two colonies, 21.VI.1982, detritic bottom off Banyuls, depth about 60 m.

Distribution. Apart from the Northern Adriatic, the species was previously recorded in the Southern Tyrrhenian (Naples - Delle Chiaie, 1841; Della Valle, 1877; Salfi, 1932) and North-Western Mediterranean (Banyuls-sur Mer: Lahille, 1890).



Description

The colony

Colonies are characterised by great thickness of the matrix. On sandy bottoms, they grow on small portions of hard substrate, usually fragments of shell or small residues of marine plants. In these cases, the colony completely covers the substrate, which is almost completely engulfed, and continues to grow, forming a globe-shaped or ovoidal body several centimetres in diameter (figs. 1a-b); the largest size recorded was 11 ? 8 ? 7 cm. When hard substrates are abundant, the colonies form fleshy, thick crusts or cushions. They present a variety of colours, as occurs in *B. schlosseri*. The species lives at depth exceeding 15-20 m.

Zooids, up to 5 mm long, are arranged in star-shaped or ovoidal *schlosseri* type systems (Brunetti, 2009) (figs. 1a-b), perpendicular to the surface of the colony. The surface is tough and does not agglutinate sand or other foreign materials. The colonial matrix, also free of foreign particles, is fleshy, transparent, and crossed by the vascular network connecting the zooids (figs. 1c-d). The matrix is tough too, making the extraction of zooids, and especially of the buds, very difficult. Marginal vascular ampullae are not present in this type of colony and spherical ampullae are abundantly scattered along the matrix vascular network and on the colony surface, among the systems.

The zooid

The zooid is cylindrical in shape, with a smooth-edged oral siphon and the atrial cone with a dorsal tongue, as usual in the genus *Botryllus* (fig. 2a). The body wall is run by muscles in an apparently chaotic arrangement (fig. 2d), only at the apertures do they form a circular band. Tentacles are greatly reduced or almost absent, except for two lateral, very large tooth-shaped ones, set slightly towards the ventral side. At their base, a large vascular lacuna is usually present, filled with blood cells (figs. 2b-c). These cellular masses are also visible on the external surface, as two spots on both sides of the oral aperture. A median longitudinal blood vessel runs along the inferior surface of them. When recognizable, tentacles number 7, an odd instead of an even number, as usual in Botryllinae. There is also a small mass of blood cells at the base of the undeveloped tentacles. All tentacles are arranged in a single ring (fig. 2b).

The branchial sac has about 15 rows of stigmata, the second row does not reach the dorsal line. There are also some other irregular stigmata at the posterior end, sometimes forming a sort of net of rounded stigmata. The pre-pharyngeal groove does not form a V at the beginning of the dorsal vessel. The dorsal tubercle has a simple circular-elliptical opening at the centre of a well defined area; the neural complex is at a short distance from it, situated at the level of the first row of stigmata. The three longitudinal vessels originate at the level of the first row of stigmata and divide the branchial wall into sectors of almost equal width (fig. 2b). The stigmata are usually arranged as follows (data from the middle of the branchial sac of two colonies): 7,6,6,9 DL 9,6,6,6 and 9,8,7,9 DL 9,7,8,9. The dorsal lamina rises from the dorsal vessel at the level of the fifth and sixth rows of stigmata. It is initially simple, then it doubles reaching its highest point before disappearing to the left of the oesophageal opening (fig. 2b). In no filtering zooids were masses of haematic cells observed on either side of the endostyle, contrary to what happens usually in recently formed systems of the Botryllinae (see

Brunetti, 2009, where the possible significance of such cells is discussed). No muscles were evident in the branchial wall.

The stomach is lateral and partially below the branchial sac, as usual in the genus (fig. 2a). It is cylindrical, with spiral folds (figs. 3a-d). The latter are 9 in number, not swollen at the cardiac end, longer on the branchial surface, with a longitudinal median groove. The ninth is much shorter than the others. The typhlosolis does not extend over the pyloric end of the stomach. The caecum, rising from the posterior half of the typhlosolis, is half the length of the stomach, and is tubular with a short tip, bent and not swollen. The duct of the pyloric gland is connected to the caecum in its third terminal tract. The intestinal loop is large and its external epithelium has cells regularly arranged in parallel transversal lines (fig. 2e). A smooth-edged anus opens about two to four rows of stigmata above the anterior edge of the intestinal loop. The surface of the intestinal loop, like that of the oesophagus, presents wrinkles (fig. 3a) but, although they have often been observed, it is not clear whether such wrinkles are artefacts due to fixation.

Gonads. The testis is fan-shaped with a number of follicles depending on the degree of maturity of the zooid. In well-developed zooids it has usually about 10 follicles. The eggs are arranged dorsally in an arc around the testis (fig. 2a). There are up to 6 eggs per side in buds at the end of their development.

The larva

At the time it leaves the maternal body, the larva is about 1.7 mm long (excluding the fin) of which 0.5 mm is represented by the trunk.

**Table 1.** Comparison between *B. schlosseri* and *B. renierii*.

| | *B. schlosseri* | *B. renierii* |
|---|---|---|
| Zooid length | up to 2 mm | up to 5 mm |
| Zooid shape | conic | cylindrical |
| Zooid posture | tilted | vertical |
| No. of developed tentacles | usually 8 | usually 2 |
| No. of rows of stigmata | about 7 | about 15 |
| Stomach shape | pear shaped | cylindrical |
| No. of stomach folds | 7 | 9 |
| Arrangement of stomach folds | longitudinal | spiral |
| Shape of pyloric caecum | not or only slightly bent | bent |
| Tip of pyloric caecum | swollen | not or only slightly swollen |



Remarks

The present species clearly differs from the littoral *Botryllus schlosseri*, apart from size, in several morphological characters such as tentacles, branchial sac and intestinal loop (table 1). However, its peculiarity is its capacity to produce and support a large quantity of colonial matrix. This peculiar feature, which drove Lamarck to create the new genus *Polycyclus*, allows the species to colonize environments lacking hard substrates such as sub-littoral sandy-muddy bottoms, which is a very rare characteristic among the ascidians. In fact, growing as a spherical body, the colonies can arrange many zooids on a surface not attached to a hard substrate. The latter is necessary only for the settlement of the larva and it may be very small.

There is disagreement about the source of the matrix and its external surface or cuticle in Botryllinae. Three possible origins have been suggested: the zooid epidermis (Zaniolo & Trentin, 1987), the endothelial cells of vascular ampullae (Torrence & Cloney, 1981) and blood cells (Smith, 1970). Probably, as suggested by Wardrop (1970), all these structures cooperate. However, one modality may predominate in a particular species. Thus, in the encrusting colonies of *B. schlosseri* (and other species with similar colonies) the matrix is probably mainly produced by the marginal vascular ampullae, while in *B. renierii* it may originate from the zooid epidermis and the endothelium of the test vascular net.

Only two other species have been described displaying such "*Polycyclus* character": *Polycyclus lamarcki* Herdman, 1886, collected in the North Atlantic at 664 m depth, and *Polycyclus jeffreysi* Herdman, 1886, collected in the Bay of Tangiers at 65 m. Herdman's accurate descriptions show that both species are different from *P. renierii*. However, all of them, in addition to presenting a very thick colonial matrix, are not shallow-water species. So, Herdman's statement (1886, p. 63) "The thickness is so considerable that there can be no doubt as to the propriety of separating this and similar forms from the genus *Botryllus*" in favour of the validity of the genus *Polycyclus* may be supported by ecological and probably also physiological characteristics. However, the general morphology of these species is so congruent with that of *Botryllus* that *Polycyclus* should still be considered a junior synonym of *Botryllus.*

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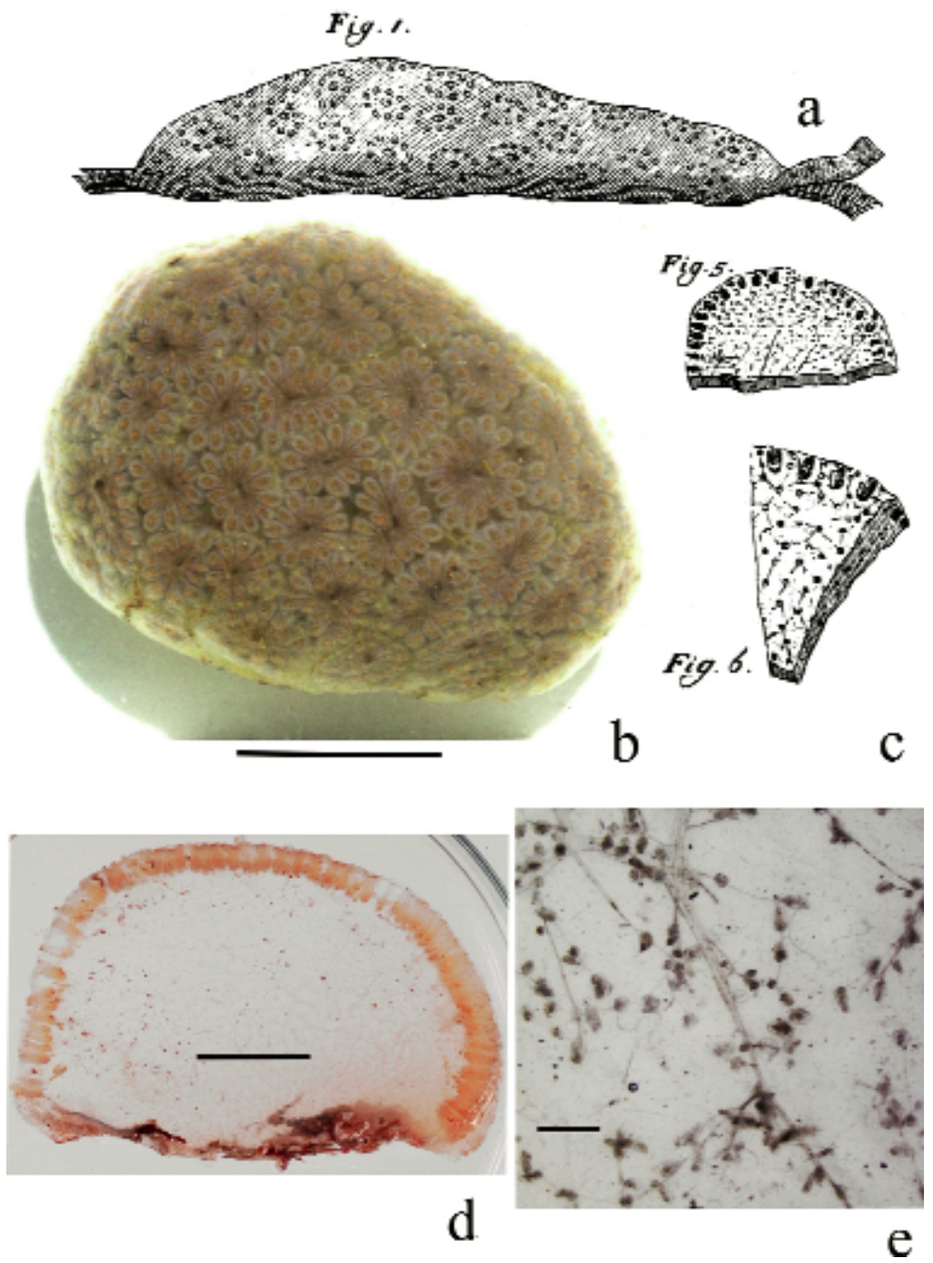


**Fig. 1. a** and **b:** colonies of *Botryllus renierii*; **c** and **d**: transverse sections of the same (a and c: original figures from Renier's letter, slightly reduced); **e**: blood vessels and ampullae into the matrix. Scales: b and d, 2 cm; e, 500 μm.

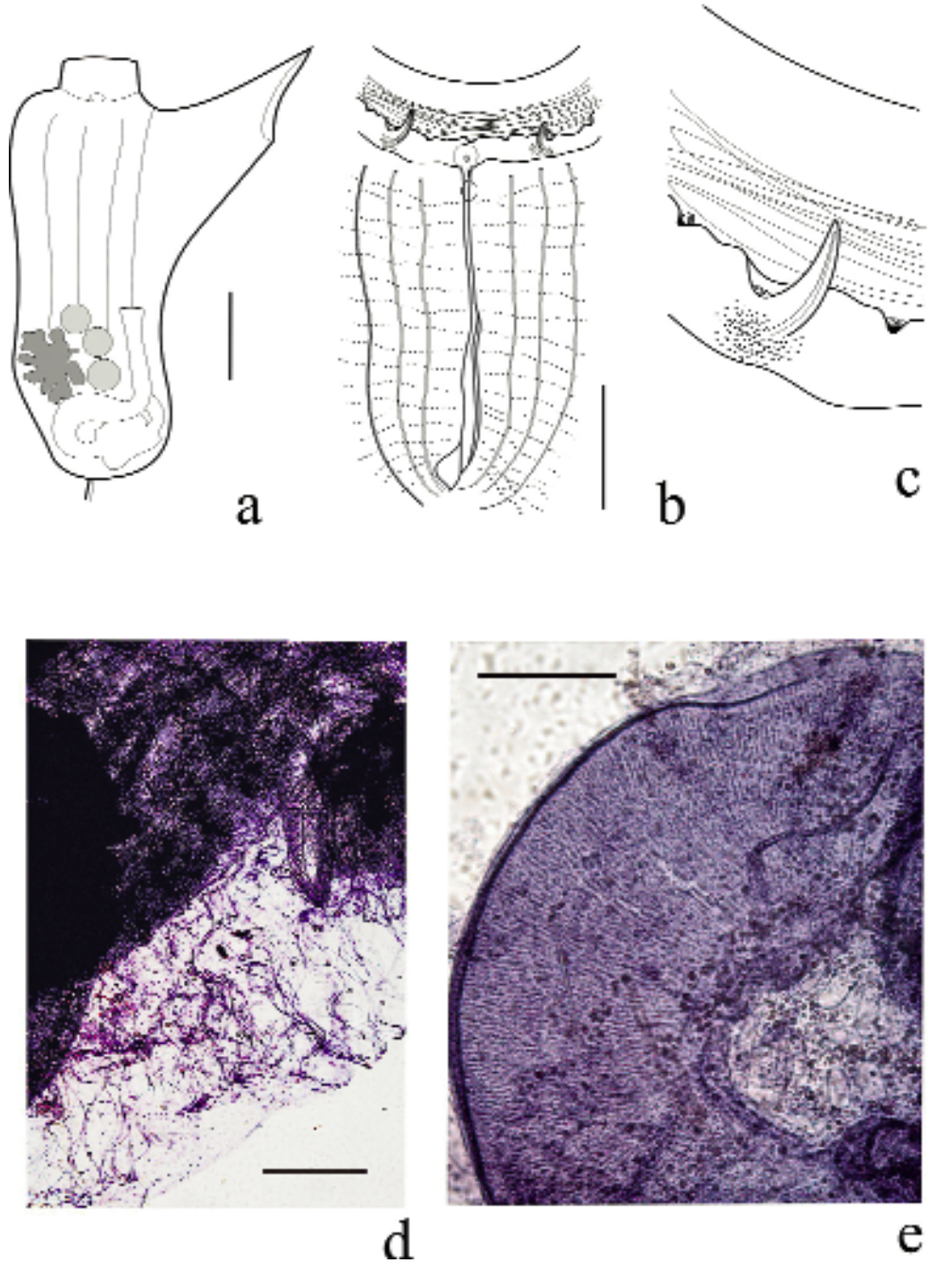


**Fig. 2. a:** zooid showing the arrangement of gonads; **b:** inner view of dissected zooid; **c:** tentacles: enlarged detail of fig. b; **d:** body wall: muscles; **e:** intestinal loop: epithelium. Scales: a and b, 1 mm; d and e, 500 µm.



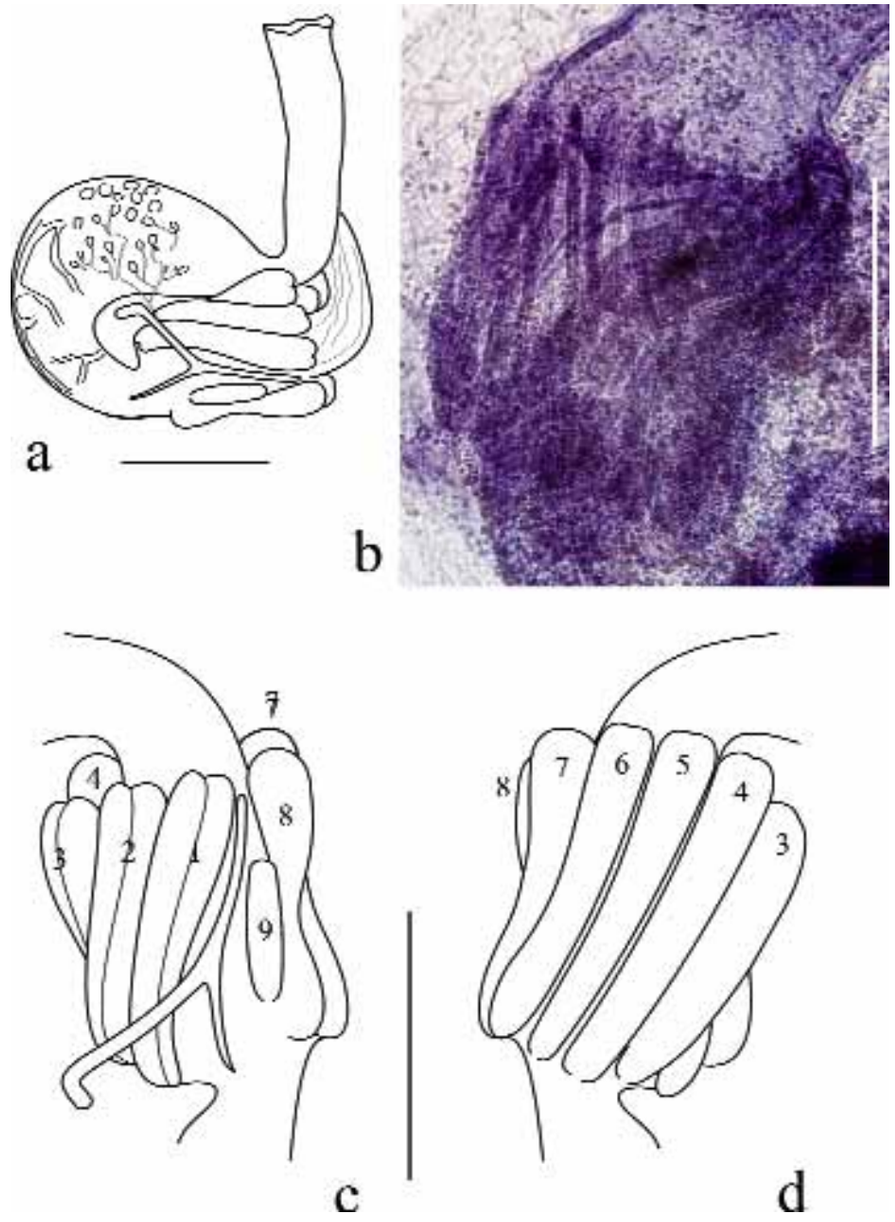


**Fig. 3. a:** gut loop (parietal side); **b**: stomach showing spiral arrangement of folds; **c** and **d**: stomach, parietal and mesial sides. Scales: 1 mm.