Morphological Ossicles of Sea Cucumber *Paracaudina australis* from Kenjeran Waters, Surabaya, Indonesia

Widianingsih Widianingsih1), Retno Hartati1), Muhammad Zainuri1), Sutrisno Anggoro2), Hermin Pancasakti Kusumaningrum3) Robertus Triaji Mahendrajaya1)

1) Department of Marine Sciences, Faculty of Fisheries and Marine Sciences Diponegoro University Jl. Prof. Soedharto, SH, Tembalang, Semarang
2) Department of Fisheries, Faculty of Fisheries and Marine Sciences, Diponegoro University. 3) Department of Biology, Faculty of Sciences and Mathematic, Diponegoro University

Corresponding author: widia2506@yahoo.com, widia2506@gmail.com

**Abstract.** This research aims to study morphological ossicles of sea cucumber *Paracaudina australis* which belongs to Ordo Molpadida and Family Caudinidae. Samples *P. australis* were taken randomly from the Kenjeran Waters, Surabaya. Samples *P. australis* were preserved by alcohol 96% for studying ossicles. According the research result, the types of ossicles in dorsal and ventral were dominated by button, rod and eyeglasses shape. In the dorsal tissue and ventral tissue, there are no rod shape bone. Calcarious ring of *P. australis* consists of 16 radial shapes and 12 interradial shapes. Ossicles shapes in the anterior tissue are more variety than in the posterior tissue. Its conclude that the eyeglasses shape and number 8 shape are dominant in all of tissue *P. australis*.

1. **Introduction**

Sea cucumber *Paracaudina australis* is one of many sea cucumber varieties found in the Kenjeran Waters, Surabaya or wellknown as “Blonyo”. This type of sea cucumber is widely exploited by fishermen because of the high demand and has high economic value and is traded as the main raw material to make trepang crackers [1] [2].

Based on observation, in the East Coast Region of Surabaya and the surrounding area, there are 7 sea cucumbers species, i.e. *Phylloporus* sp., *Paracaudina australis*, *Colochirus quadrangularis*, *Holothuria* sp., *H. sanctori*, *H. forskali* and *H. turricelsa* [1]. From the seven species, there are 2 species of *Phylloporus* sp and *P. australis*, which are mostly captured in the Kenjeran waters, Surabaya.

Sea cucumber *P. australis* belongs to Phylum Echinodermata [3] [4] [5]. Family Caudinidae has 15 tentacles, each with 2 pairs of digits, no unpaired terminal digit [6].

Phylum Echinodermata,

Class : Holothuroidea,

Ordo : Molpadida

Family: Caudinidae
Genus: Paracaudina
Species: Paracaudina australis

Trepang can be identified according to its morphological form [7] and DNA [8]. However, in various preserved sea cucumbers, it can often damage the morphological appearance so that sea cucumbers are difficult to identify. During the preservation process, the specimen often loses its external characteristics. Thus the preserved species depends very much on the characteristics of the ossicles on the body wall.

Based on the observations, ossicles have a composition which is very different and specific for each species. Therefore, ossicles can be used as a way to identify [5][7]. Research results in Cook Strait, New Zealand have successfully mapped several types of 235 species of Holothurian including genus Paracaudina [3]. According to the study the ossicles taken from the body walls of the middle sea cucumber, in New Zealand waters such as Paracaudina ambigua, P. australis, P. chilensis, P. bacillis, P. cortacea, P. cuprea, P. keablei, P. luticola, P. tetrapora and P. tripoda. [3][6].

This research aims to morphologically identify trepang by investigating characteristics of the ossicles in the species P. australis.

2. Research methods
Sea cucumber P. australis samples were taken from the Kenjeran Waters, Madura Strait, Surabaya randomly at several locations. In the study of morphology P. australis, the researchers conducted sea cucumber sampling and prepared the equipments such as sample bottles, cameras, dissecting kits, pipettes, object glass, compound microscopes, lucida cameras, ethanol and NaClO (compounds used for bleaching).

In observing the phenotype of sea cucumber P. australis morphological ossicles, we investigated the calcareus ring and ossicles from tentacle tissue, dorsal tissue, ventral tissue, anterior tissue and posterior tissue [5][9].

The stages that will be carried out observing the ossicles are: (a) Cut the tissue 1-5 mm2 for each part of the body which includes the dorsal, ventral, anterior, ventral and tentacles; (b) Place the piece of tissue on a 25 ml glass beaker then soak it with NaClO bleaching solution for 30-45 minutes, until the entire muscle tissue is destroyed.; (c) After that wash the ossicles sample with distilled water 4 - 7 times; (d) Then the ossicles sample is placed on a concave glass object, and after that the ossicle is measured; (e) The sample is ready to be observed under the compound microscope and then the picture is taken using the camera connected with microscope.

3. Results and Discussion
3.1. Ossicle Sea Cucumber
The body shape of P. australis is cylindrical with the posterior part having a small tail shape. The length of this biota can reach 230 mm in fresh conditions. This type of sea cucumber has no tube legs, with a smooth body. In fresh conditions, the sea cucumber body has a transparent pink colour and slightly ivory white. While in the anterior part, especially in the tentacles, the color is rather yellowish. Whereas in the form of preserved the P. australis is pale white and tends to grayish brown [5][6]. In the preserved body part, this sea cucumber loses yellow color, but in the anterior part it still looks pale yellow (Figure 1). According to [5] the P. australis from North-Eastern Australia has characteristic deeper mid body wall with irregular rods, less than 50 um long, colour never yellowish -red (rusty).

3.2. Calcareous Ring
Based on the observation of the shape of the calcareous ring, Paracaudina australis consists of 16 radial shapes and 12 interradial shapes (Figure 1). This is in accordance with the research, that there is an interradial shape with ivory white color between the radial shape [5][6].
3.3. Anterior
In the anterior tissue there are several ossicle shapes such as the shape of the glasses with a length of 6.24 ± 1.04 µm, peanut shape is of 5.52 ± 0.64 µm, length round shape is 6.69 ± 3.77 µm diameter, oval shape is 7.52 ± 2.04 µm length, number 8 shape is 7.90 ± 0.18 µm length and rod shape is 12.95 ± 2.55 cm length (Figure 2). In some references shapes peanut, round, oval and number 8 shape it is commonly referred to as the shape of "grain shape" while the shape of the rod is known as the form of "rod" [5] [6].

![Figure 2](image)

**Figure 2.** Ossicle shape in anterior tissue (a) eyeglasses shape; (b) nut shape; (c) round shape; (d) oval shape; (e) the shape of glasses; (f) rod shape.

3.4. Posterior
According to observations on the posterior tissue of the trepang body, there is a form of glass with an average length of 8.99 ± 1.14 µm, round shape with an average diameter of 3.70 ± 0.17 µm, the shape of figure 8 has a size the average length is 9.27 ± 1.88 µm and the rod shape is like a bone with a size of 9.04 ± 1.62 µm (Figure 3). In this posterior tissue there is no rod shape as in the anterior tissue.

![Figure 3](image)

**Figure 3.** Ossicle shape in posterior tissues (a) eyeglasses shape; (b) number 8 shape; (c) rod shaped bone; (d) round shape.

3.5. Dorsal
In dorsal tissue there is a form of eyeglasses with 7.89 ± 0.68 µm average length, number 8 shape with an average length of 7.01 ± 0.96 µm, pea nut shape with a length of 6.11 ± 0.87 µm and round shape with 4.82 ± 0.07 µm diameter (Figure 4). All of the ossicles shape in the dorsal tissue can be catagorized as grain shape [5] [6] [10].
3.6. Ventral
In the ventral body tissue, the shape of the glass is found a length of 6.02 ± 0.22 µm, the figure 5 has an average length of 7.06 ± 1.16 µm, round shape with an average diameter of 6.00 ± 0.89 µm and cylindrical shape with an average length of 6.31 ± 0.84 µm.

3.7. Tentacle
In the tentacle network there is a form of eye glass with an average length of 10.99 ± 1.24 µm, number 8 with an average length of 9.94 ± 1.57 µm and the appearance of irregular rod shapes with the average length is 10.60 ± 0.88 µm (Figure 6). The irregular rod shape in the tentacle tissue is one of the shape in Paracaudina sp. [5] [6]

4. Conclusion
The shapes of the ossicles found in all tissues observed are the rod shape, eyeglass shape and number 8 shape. The round shapes, such as nut and oval shapes can be categorized as grain shapes. The irregular rod shape is only found in tentacle tissue.
Acknowledgments. I would like to say thank you for all colleagues who helped in this research.

References
[1] Winarni, D., M. Affandi, E.D. Masithah, H. Damayanti. 2012. Proceeding of The International Fisheries Symposium IFS 2012, Can Tho City-Viet Nam, 6 – 8 December 2012.
[2] Widianingsih, M. Zainuri, S. Anggoro and H.P. Kusumaningrum. 2016. Aquatic Procedia 7: 271-276.
[3] Pawson, D.L. 1963. Zoologi Publication from Victoria University of Wellington.
[4] Pawson, D.L. 2007. Zootaxa, 1668: 749-764.
[5] O’Loughln, P.M., S. Barmos, D.V. Spiegel. 2011. Memoir of Museum Victoria 68: 37-65.
[6] Davey, N & M.O’Loughlin. 2013. Zootaxa 3613 (4): 357-368
[7] Massin, C. 1996. Zool. Verh. Leiden 307: 1-53.
[8] Sun, L., M. Chen, H. Yang, T. Wang, B. Liu, C. Shu & D.M. Gardiner. 2011. Comparative Biochemistry and Physiology Part D 6: 195-205.
[9] Samyn, Y.D. Vandenspiegel & C. Massin. 2006. Zootaxa 1138: 53-68
[10] Pawson, D.L. 1970. Mem. N.Z. Oceanogr. Inst. No. 52: 1-69.