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Research Article

Anatomical description of a tropical sea pen, *Virgularia gustaviana* (Herklots, 1863), from the Strait of Hormuz

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Abstract

The present investigation aimed to study the anatomical characteristics of *Virgularia gustaviana* (Herklots, 1863) from the Strait of Hormuz in the northwest of the Persian Gulf. In this regard, 25 sea pens, which were identified as *V. gustaviana* (Herklots, 1863), were collected from the intertidal zone of the Suru and *Bandar Abbas* coasts (the northeast of the Persian Gulf). The samples were fixed in the alcohol formalin glacial acetic (AFA) fixative for a week and were then studied using a stereomicroscope. According to the results, the body of *V. gustavia* anatomically consisted of two main parts (rachis and peduncle), both of which originate from a primary polyp. The rachis was the upper part of the body with leaf-like leaves. The peduncle, with a smooth, straightforward structure, was the lower part of the body. The caudal part of the peduncle was bubble-like. A mineralized axis was observed at the center along the body, surrounded by mesenteric septa. Although all sea pens share the same basic anatomical structure, colony shape varies considerably in different species in terms of evolution.

Keywords: *Virgularia gustaviana*; Pennatulaceans; Sea pens anatomy, Rachis

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Introduction

Sea pens or pennatulaceans (Phylum Cnidaria, class Anthozoa and Order Pennatulacea) are the most advanced octocorals due to the complexity of the colony, the specific function of the polyps, and the colony integrity (Brusca and Brusca, 2003). These animals are one of the most exciting non-moving megabenthos in marine environments at depths ranging from 15 to 200 m or more (Williams, 2011).

Unlike other octocorals, the colonial body of sea pens originates from a single large primary polyp anchored into substrate by a peduncle which in turn creates a rachis expanding to the distal end. Secondary polyps originate from the primary polyp. They are responsible for nutrition and reproduction (autozooids), and water intake (siphonozooids). Most of the sea pens have a calcified central rod, called an axis, surrounded by the internal sclerites that provide firmness (Antcliffe and Brasier, 2008).

Under optimal conditions, sea pens may form large aggregations called sea pen fields (Kenchington *et al.*, 2014) that provide biogenic structures for benthic invertebrates and fish, especially in sandy and muddy grounds with little physical habitat (Malecha *et al.,* 2005; Baillon *et al.,* 2014; De Clippele *et al.,* 2015). Therefore, these animals are considered important components of muddy and sandy marine ecosystems that are found in the deep sea as well as shallow waters (Williams, 2011). The muscular peduncle lets them stick to sandy beds, where fewer creatures can settle in these areas. Sea pens are able to move through their peduncles (Williams and Alderslade, 2011; Imahara *et al.,* 2014). Most of these animals feed on a variety of small plankton, such as invertebrates, rotifers, suspended detritus, planktonic bacteria, and possibly phytoplanktons (Borneman, 2001).

Sea pens show great variation in appearance and morphological changes, such as bilateral symmetry, decrease in the number and size of sclerites, density, and position of feeding polyps, which can be described as evolutionary events of different lineages (Williams, 2015). In this regard, the present study aimed to evaluate the anatomical characteristics of *Virgularia gustaviana* (Herklots, 1863) from the northwest of the Persian Gulf.

Materials and methods

25 sea pens identified as *V. gustaviana* (Herklots, 1863) in appearance (Fig. 1) were collected on July 2017 by patrolling on the intertidal zone of the Suru and *Bandar Abbas* beaches, situated at the Strait of Hormuz at the northeast of the Persian Gulf (Fig. 2) (Miatta and Snelgrove, 2022). First, biometric assays including measurement of the weight, length, and thickness of the animal body were conducted. The samples were then fixed in an alcohol-formalin-acid (AFA) solution (containing 80 ml of ethanol, 10 ml of formalin, and 5 ml of acetic acid) for a week and then moved to 70% ethanol (Miatta and Snelgrove, 2022). For anatomical analysis, samples were taken

from different parts of the animal body and studied using a stereomicroscope (Olympus- Japan) and digital images were taken using the Dino lite lens (with Dinocapture software, FDP2, New Taipei City, Taiwan) (García-Cárdenas and López-González, 2023).

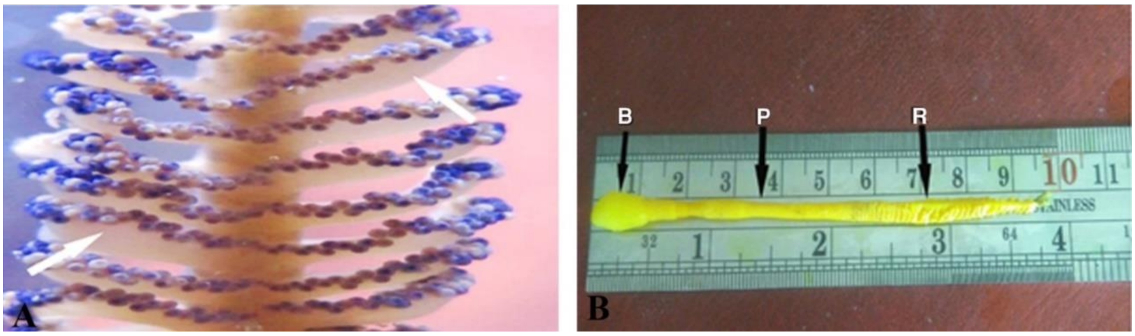

Figure 1: *Virgularia gustaviana* (Herklots, 1863): A. live sample, autozooids (white arrows); B. Fixed sample: R: rachis, P: peduncle, B: bulb; C. Schematic image of *V. gustaviana.*

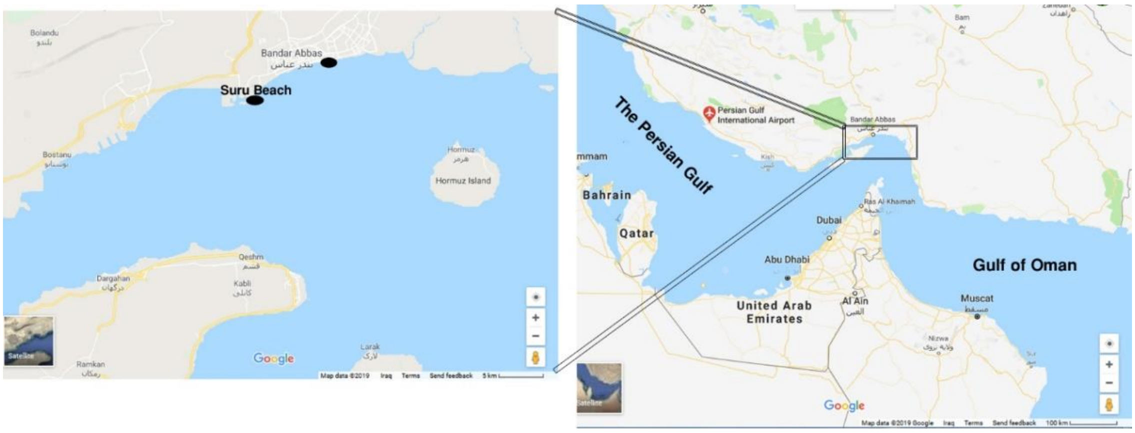

Figure 2: Sampling areas in the Suru and Bandar Abbas beaches (at the Strait of Hormuz at the northeast of the Persian Gulf).

Results

Identification of *V. gustaviana* was done using identification keys based on morphological characteristics and shape of spicules. Two types of spicules were detected in this species: 1. Three-edged spicules in autozooids and siphonozooids, and 2. Brass-like spicules in peduncle (Fig. 3). Spicules contain color pigments and therefore, the color of the colonies is affected by these colored pigments.

According to the results, the body of *V. gustavia* anatomicaly consisted of two main parts (rachis and peduncle), both of which originate from a primary polyp (Fig. 1B, C).

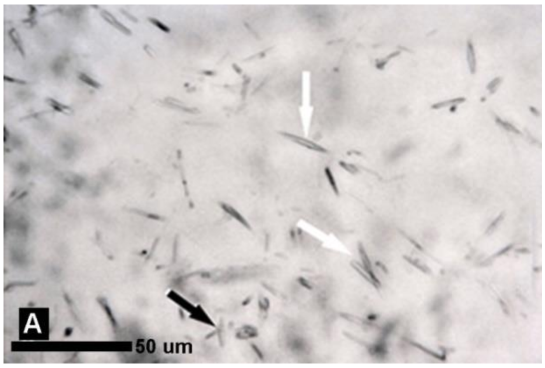

Figure 3: Two types of spicules were detected in *Virgularia gustaviana*. Three-edged spicules (Black arrow), brass-like spicules (white arrows).

A rachis with leaf-like leaves was the upper part of the body, extending from the primary bud to the tip of the polyp (Fig. 1B, C). The lower part of the body is called the peduncle with a smooth and straight structure with a bubble-like tail (Fig. 1B,C). Table 1 shows the biometrical characteristics of *V. gustavia*.

Table 1: The biometrical characteristics (mean±SE) of *Virgularia gustaviana* collected from the Suru and *Bandar Abbas* beaches (the northeast of the Persian Gulf).

Total length (cm)	Length of the rachis (cm)	Length of the peduncle (cm)	Rachis diameter (cm)	Peduncle diameter (cm)	Bulb diameter (cm)	Total weight (gr)
8.3±1.04	5.3±0.54	3.01±0.43	0.5±0.06	0.3±0.05	0.4±0.11	0.64±0.19

Rachis had two ventral and dorsal surfaces. Autozooids were leaf-like plates with contractile strength located on the ventral surface of the rachis (Fig. 4A, B). The plates were open in a relaxed state and collapsed and fell into the mud under stress.

Autozooids had hollow button-shaped prominences called tentacles, each of which had many finger-like bumps (usually more than 20) on the outer edges (Fig. 4B, C). The mouth opening (Fig. 4C) located in the center of the tentacles involved in the transport of food by autozooids into the polyp. The siphonozooids were visible as two non-contractile smooth bands on the dorsal surface of the rachis (Fig. 4A). They play a role in the transport and circulation of water.

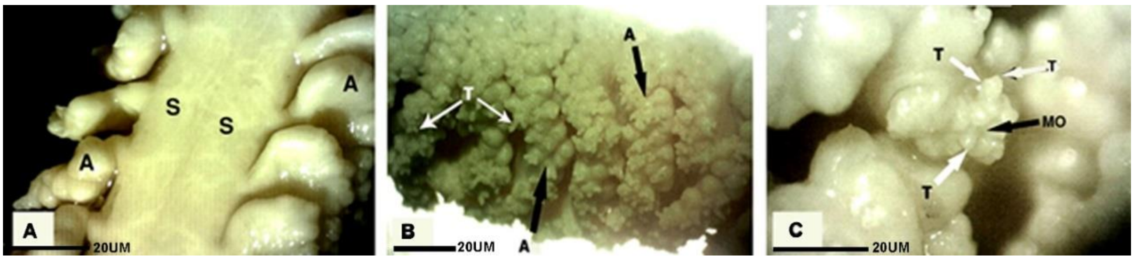

Figure 4: Stereomicroscopic images of rachis and peduncle in *Virgularia gustaviana*: Siphonozooids (S), Autozooids (A), Tentacles (T), Mouth opening (MO).

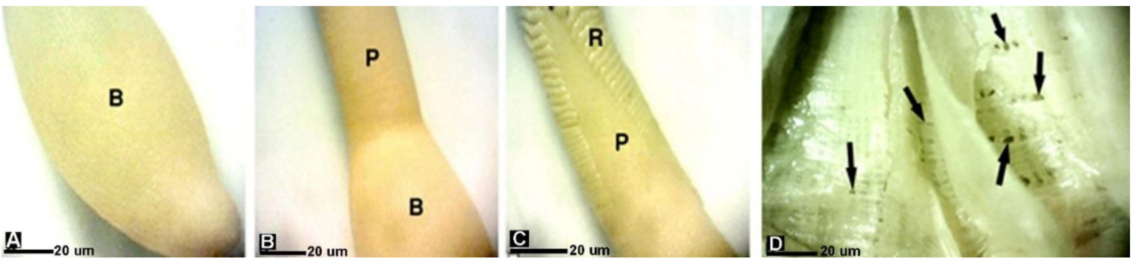

Figure 5: Stereomicroscopic images of bulb, peduncle, and rachis in *Virgularia gustaviana:* bulb (B), peduncle (P) and rachis (R), pores in the wall of the peduncle (black arrows).

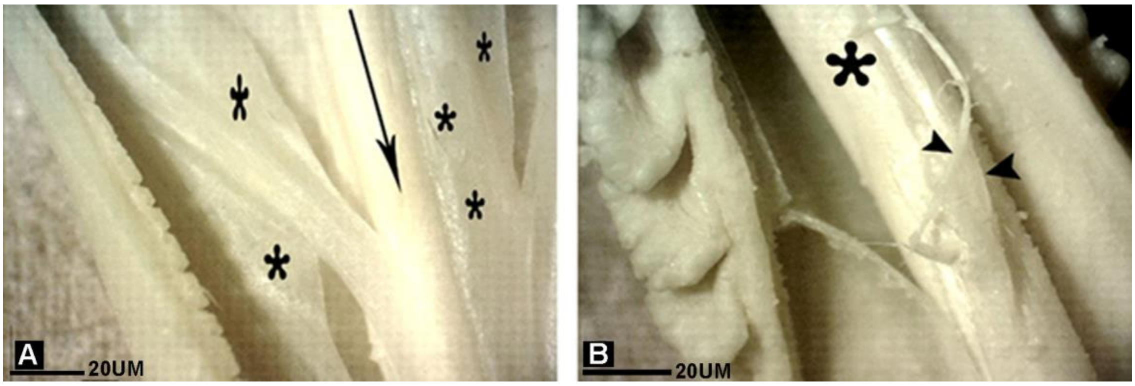

Figure 6: Stereomicroscopic images of axis and surrounding mesenteric septa in peduncle (A) and rachis (B): A. axis (black arrow), surrounding mesenteric septa (black stars); B. axis (black star), surrounding mesenteric septa (black arrowheads).

The axis with a constant length originates from the primary polyp. In the expanded colonies, the axis rarely penetrates the tip of the rachis and the end of the peduncle.

The peduncle is the lower part of the polyp, which extends from the end of the sea pen (that flows into the mud) to the autozooid buds. The caudal end of the peduncle was a swollen bulb (Fig. 5A) with an anchor-like function that anchored the sea pen in the mud. Unlike the rachis, the peduncle had a smooth surface (Fig. 5B, C) with plenty of pores in the wall (Fig. 5D).

In the transverse section, a mineralized axis was observed at the center along the body, surrounded by mesenteric septa that expanded from the inner wall of the sea pen to the axis (Fig. 6).

In fully contracted specimens it is observed throughout the colony. The four mesenteric septa were extended from the inner wall of the body towards the axis (Fig. 7). In the rachis, the cross-section of the axis was circular (or square in some cases) and was finally merged with mesenteric septa (Fig. 7A, B, C). In the peduncle, the axis was square and four flat muscle strips along the mesenteric septa from the inner wall were connected to each side of the axis (Fig. 7D). The axis was narrow and hook-like at the apex and the end of the polyp (Fig. 7E, F). The axis was surrounded by a sponge-like tissue, called sclerite, and mesenteric septa all along its length (Fig. 7).

The four mesenteric septa around the axis divided the primary polyp cavity into five canals: a ventral canal, a dorsal canal, two lateral canals, and a central canal where the axis was located (Fig. 8). The size of the canals varied over the colony; lateral canals were usually small; however, the dorsal and ventral canals were usually larger in the rachis and peduncle, respectively (Fig. 8A, D). The size of the canals decreased to the top of the rachis until they finally merged (Fig. 8B). Unlike other parts of the peduncle, the inner space of the bulb was divided into two canals by one mesenteric septum (Fig. 8E, F).

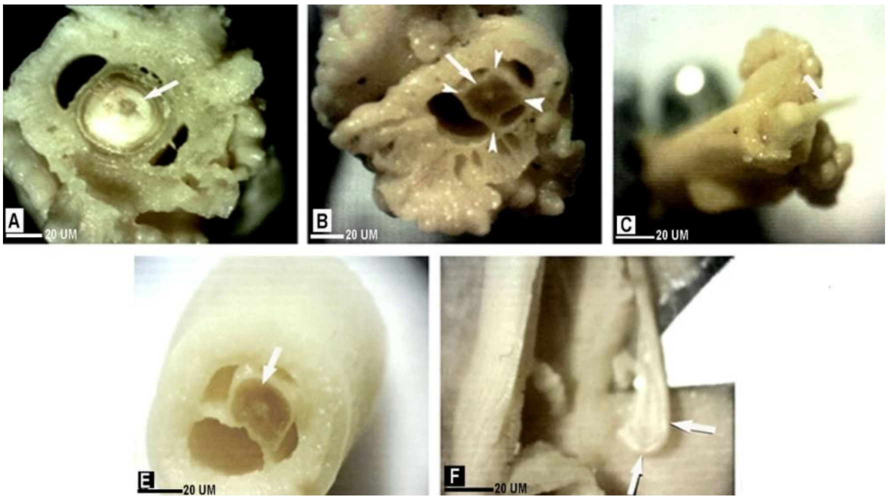

Figure 7: Stereomicroscopic images of axis in the rachis (A,B, and C) and peduncle (E,F, and D); axis (white arrow), mesenteric septa (white arrowheads).

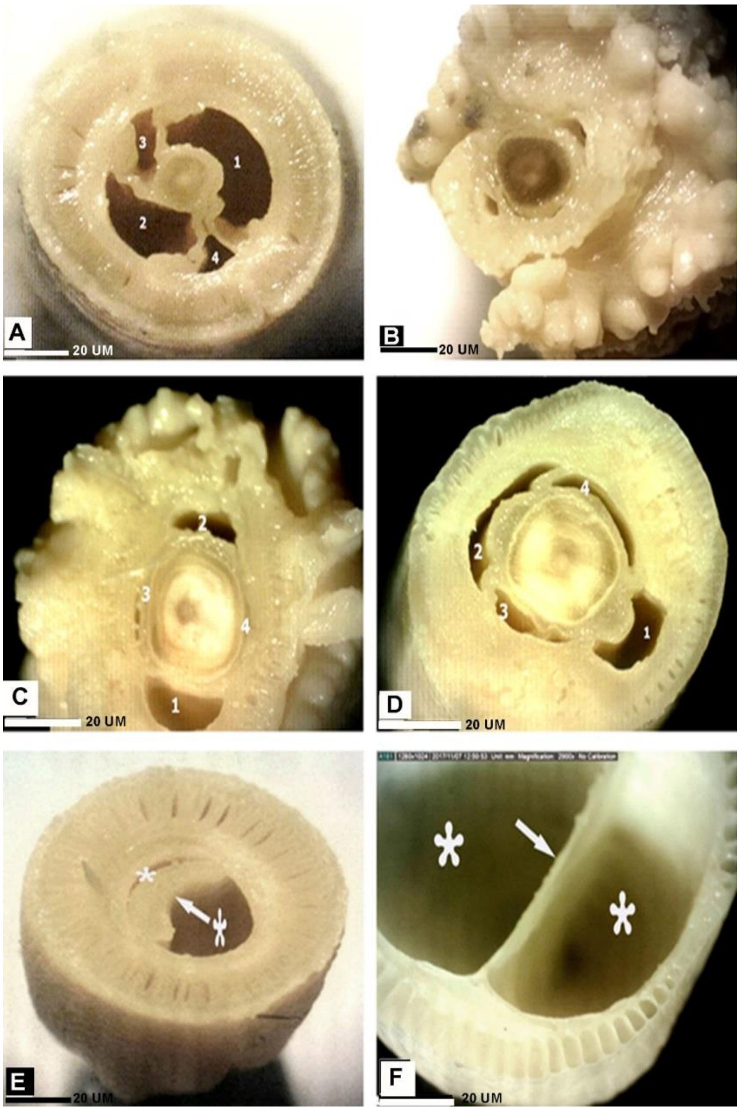

Figure 8: Stereomicroscopic images of axis and surrounding mesenteric canals in rachis (A and B), peduncle (C and D) and bulb (E, F); mesenteric canals (1,2,3, and 4), mesenteric canals (white star), mesenteric septum (white arrow).

Discussion

Due to its geographical location and special weather conditions, the Persian Gulf has very diverse organisms, so it is necessary to investigate and identify the different species living in it, especially the species that are less studied. According to available reports, *V. gustaviana* is the dominant species of sea pens on the Iranian coasts of the Persian Gulf (Safaeian *et al.*, 2009; Sharifi *et al.*, 2015). Because of the few studies conducted regarding this species in Iran, the present study was carried out to investigate the morphological and anatomical characteristics of this species.

William (2011) identified two types of spicules in *V. gustaviana* using an electron microscope, in the form of three edges in atosoids and siphonozooids, and rice grains in the peduncle. In the present study, similar spicules were detected.

According to López-González *et al.* (2000) and García-Cárdenas and López-González (2023) sea pens consist of a primary polyp, which turns into a rigid stem with many tubercles (tentacles) called rachis, and some polyps without tentacles called peduncle. Based on the available reports in many species of the family Veretillidae, the rachis has leaf-like tentacles and its length is more than half the length of the body. The peduncle is the lower part of the sea pen's body without tentacles. The bubble-shaped bottom of the peduncle sinks into the seabed like an anchor and helps to keep the animal stable (López-González *et al.,* 2000; García-Cárdenas and López-González, 2023). All of these findings were in agreement with the anatomical structure of *V. gustaviana.*

López-González *et al.* (2000) stated that the pennatulaceans rachis consist of two with and without tentacle sections called autozooids and siphonozooids, respectively. Autozooids are feeding structures on the ventral surface of the sea pen. Siphonozooids are two flat strips along the dorsal surface of the sea pen involved in the water circulation (López-González *et al.*, 2000). According to William (2011) the beautiful appearance of the sea pens is due to the expansion of autozooids and tentacles to absorb suspended food in the water column. Batie (1971) reported that the middle part of the tentacles is directly related to mesenteric canals in all species of order Pennatulacea. He also stated that the mesentery continues into the polyp and connects the wall of the digestive tract with the body wall of the polyp (Batie, 1971). Mesentery then disappears towards the base of the polyps, so the wall of the digestive tract is merged with the wall of the polyp (Batie, 1971; Miatta and Snelgrove, 2022).

According to William (2015), the peduncular wall is very porous, which allows it to swell a lot. Two rows of canals create the sponge layer of the peduncular wall. The outer layer of the canal is longitudinal or parallel to the peduncle. There is a secondary porous layer in the middle of this spongy layer that surrounds the inner mesentery and the axis (William, 2015). In the present study, the porous wall of *V. gustaviana* was confirmed.

Based on the reports of William (2011), most pennatulaceans have a central axis, mainly composed of calcium carbonate. Nonaka *et al.* (2012) also reported that the central axis, with a circular or elliptical cross-section, extended throughout the colony in the Virgulariidae family. These findings are in agreement with the results of the present study. Wilson *et al.* (2002) found that in *Halipteris willemoesi*, the cone-shaped axis extended throughout the colony *and* its diameter decreased towards the end of the peduncle.

In conclusion, though all sea pens have the basic anatomical structure as explained, the form of the colony in different species differs significantly in terms of evolution, particularly their environmental compatibility. Upcoming investigations will emphasize on the comparative morphometric and anatomic analysis of different species of sea pens from the Persian Gulf.

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