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# Diversity of Sea Cucumber Types in Shallow Sea Waters of Katapang Sukarame Carita Labuan Pandeglang Banten Beach

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#### Abstract

Cucumbers of the sea are one of the animals from the phylum Echinodermata that have ecological and economic roles. Biogeographically, the types of sea cucumbers from the Holothuriidae family are not yet well known in the waters of Shallow Waters of Katapang Sukarame Carita Labuan Pandeglang Banten Coast, particularly the information on the types and their phylogenetic relationships. This study aims to determine the diversity and phenetic relationship between types of sea cucumbers in Shallow Waters of Katapang Sukarame Carita Labuan Pandeglang Banten Coast. The research has stages which include preliminary observations of sea cucumber species. Preliminary observations were carried out by surveying the location of the sea cucumber catch and interviewing local fishermen. The results of the survey on preliminary observations were used as the basis for determining the sampling station. Sampling of the specimens was carried out using a cruising technique based on a predetermined sequence of sampling stations. Observations and species determination were carried out to obtain data as material for the analysis of taxonomy and phenetic relationships. The research results show that the diversity of sea cucumber species found in the Shallow Waters of Katapang Sukarame Carita Labuan Pandeglang Banten Coast are S. horrens, H. atra, and H. leucospilota. There is a phylogenetic relationship among the three sea cucumber species in the Shallow Waters of Katapang Sukarame Carita Labuan Pandeglang Banten Coast. Group 1, which consists of H. atra, has a similarity of 85.393% with H. leucospilota. Group 2, which includes S. horrens, has a similarity of 64.130% with H. atra and H. leucospilota.

Keywords: Phenetics; Diversity; Sea Cucumbers.

#### INTRODUCTION

Indonesia is an archipelago with 17,508 islands and an area of 8.3 million km2, making it the largest archipelagic country in the world. Indonesia's coastline stretches 81,000 km, creating diverse ecosystems in coastal areas, including mangrove forests, coral reefs, and seagrass beds. These ecosystems boast high biodiversity, as evidenced by the presence of nearly every phylum, including Coelentrata, Mollusca, Annelida, and Holothuroidea (phylum Echinodermata). Holothuroidea play a crucial role in marine ecosystems (Hedriansyah et al., 2018).

Holothuroidea, or sea cucumbers, are a marine resource with enormous potential in Indonesia. Sea cucumbers are distributed throughout the world, and in Indonesia, they are found throughout the ocean from Sabang to Merauke (west to east). Sea cucumbers are found along almost all coastlines, from shallow to deep sea areas (Agustina & Sulaiman, 2021).

Globally, there are approximately 1,135 identified species of sea cucumbers, and in Indonesia, there are approximately 257 species of sea cucumbers distributed

throughout Indonesian waters. Of these, 60 species are generally known to the public. Only 23 species of sea cucumbers in Indonesia have been utilized, exploited, and consumed by the public. Five of these 23 species of sea cucumbers have high economic value: Holothuria scabra, Holothuria nobilis, Holothuria vacabunda, Holothuria vatiensis, and Holothuria marmorata. The most widely exploited, captured, and traded sea cucumber is the sand sea cucumber (Holothuria scabra).

Of these five sea cucumber species, the sea cucumber has an elongated body shape and tentacles around the mouth opening used to capture prey. The sea cucumber's tubular legs (podia) are located on the ventral side of the body, representing pseudopods. The cross-section of a sea cucumber's body is round, semicircular, trapezoidal, or square, and can be elongated. The sea cucumber's body is 80-90% water, and it will protrude from the body when not in water. The sea cucumber's anus is located at the tip and can open and close regularly. Some sea cucumber species can secrete Cuvier's tubules (sticky white threads) as protection against physical or chemical disturbances (Setiawan et al., 2017).

Each sea cucumber has spicules, tentacles, papillae, and podia. Sea cucumber spicules are microscopic spines located within the integumentary tissue. Sea cucumber spicules are composed of chalk that dissolves in acidic solutions. The shape and composition of spicules vary depending on the sea cucumber species, becoming a distinctive characteristic of sea cucumbers at the genus and species levels. Variations in spicule shape include stem-shaped, branched stem-shaped, plate-shaped, rosette-shaped, button-shaped, anchor-shaped, and table-shaped. The body color of sea cucumbers varies by species and genus, ranging from black, gray, brownish, reddish, yellowish, and white (Wulandari et al., 2012).

Sea cucumbers have important economic and ecological roles. They are a crucial component of the ecosystem's food chain because they serve as a food source for various reef fish species (Hedriansyah et al., 2018). Sea cucumbers' economic importance, including their use as food, pharmaceutical, and industrial raw materials, is due to their high nutritional content. When dry, sea cucumbers contain protein (82%), fat (1.7%), water (8.9%), ash (8.6%), and carbohydrates (4.8%) (Komala, 2015).

Sea cucumbers contain EPA (Eicose Pentaenoic Acid) and DHA (Docose Hexaenoic Acid), saturated fatty acids that are beneficial as wound healing agents, antithrombotic agents, and accelerate cell regeneration, as well as anti-cholesterol, stroke, and anti-aging agents. Sea cucumbers contain minerals such as calcium, sodium, phosphorus, chromium, manganese, iron, cobalt, zinc, and vanadium, all of which are highly beneficial. Sea cucumbers are used medicinally as a source of testosterone, antigens, steroids, collagen, vitamin C, and minerals such as chromium, iron, cadmium, manganese, nickel, cobalt, and zinc (Roni et al., 2020).

Sea cucumbers have high nutritional value and extraordinary potential, leading to their massive exploitation without considering their sustainability. Export demand for sea cucumber products in Indonesia and the increasing price of sea cucumbers on the international market have further fueled the increase in exploitation and large-scale harvesting of sea cucumbers from their natural habitat (Elfidasari et al., 2012).

The large-scale exploitation and harvesting of sea cucumbers from their natural habitat has resulted in the intensive harvesting of sea cucumber species without regard for their species and size. Even sea cucumbers with no economic value are exploited, resulting in a significant decline in natural sea cucumber populations, which can lead to the extinction of certain sea cucumber species, which in turn leads to the loss of germplasm within the ecosystem (Setiawan et al., 2017).

Banten Province is a region with a diverse aquatic ecosystem. One of the regencies within Banten Province, Pandeglang Regency, boasts a fairly extensive aquatic ecosystem and pristine and healthy coral reefs. The coral reefs along the west coast of Pandeglang Regency,

particularly on Liwungan Island, Badul Island, and Karang Badul, are above 60%. The coral fish diversity index (CFDI) on the west coast of Pandeglang Regency is 106, with an estimated total fauna of 338,745 species. This indicates that the west coast of Pandeglang Regency has moderate species diversity. The average fish abundance in the waters of Pandeglang Regency as a whole is 9,783 individuals/ha (KKHL, 2021).

In the waters of Pandeglang Regency, particularly in the shallow waters of Katapang, Sukarame, Carita, Labuan, Pandeglang, Banten, various species of sea cucumbers are estimated to be present, and local communities have exploited them. Biogeographically, little is known about the Holothuriidae species of sea cucumbers in the shallow waters of Katapang, Sukarame, Carita, Labuan, Pandeglang, Banten, especially regarding their species and phenetic relationships. Therefore, identifying the diversity and phenetic relationships of sea cucumber species on the west coast of Pandeglang Regency is crucial.

#### RESEARCH METHODS

The type of research conducted is Survey and Exploration research.

#### **Time and Place Research**

The research was conducted at sampling points in the shallow waters of Katapang Sukarame Carita Beach, Labuan Pandeglang, Banten, which is an underwater nature conservation area. Specimen collection was carried out at low tide, considering safer sea waves (often uncertain due to weather and lunar gravity), low tide at 10/11 a.m. and high tide at 1 p.m. The specimen collection results were then identified at the Integrated Laboratory of FSFK UNMA Banten.

# **Tools and materials**

Specimen collection in the field using snorkeling equipment, hooks or clamps. Microhabitat observation of captured specimens using a digital camera device (Gadget/Smartphone: Realme 8, Model: RMX3085, Camera: 64 MP AI Quad Camera). 5 sample containers for preserved species and 5 containers for live species. Dissection of the sea cucumber body for observation of internal organs using surgical equipment (dissecting kit). Macroscopic observation of external body parts and visceral organs using a magnifying glass. Spicule observations were carried out using a microscope and digital imaging results. The main material consists of sea cucumber specimens for observation of species diversity and phenetic relationships. Specimens for observation of species diversity are groups of sea cucumbers that have been treated with anesthesia and preservation. The chemicals used for anesthesia and relaxation of sea cucumber tissue are 70% MgCl2 solution, 70% Ethanol

preservative solution and NaClO (a compound used for bleaching)

# Ways of Working

# Preliminary observation of sampling locations

Preliminary observations through exploration were conducted in the sampling area, namely the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast to determine the sampling point. The sampling point was determined based on the presence and abundance of sea cucumbers in both the intertidal and subtidal zones, which are the usual locations for sea cucumber catches. The research location was in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast.

# Sampling

Sampling was conducted in the intertidal zone during low tide in coastal areas. The diving team and fishermen were assisted in sampling (weather and natural conditions significantly influence the sampling process). Sampling in the subtidal zone was conducted with the assistance of fishermen and using hooks or tongs to catch sea cucumbers. Sea cucumber sampling was conducted at sampling points determined based on preliminary observations. The sampling points were determined based on the diving team's findings, namely the distance from the shoreline (10 to 50 meters from the shoreline) with a depth of approximately (3 to 5 meters).

# Sample Management

Sea cucumber samples from the field were washed with clean water and cleaned of any adhering dirt. Samples were grouped based on morphological characteristics to observe species diversity and phenetic relationships.

The specimens obtained for species diversity observation were anesthetized using a 2000 mL MgCl2 solution mixed with seawater and subjected to tissue relaxation. The sea cucumbers were preserved in specimen containers containing 70% ethanol for laboratory observation and identification.

# Specimen identification

The sea cucumber specimens were identified and dissected. Their external and internal morphology was observed, and their morphometric measurements were taken. A section of the body wall was taken for spicule observation. The samples were bleached and the spicules were observed under a microscope. The resulting spicules were matched for identification.

Soaking the internal body wall fragments loosens the integumentary tissue, causing the spicules to detach from the tissue. The spicule composition observed was derived from the anterior, dorsal, posterior, and ventral body wall fragments. The spicule composition was observed using a digital microscope and digital imaging. Spicule observation serves as a means of distinguishing sea cucumber species.

The steps taken to study the ossicles include cutting 1–5 mm2 of tissue from each body region, including the anterior, dorsal, ventral, and tentacles. Place the tissue fragments in a 25 ml beaker and soak them in a NaClO bleaching solution for 45–60 minutes, until all muscle tissue is destroyed. Afterward, the ossicle samples were washed with distilled water 4–7 times. The ossicle samples were then placed back on a concave slide, where measurements and images were taken. The samples were then ready to be observed under a microscope and photographed with a camera (Widianingsih et al., 2015).

Morphological observations of sea cucumbers included the external body parts (external) and internal parts of the sea cucumber, including the following organs:

- 1. The body consists of length: width, tip (anterior), base (posterior), cross-section, ventral side, dorsal side (lateral).
- 2. The mouth consists of shape, diameter and location.
- 3. The anus consists of shape, diameter (length), location, color, anal teeth.
- 4. Body wall, namely thickness/texture
- 5. Tentacles consist of type, color (tip of tentacle), diameter, protective papillae, number, circle, and length (stalk).
- 6. The epidermis (dorsal lateral) consists of the surface, color (surface), grooves (specific), *tuberkel and pediselus*.
- 7. Tube feet consisting of arrangement, length, shape and number.
- 8. Papillae consist of tips (color), shape, number, arrangement and location.
- 9. Ambulacral grooves consisting of number and arrangement.
- 10. Gonads consist of shape (1 lobe/2 lobes) and presence or absence.
- 11. Respiratory tree consisting of the presence or absence, shape of tubules and shape of the ends of tubules.
- 12. Cuverian tube (presence or absence).
- 13. The mesentery consists of color, number of dorsal attachments and number of ventral attachments.
- 14. The chalk ring consists of a conical curve of the radial tip, a conical curve of the radial base, the diameter of the ring and the height of the ring plate.
- 15. Digestive tract (intestinal length:body length ratio).
- 16. Spicules consist of dominant, C-shaped, table-shaped, button-shaped, rosette-shaped, plate-shaped, rod-shaped, grain-shaped and typical spicules.

Morphometric observations of sea cucumbers include organs: body, mouth, anus, body wall, tentacles, respiratory tree, digestive tract, Cuverian tube, calcareous ring, tube feet, gonads, papillae and longitudinal muscles.

# Data analysis

# Taxonomic Analysis

The results of the sea cucumber identification then made a taxonomy of the diversity of each type of sea cucumber that was found and identified.

#### Phenetics Relationship Analysis

Characters for identification are determined according to the stages of determining the Operational Taxonomic Unit (OTS), selecting characters, assigning character values, and calculating taxonomic distances. Taxonomic characters are measurable data that is then coded with numbers for clustering analysis. Clustering analysis uses the UPGMA (unweight pair group with arithmetic average) method using the MVSP 3.1 program. The results of the analysis are in the form of a dendogram to determine the groups/clusters of each OTS based on similarity.

#### RESULTS AND DISCUSSION

#### Results

Based on observations, the species found in the shallow waters of Katapang, Sukarame, Carita, Labuan, Pandeglang, Banten are presented in Table 1.

Table 1. Sea Cucumber Species Discovery Results.

No	Famili	Spesies
1	Stichopodidae	Stichopus horrens
2	Holothuriidae	Holothuria atra
3	Holothuriidae	Holothuria leucospilota

# Comparison of Macroscopic Observations of Sea Cucumber Organs

The results of macroscopic comparative observations on the organs of sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota in the organ sections: body, mouth, anus, body wall, tentacles, epidermis (lateral dorsal), tube feet, papillae, ambulacral grooves, gonads, respiratory tree, Cuverian tube, Mesentery, lime ring, digestive tract, spicules.

#### **Body**

Comparison of observations of body organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota, is presented in Table 2.

Table 2. Comparison of Body Organs in Sea Cucumber Species.

Body organs	S. horrens	H. atra	H. leucospilota
Length: Width	240 mm: 78 mm	126 mm : 45 mm	115 mm : 39 mm
Tip (Anterior)	Obtuse	Tapered	Tapered
Base (Posterior)	Obtuse	Blunt	Blunt
Cross- section	Square/Trapezoid	Rounded	Rounded
Ventral Side	Flat	Rounded	Rounded
Dorsal Side (Lateral)	Square	Rounded	Rounded

#### Mouth

Comparison of observations of the mouth organs of sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota, is presented in Table 3.

Table 3. Comparison of Mouth Organs in Sea Cucumber Species.

Mouth organs	T. horrens	I. atra	H. leucospilota
Shape	circle	circle	oval
Diameter	10,8 mm	5,2 mm	8,6 mm
Location	subterminal	subterminal	subterminal

### Anus

A comparison of anal organ observations in sea cucumber species found in the shallow waters of Katapang, Sukarame, Carita, Labuan, Pandeglang, Banten, namely S. horrens, H. atra, and H. leucospilota, is presented in Table 4.

Table 4. Comparison of the Anus Organs in Sea Cucumber Species.

Anus Organs	U. horrens	J. atra	H. leucospilota
Shape	Round	Round	Round
Diameter (Length)	6 mm	4,8 mm	5 mm
Location	Terminal	Terminal	Terminal
Color	Reddish black	Black	Black
Anal teeth	None	None	None

# **Body Wall**

Comparison of observations of body wall organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota, is presented in Table 5.

Table 5. Comparison of Body Wall Organs in Sea Cucumber Species.

Body wall	V. horrens	K. atra	H. leucospilota
Thichness/texture	6,3 mm	0,6 mm	0,7 mm

#### **Tentakel**

Comparison of observations of tentacle organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota, is presented in Table 6.

Table 6. Comparison of Tentacle Organs in Sea Cucumber Species.

Organ Tentakel	S. horrens	H. atra	H. leucospilota
Type	Shield	Shield	Shield
Color (Tentacle tip)	Translucent	Black	Black
Diameter	6,9 mm	2,3 mm	3,2 mm
Protective Papillae	Available	Available	Available
Number	19	20	21
Circle	1 circle	1 circle	1 circle
Length (stalk)	97 mm	19 mm	39 mm

### **Epidermis (Lateral Dorsal)**

Comparison of observations of the epidermis organs (dorsal lateral) in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 7.

 Table 7. Comparison of Dorsal Lateral Organs in Sea Cucumber Species

Epidermis (lateral dorsal)	S. horrens	H. atra	H. leucospilota
Surface	Uneven	Slippery and	Slippery
Color (surface)	Dark green	Slimy	Black
Grooves (specific)	Grooves present	None	None
Tubercles	Available	None	None
Pedicellus	None	None	None

# **Tube feet**

Comparison of observations of tube foot organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 8.

Table 8. Comparison of Tube Feet Organs in Sea Cucumber Species.

Tube feet	W. horrens	L. atra	H. leucospilota
Arrangement	Numerous, 4 line	Regular	Irregular
Length	1-9 min	2,8 mm	3-5 mm
Shape	Tubes	Short tube	Short tube
Name le ou	Numerous,	Numerous,	Numerous,
Number	regular	irregular	irregular

# Papila

Comparison of observations of Papilla organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 9.

Table 9. Comparison of Papilla Organs in Sea Cucumber Species.

Organ papila	H. horrens	H. atra	H. leucospilota
Tip (color)	According to the pattern	Black	Black
Shape	Conical and blunt tapered	Short tapered	Tapered
Number	Many	Many	Many
Arrangement	Irregular	Irregular	Irregular
Location	Outer body wall (except in the ventral)	Outer wall of the body (except in the ventral)	Outer wall of the body (except in the ventral)

#### Alur Ambulakral

Comparison of observations of the ambulacral groove organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 10.

Tabel 10. Comparison of Ambulacral Arul Organs in Sea Cucumber Species.

Arul ambulakral	H. horrens	H. atra	H. leucospilota
Amount	4 lines	None	None
Arrangement	None	None	None

# Gonad

Comparison of gonad organ observations in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota, is presented in Table 11.

Table 11. Comparison of Gonad Organs in Sea Cucumber Species.

Gonad	H. horrens	H. atra	H. leucospilota
Shape (1lobe/2 lobes)	-	-	-
Presence or absence	Not found	Not found	Not found

# **Respiratory Tree**

Comparison of observations of respiratory tree organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 12.

**Table 12.** Comparison of Respiratory Tree Organs in Sea Cucumber Species.

Respiratory tree	X. horrens	M. atra	H. leucospilota
Availability	Available	Available	None
Tubule shape	Fine thread	Fine thread	None
End shape tubules	Alveoli	Alveoli	None

#### Cuverian Tube

Comparison of observations of Cuverian tube organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 13.

Table 13. Comparison of Cuverian Tube Organs in Sea Cucumber Species.

Cuverian Tube	H. horrens	H. atra	H. leucospilota
Availability	None	None	Available

#### Mesenterium

Comparison of observations of the Mesentery organ in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 14.

Table 14. Comparison of Mesentery Organs in Sea Cucumber Species.

Mesenterium	H. horrens	H. atra	H. leucospilota
Color	Transparent	Brownish white	Transparent yellow
Number of attachments dorsal	1	2	2
Number of attachments ventral	1	1	2

## Chalk ring

Comparison of observations of the lime ring organ in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 15.

Table 15. Comparison of Cretaceous Ring Organs in Sea Cucumber Species.

Chalk ring	H. horrens	H. atra	H. leucospilota
Conical curve apostle tip	Available	Available	Available
Conical curve	None	Available	None
Radial base ring diameter	23,2	16,1	14,2
Piece height ring	4,2	2,1	2,5

# **Digestive tract**

A comparison of observations of the digestive tract organs of the sea cucumber species found in the shallow waters of Katapang, Sukarame, Carita, Labuan, Pandeglang, Banten, namely S. horrens, H. atra, and H. leucospilota, is presented in Table 16.

Table 16. Comparison of Digestive Tract Organs in Sea Cucumber Species.

Digastive tract	H. horrens	H. atra	H. leucospilota
Intestinal length	926 mm	417 mm	350 mm
Body length	286 mm	130 mm	139 mm
Ratio	926 mm :	417 mm :	350 mm: 139
Kano	286 mm	130 mm	mm

# **Spicule**

Comparison of observations of spicule organs in sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota is presented in Table 17.

Table 17. Comparison of Spicule Organs in Sea Cucumber Species.

Spicule	H. horrens	H. atra	H. leucospilota
Dominant	Table	Plate shape (perforation)	Button shape
C-shape	Yes	No	No
Table shape	Yes	Yes	Yes
Button shape	No	No	Yes
Rosette shape	No	No	No
Plate shape	Yes	Yes	Yes
Rod shape	No	No	No
Grain shape	No	No	No
Typical spicules	Yes	No	Yes

# Comparison of Morphometric Observations of Sea Cucumber Organs

The results of comparative morphometric observations on the organs of sea cucumber species that were successfully found in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast, namely S. horrens, H. atra and H. leucospilota in the organ sections: body, mouth, anus, body wall, tentacles, respiratory tree, digestive tract, Cuverian tube, lime ring, tube feet, gonads, papillae, longitudinal muscles.

Table 18. Comparison of Organ Morphometrics in Sea Cucumber Species.

Organ	Characteristics	H. horrens	H. atra	H. leucospilota
Body	Length	212,5 mm	166,67 mm	196,33 mm
•	Width	67,1 mm	44,30 mm	41,40 mm
Mouth	Diameter	9 mm	7,37 mm	8,80 mm
Anus	Diameter	14,65 mm	15,27 mm	14,40 mm
Wall thickness	Tentacle	4,45 mm	1,80 mm	1,40 mm
Tentacle	Diameter	4,05 mm	3,07 mm	3,03 mm
	Stalk Length	26,45 mm	10,13 mm	22,67 mm
Respiratory tree	Length (Organ)	471,15 mm	34,30 mm	-
Digestive tract	Intestine Length/Height Ratio	670 mm	601,67 mm	560,67 mm
Cuverian tube	Size			
	Length/Width	-	-	55,07 mm
	Radial/interradial	18,45 mm	21,97 mm	22,93 mm
Calcareous ring	Plate height	2,75 mm	3,00 mm	2,30 mm
Tube feet	Length	2,55 mm	2,97 mm	3.,10 mm
Gonad	Length/width	_	-	-
Papila	Length	1-9 mm	2 mm	3-5 mm
Long length of	Integument	2 baris	2 baris	2 baris

# **Taxonomic Analysis**

#### Stichopus horrens

The classification (grouping) of Stichopus horrens in the taxonomic system is as follows:

Kingdom : Animalia
Phylum : Echinodermata
Class : Holothuroidea
Order : Synallactida
Family : Stichopodidae
Genus : Stichopus
Species : Stichopus horrens

Local Name: fried peanut sea cucumber, taikongkong, kacang, susu, rengget. International Name: Dragonfish. Brief description: its body is solid, fleshy, thick, folded, and soft. Its body color is yellowish green with small squares and blackish brown stripes. White, long, and small papillae with large protrusions at the base. These protrusions are greenish white and somewhat transparent, spread across the dorsal surface. Tube feet are arranged in three longitudinal rows on the ventral surface.

# Holothuria atra

The classification (grouping) of Holothuria atra in the taxonomic system is as follows:

Kingdom: Animalia
Phylum: Echinodermata
Class: Holothuroidea
Order: Holothuriida
Family: Holothuriidae
Genus: Holothuria
Species: Holothuria atra

Local Name: black or brown takling sea cucumber, black, dara, keling, cera. International Name: Lollyfish / Black trepang. Short Description: The body is elongated, medium-fleshed and relatively hard. The entire body is reddish-black and covered with long, small, and dense papillae on the dorsal surface. The ventral surface of the body is also covered with small, long, densely arranged tube feet.

#### Holothuria leucospilota

Klasifikasi (Brandt, 1835):

Kingdom : Animalia
Phylum : Echinodermata
Class : Holothuroidea
Order : Holothuriida
Family : Holothuriidae
Genus : Holothuria

Species : Holothuria leucospilota

Local Name: Timun laut gum, cera, jepun, keling, talengko. International Name: -. Short Description: The body is elongated, medium-fleshed and soft. The entire body is black with large and long papillae, densely arranged on the dorsal surface. Tube feet are similar to papillae and densely arranged on the ventral surface.

# **Phenetics Relationship Analysis**

Analysis of phenetic relationships with a dendrogram of kinship relationships of 3 sea cucumber species based on morphological characteristics in the Operational Taxonomic Unit (OTU) (Table 19).

Table 19. Characters in Operational Taxonomic Units (OTS).

I	Body organs	Criteria
Body	Tip (ante)	0 = tapered
		1 = blunt
	Base (post)	0 = tapered
		1 = blunt
	Cross Section	0 = square
		1 = rounded
	Ventral side	0 = flat
		1 = rounded
	Dorsal side (lateral)	0 = square
		1 = rounded
Mouth	Shape	0 = oval
	•	1 = circle
	Location	0 = terminal
		1 = subterminal
Anus	Shape	0 = square
Allus	Shape	1 = round
	Location	0 = subterminal
	Location	
		1 = terminal
	Color	0 = not black
		1 = black
	Anal teeth	0 = present
		1 = absent
Tentacles	Туре	0 = pinnate
		1 = shield
	Color	0 = black
	20101	1 = not black
	Protective papilae	0 = absent
	1 fotective papilae	
	C	1 = present
	Surface	0 = uneve
		1 = smooth and slimy
Epidermis (lateral dorsal)	Grooves	0 = present
		1 = absent
	Tubercles	0 = present
		1 = absent
	Pedicellus	0 = present
		1 = absent
	Arrangement	0 = present
		1 = absent
Tube feet	Length	0 = short
Tube leet	Lengui	1 = long
	Chana	0 = not tube
	Shape	
		1 = tube
	Number	0 = few
		1 = many
	Tip (color)	0 = patterned
		1 = black
Papillae	Shape	0 = tapered
-	-	1 = blunt
	Number	0 = few
	<del></del>	1 = many
	Arrangement	0 = regular
	/ mangement	0 – regular 1 = irregular
Ambulakral arasya	Number	0 = available
Ambulakral groove	INUITIOCI	
	<u> </u>	1 = none
	Arrangement	0 = available
		1 = none
Gonad	Availability	0 = available
		1 = none
Respiratory Tree	Availability	0 = available
-	,	1 = none
	Tubular shape	0 = available
	I double brupe	o available

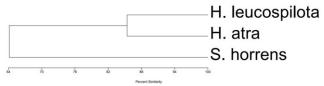
		1 = none
	Tubular tip shape	0 = available
		1 = none
Cuverians Tube	Availability	0 = available
		1 = none
Mesenterium	Color	0 = available
		1 = none
Calcareous ring	Conical curve at tip	0 = available
		1 = none
	Radius	0 = available
	-	1 = none
	Conical curve at base	0 = available
		1 = none
	Radius	0 = available
		1 = none
Spikula	Dominant	0 = available
		1 = none
	Table shaped	0 = available
		1 = none
	Button shape	0 = available
	<del></del>	1 = none
	Rosette shape	0 = available
		1 = none
	Plastic shape	0 = available
	D 11	1 = none
	Rod shape	0 = available
		1 = none 0 = available
	Grain shape	
	Typical spicules	1 = none 0 = available
	Typical spicules	0 - avanable 1 = none
Body	Body length: width	0 = small
Body	Body length, width	1 = big
Mouth	Diameter	0 = small
Wodin	Diameter	1 = tall
Anus	Diameter (panjang)	0 = short
	(f <b>g</b> )	1 = tall
Body wall	Thickness/texture	0 = thin
		1 = thick
Tentacles	diameter	0 = short
		1 = tall
	Length (stalk)	0 = short
		1 = tall
Calcium ring	Ring diameter	0 = short
2	C	1 = tall
	Ring lobe height	0 = low
		1 = tall
Tentackels	Number	0 = few
		1 = many
	Circle	0 = available
		1 = none
Gonad	Shape (1 lobus/2 lobus)	0 = available
	- ` /	1 = none
Mesentarium	number	0 = available
		1 = none
	Number of ventral attachment	0 = available
		1 = none

After conducting clustering data analysis to determine the close relationships between each sea cucumber species studied, 57 characteristic data from the three sea cucumber species were further processed

using the MVSP 3.22 computer program. This data analysis process can be used as supporting evidence for the close relationships based on morphological characteristics among the three sea cucumber species.

The data processing grouped the morphological characters of each sample based on binary numbers 0 and 1 to obtain clustering analysis using the UPGMA classification analysis.

Based on the clustering analysis, a dendrogram of the phylogenetic relationships of the three sea cucumber species was created based on morphological characteristics using the UPGMA classification analysis. The UPGMA analysis in MVSP 3.2 software was transferred to the Jaggard similarity index tree construction method (Figure 1).



**Figure 1.** Dendogram of the kinship relationship of 3 species of sea cucumber in the shallow sea waters of Katapang Sukarame Carita Labuan Pandeglang Banten coast

#### **DISCUSSION**

# Diversity of Sea Cucumber Species in the Shallow Waters of Katapang Beach

### Sukarame Carita Labuan Pandeglang Banten

Based on morphological observations, several species of sea cucumbers have been found in the shallow waters of Katapang, Sukarame, Carita, Labuan, Pandeglang, Banten, including S. horrens, H. atra, and H. leucospilota. Morphologically and anatomically, each type of sea cucumber differs. These differences are clearly visible in the shape, color, and color pattern of the sea cucumbers.

The variety of habitats for sea cucumbers, combined with different ecological conditions, leads to differences in the species composition, number, and distribution of sea cucumbers in each location. The clustering of certain sea cucumbers in a particular habitat indicates that the presence of these animals is influenced by the availability of food sources in that habitat. Sea cucumbers feed on plankton, detritus, and other organic matter found in mud or sand. Other food sources include small organisms, protozoa, filamentous algae, seaweed, small pieces of marine animals and plants, and sand particles (Elfidasari et al., 2012).

S. horrens is rigid, nearly rectangular, with a body diameter or thickness of 0.20 cm. Its body is white to grayish with irregular brown wart-like nodules all over the body except on the ventral surface, and it lacks Cuvierian tubes. The dorsal surface of the specimen is dark brown with light brown spots. The bases of the papillae are enlarged and form circles, some of which are white. The papillae's exit points are black. The ventral surface of the specimen is cream-colored with distinct

tube feet. The mouth is located on the anterior ventral surface with 20 shield-shaped (peltate) tentacles.

Observations of the spicules in S. horrens revealed a dominant table-shaped structure, with C-shaped, table-shaped, plate-shaped, and distinctive spicules also found. The spicules on the dorsal body wall of the specimen consist of table-shaped spicules with spires forming a crown of thorns, tack-like tables, rosettes, and S- and C-shaped stems. The ventral body wall spicules consist of table-shaped spicules with spires forming a crown of thorns, rosettes, S- and C-shaped stems, stems with spiny surfaces and holes in the middle and edges. The tentacle spicules consist of curved stem spicules with fine, short spines on their surfaces.

Identification results for the sea cucumber H. atra indicate that this sea cucumber has an oval and elongated morphology with a body length of approximately 20 cm and a body weight of approximately 200 grams. The sea cucumber H. atra has a dense black color pattern covering its entire body, making it difficult to distinguish between the dorsal and ventral parts. However, the ventral part usually has a reddish color that forms a straight line from the anterior to the posterior end. The integumentum of this sea cucumber feels rough due to the presence of papillae (small protrusions on the dermis) covering its body.

The sea cucumber H. atra has five white muscles, extending from the anterior to the posterior end and attached to the inner dermis. The intestinal tract resembles a thin, transparent membrane and forms most of the digestive tract, ending in the cloaca. Observations of the spicules in H. atra found that they are predominantly plate-shaped, with some also appearing table-shaped.

Purwati et al. (2008) explained that sea cucumbers possess a micron-sized skeletal structure embedded within the skin tissue, podia, and tentacles, commonly called spicules, or ossicles in international parlance. These spicules can be isolated by removing them from the surrounding tissue using domestic bleaching fluid. Darsono (1998) also explained that the main compound forming spicules is calcium carbonate, which is soluble in acidic solutions. Sea cucumber spicules have a porous structure similar to that seen in other echinoderms and can account for more than 50% of the total endoskeleton volume. The shape and type of spicules vary among species. Therefore, these spicules can be used to characterize sea cucumbers at the genus and species levels.

The spicule isolation technique was based on the method used by Samyn et al. (2005). The observed spicules originated from several body parts, namely the dorsal and ventral body walls, dorsal papillae, ventral tube feet, and tentacles.

Based on research by Aba & Rusliadi (2020), H. atra has a round, elongated body shape with a black, finecolored speckled surface and whitish-yellow tentacles. It measures approximately 20 cm long and 4 cm wide. This sea cucumber typically lives in rocky and sandy waters, often overgrown with seagrass beds. According to Purwati (2008), H. atra is known as the blood sea cucumber because of the reddish fluid it produces when rubbed. Holothuria atra prefers open areas that are constantly inundated during low tides, typically with hard, rough substrates.

According to Massin (1996), H. atra has a cylindrical body shape, measuring 15-30 cm in length. Its body is completely black, both dorsally and ventrally. Papillae and tube feet are irregularly distributed. The mouth is located at the anterior end of the ventral surface, with 20 shield-shaped (palate) tentacles. The anus is located at the posterior end. The dorsal spicules are tables and rosettes, while the ventral spicules are pseudoplates. The tentacles lack spicules. H. atra thrives at temperatures of 23-24°C, with a water pH (acidity) of 7. According to Sutaman (2013), the optimal temperature for sea cucumber survival is between 22-32°C. The optimal pH for H. atra is between 6.5 and 8.

H. leucospilota is brown with black spots on the ventral surface. The ventral surface is pale brown. H. leucospilota has prominent morphological characteristics: an elongated, cylindrical, black body. Its body is soft, flexible, and covered with soft papillae. When contracted, it takes on a pear-shaped shape. It has a soft tegument and a Cuvierian tube.

H. leucospilota has a rounded body cross-section, with a wider posterior section than the anterior section. The ventral surface tends to be flat, and the anal opening is round. The dorsal side is black and the ventral side is dark brown. Its skin is thin and soft. H. leucospilota has Cuvier's tubules. Twenty tentacles are visible. The spicules on the dorsal integument are predominantly button-shaped, with table-shaped, plate-shaped, and distinctive spicules also found.

Based on research by Aba & Rusliadi (2020), H. leucospilota is brown with black spots on the dorsal side. The ventral side is yellowish-white. This sea cucumber, when touched, exudes a white sap. It has the habit of clinging to dead coral, especially on its posterior side, while its anterior side often extends above the sand surface. This species lives in seagrass beds, sandy rocks, and coral reefs. It is commonly found in sandy, coral, and seagrass-covered substrates with a temperature of 23-26°C and a water pH (acidity) of 6-7.

According to Gultom (2004), sea cucumbers typically grow in sandy areas mixed with coral fragments and abundant marine plants or seagrass. According to Lewerissa (2014), the ideal temperature range for sea cucumber growth is 27–30 °C. According to Wibowo et al. (1997), the optimal pH range is 6.5–8.5.

# Phenetic Relationships Between Sea Cucumber Species in West Coastal Waters

# Pandeglang, Banten

The distinguishing features within the class of sea cucumbers (Holothuroidea, Echinodermata) are external morphology, internal organs, and spicules. These characteristics can show a high degree of similarity within certain families, including the Stichopodidae. This family is one of the Holothuroidea (Echinodermata) families that contains most of the commercially important species widespread in shallow tropical waters. To date, the evolution of sea cucumbers based on morphological characteristics used to determine the status of characteristics applicable to phylogenetic analysis remains debated. In fact, no phylogenetic research has yet been conducted on the Stichopodidae family (Wirawati & Purwati, 2016).

Analysis of phenetic relationships among sea cucumber species in the West Coastal Waters of Pandeglang, Banten, used cluster analysis to group similar elements into distinct clusters. Cluster analysis is useful for summarizing data by grouping objects based on shared characteristics.

The sea cucumbers studied in the West Coastal Waters of Pandeglang, Banten, obtained 57 morphological characteristics from three sea cucumber species. These data were further processed using the MVSP 3.22 computer program to obtain clustering analysis groups using the UPGMA method.

The external morphology observed included body cross-sectional shape, maximum body length, arrangement of papillae and tube feet, the presence or absence of papillae protrusions, the position of the mouth and cloaca/anus, the presence or absence of cloaca/anus modifications, and the number of tentacles. The observed morphological characteristics included the shape and size ratio of radial to interradial calcareous rings, the number of gonads, the shape of the madreporite, the number and length of polyan vesicles, and the presence or absence of Cuvier's organs.

The results of the cluster analysis are displayed in the form of a dendrogram, which shows how the data sets are hierarchically interconnected. Based on the obtained similarity matrix, the clustering analysis was performed using the UPGMA method. UPGMA is the most common and recommended method. It also minimizes the amount of distortion that occurs between the dendrogram and the similarity index (Hayati et al., 2018).

The UPGMA method is a simple algorithm for tree construction that assumes the average change along the tree, expressed as a distance. The UPGMA method begins with the distance between the most closely related characters, then averages the distance between the species and the next species, continuing until all species are included in the tree (Andriani, 2016).

The UPGMA analysis in network software was transferred to the Jaggard similarity index tree construction method. Next, the distribution of morphological characteristics of each sea cucumber sample in the West Coastal Waters of Pandeglang, Banten, was analyzed. These were first transformed into binary form and arranged according to the specified characteristics for each listed character. The Jaggard method is one method used to calculate the similarity between two objects (items). Like the cosine distance and matching coefficient, this method is generally calculated based on a vector space similarity measure (Ubaidillah, 2018).

Phenetic relationships with dendrograms of kinship relationships between three sea cucumber species in the West Coastal Waters of Pandeglang, Banten, based on morphological characteristics, with the following criteria: body, mouth, anus, tentacles, epidermis (dorsal lateral), tube feet, papillae, ambulacral grooves, gonads, respiratory tree, Cuverian tubes, mesentery, calcareous ring, spicules, and body wall. The results of the cluster analysis between three sea cucumber species in the West Coastal Waters of Pandeglang, Banten, depicted in a dendogram, demonstrate grouping and demonstrate the closeness of kinship between the three sea cucumber species in the West Coastal Waters of Pandeglang, Banten. The dendogram above shows that the smaller the similarity value of the line connecting one individual to another, the greater the differences between them.

The dendrogram revealed phenotypic similarities between three sea cucumber species in the West Coastal Waters of Pandeglang, Banten. Group I, H. atra, is 85.393% similar to H. leucospilota; group II, S. horrens, is 64.130% similar to both H. atra and H. leucospilota. Both phylogenetic trees or dendograms show that the separation of the two genera is stable and consistent with the currently recognized taxonomy of sea cucumbers at the Stichopodidae family level (Massin, 1999; Samyn et 2005). separation This supports previous phylogenetic studies using mitochondrial DNA analysis (Byrne et al., 2010) and morphological characteristics (Appletans, 2002; Samyn et al., 2005). Furthermore, research by Kerr & Kim (2001) showed that Stichopodidae and Holothuriidae are very closely related (sister groups) (Wirawati & Purwati, 2016).

The results of descriptive analysis indicate that there are differences and similarities between the studied species. The morphological similarity of an organism has a relative value because the characteristics shared do not have significant similarity values. The kinship of living organisms can be determined through similarities in morphological characteristics. The more similar characteristics shared by groups of living organisms, the closer the kinship is considered. The kinship relationship between two individuals or populations can be measured based on similarities in a number of characters, assuming that differences in characters are caused by differences in

genetic makeup. Characters in living organisms are controlled by genes. Genes are pieces of DNA whose activity (expression) can be observed through changes in morphological characters that can be caused by environmental influences (Andriani, 2016).

Based on the results of the dendrogram analysis, the groupings formed in this study can explain the closeness of relationships based on similar characteristics, especially phenotypic (morphological) characteristics. Form or morphological characters are generally the best data for delimiting a taxon because good taxonomic delimitation is achieved using easily visible characters, rather than hidden ones. For this reason, morphological characters can be used as a source of taxonomic evidence. Furthermore, the results of research using morphological characters (phenotypic characters), as conducted in this study, indicate that morphological characters as taxonomic evidence are indeed very effective for identifying and analyzing the diversity of sea cucumber species in the West Coastal Waters of Pandeglang, Banten, and for determining their close kinship relationships.

#### CONCLUSION AND SUGGESTION

#### Conclusion

Based on the results and discussion, the following conclusions can be drawn:

- The diversity of sea cucumber species found in the West Coastal Waters of Pandeglang, Banten, includes S. horrens, H. atra, and H. leucospilota.
- The dendrogram reveals phenetic similarities between the three sea cucumber species in the West Coastal Waters of Pandeglang, Banten. Group I, H. atra, is similar to H. leucospilota with a similarity percentage of 85.393%; group II, S. horrens, is similar to both H. atra and H. leucospilota with a similarity percentage of 64.130%.

#### **Suggestion**

Based on the discussion and conclusions, the following recommendations are made:

- More thorough exploration and identification of sea cucumber diversity in the West Coastal Waters of Pandeglang, Banten is needed.
- Further research is needed to identify sea cucumbers in the waters of the West Coast of Pandeglang, Banten.
- Future studies should utilize molecular data for comparison with existing phylogenetic studies of the Stichopoda and Holothuriidae families.
- Morphological characteristics such as color patterns during life, body wall thickness, and so on should be added, so that the number of spicule characteristics used is balanced with the number of external morphological characteristics.

 Local communities are expected to maintain and preserve the ecosystem that serves as the habitat for sea cucumbers so that they remain preserved in nature.

**Competing Interests:** The authors declare that there are no competing interests.

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