Egyptian Journal of Aquatic Biology & Fisheries Zoology Department, Faculty of Science, Ain Shams University, Cairo, Egypt. ISSN 1110 – 6131 Vol. 28(1): 2007 – 2030 (2024) www.ejabf.journals.ekb.eg



Composition and Density of Benthic Diatoms in Sediments with Different Compost Mixtures on Cultivation of Sea Cucumbers (*Holothuria scabra*) Using Floating Net Cages

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ARTICLE INFO Article History:

Received: May 26, 2023 Accepted: Dec. 27, 2023 Online: Feb. 28, 2024

Keywords: Sago waste, Chicken manure, Seagrass leaves,

Natural feed

ABSTRACT

Diatoms, a group of phytoplankton known as Bacillariophyceae, serve as food for benthic organisms such as sea cucumbers (Holothuria scabra). Sea cucumber raising has the potential to alleviate wild fishing pressure, satisfy a sizable market demand, and improve the water quality in aquaculture settings. This study aimed to analyze the composition and density of benthic diatoms as a natural feed for sea cucumbers deposited in sediments. The research was carried out in the waters of the Inner Ambon Bay, Maluku, Indonesia. The rearing of sea cucumbers involves using floating net cages with the off-bottom method. This method utilizes suspended containers and sediment, with the addition of seagrass leaves, sago waste, and chicken manure to stimulate the growth of benthic diatoms, which serve as a natural feed for sea cucumbers. This study used three treatments: i) treatment A (seagrass leaves + chicken manure), ii) treatment B (seagrass leaves + sago waste), and iii) treatment C (seagrass leaves + chicken manure + sago waste). Data analysis was descriptively carried out and displayed in the form of tables and graphs. The research results showed that two orders, 14 families, 25 genera and 66 species of benthic diatoms were found in the sediments of sea cucumber cultivation floating net cages. Density of benthic diatom species, composition, and quantity were influenced by varying meal compositions. Treatments involving seagrass leaves, sago waste, and chicken manure resulted in increased diatom densities and genera composition. The benthic diatoms found were dominated by the order Pennales, in particular the different species belonging to the genera of Navicula. The results of this study found that treatment with a more diverse compost composition was able to support the high number and density of benthic diatoms since it was suspected that they had a higher nutrient content. The results of this study provide information about the effectiveness of using sago waste + chicken manure + seagrass leaves as an alternative compost to stimulate the growth of benthic diatoms as a natural feed in sea cucumber cultivation.

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INTRODUCTION

Benthic microalgae are one of the autotrophic organisms contributing to primary productivity in aquatic environments since they contain chlorophyll as an absolute requirement for the process of photosynthesis (Wibowo *et al.*, 2004; Padang, 2012; Latuconsina, 2020; Padang *et al.*, 2021). Diatoms are one of the benthic microalgae that form assemblages on sediments and other hard substrates known as microphytobenthos, and they play an important role in the biogeochemical cycles of carbon, nitrogen, phosphorus and silicon (Wilhelm *et al.*, 2006). Although often overlooked, benthic diatoms are a key component in the primary productivity in any aquatic habitat (Karsten *et al.*, 2021). The high diversity of benthic diatoms supports high primary productivity, and the diversity of benthic diatoms is supported by the content of organic matter in the sediments in addition to other environmental factors (Virta *et al.*, 2019).

Diatoms/ Bacillariophyceae also play a radical role as a natural feed for benthic organisms such as sea cucumbers (Lalli & Parsons, 1997; Huliselan *et al.*, 2006; Nontji, 2008; Padang *et al.*, 2014a). Sea cucumbers (Holothuridae) as benthic organisms by means of deposit feeding consume benthic phytoplankton and zooplankton stored in the sediment substrate (Xie *et al.*, 2016; Jiang *et al.*, 2020). Sea cucumbers (Holothuridae) are highly nutritious fishery commodity and hold important commercial value (Padang *et al.*, 2015). High market demand and uncontrolled exploitation of sea cucumbers are problems that must be overcome, one of which is with the pen culture method (Padang *et al.*, 2016), using the floating net cage method (Padang *et al.*, 2017), or through the development of marine farms (Hartati *et al.*, 2021).

The basic principle of ecology in the sustainable development of aquaculture is the effort to maintain the carrying capacity of the environment to ensure a positive reciprocal relationship between environmental factors and aquaculture commodities through the support of optimal water environmental quality (Latuconsina, 2020). Sea cucumbers play an important role in improving the environmental quality of marine waters, with the aim of being developed in aquaculture to improve environmental quality (Isgoren-Emiroglu & Gunay, 2007). Diatoms as a natural food for sea cucumbers (Holothuridae) are effective in reducing total nitrogen and phosphorus in the water column, thereby improving water quality to support the environmental carrying capacity for aquatic biota (Jiang *et al.*, 2020).

Sediment as a part of the container for rearing benthic organisms allows the growth of benthic diatoms as a natural feed for sea cucumbers. Based on the studies of **Little** (2000) and Widianingsih *et al.* (2009), Bacillariophyceae/Diatom have special attachment tools to the substrate, therefore they are quite abundant in sediments. Xie *et al.* (2016) reported that the benthic fiatom Navicula sp. can increase growth performance and digestive enzyme activity as well as immunity in the early growth of sea cucumber (*Apostichopus japonicus*). Jiang *et al.* (2020) reported that diatom Nitzschia sp. can grow and effectively utilize the nutrients released by the decomposition of macroalgae carriers,

with the aim of being applied as a natural feed for juvenile sea cucumber (*Stichopus japonicus*).

Sago waste, chicken manure and seagrass leaves can be used as compost to support the growth of benthic microalgae. Sago waste is generally used as an organic fertilizer since it contains carbon (C), nitrogen (N), phosphorus (P), pottasium (K) and the C/N ratio to support the growth of plant (Wahida & Limbingan, 2015; Zaimah & Prihastanti, 2015). Chicken manure is used as compost since it has a better nutrient content compared to other livestock manure (Utama & Jannah, 2014), while seagrass leaf litter is a source of nutrients (Liu *et al.*, 2018; Prasad *et al.*, 2018). It has been used as a liquid organic fertilizer (Dewi *et al.*, 2016).

Scientific information regarding the use of sago waste, chicken manure and seagrass leaves to stimulate the growth of benthic diatoms as a natural feed for sea cucumbers (Holothuridae) is limited, therefore this research was important to determine the composition and density of benthic diatoms as a natural feed for sea cucumbers, which are deposited in the sediment of rearing containers by providing different feed compositions in the form of sago west, chicken manure and seagrass leaves. The output of this research can be used as scientific information that contributes to efforts to develop environmentally friendly and sustainable sea cucumber cultivation.

MATERIALS AND METHODS

1. Study area

This research was conducted in the coastal waters of Hunut Village, the Inner Ambon Bay, Maluku - Indonesia, with the geographical position of floating net cages located at 338"6.0036" S and 12812'52.164" E. Map of research locations and method of placing sea cucumber containers for rearing in floating net cages are shown in Fig. (1).



Fig. 1. Map of the research location marked with a red spot (left), and the container placed for raising sea cucumbers into a sack and hanging it on the floating net cage box (right)

The HDPE type floating marine cages used belong to the Marine Fisheries and Cultivation Center of Ambon which are placed in the waters of the Inner Ambon Bay in the amount of 1 unit totaling 4 boxes, with each measuring $3 \times 3 \times 3m$. The floating net cages are used in the cultivation of several marine fishes commodities, namely; Grouper (Serranidae), Trevally (Carangidae), and Barramundi (Centropomidae). Floating net cages can also be used to develop sea cucumber cultivation. Sea cucumber (*Holothuria scabra*) cultivated in floating net cages is placed in a rearing container in the form of a serving lid measuring 60 x 45 x 15cm and covered with a net and tied at the top to facilitate the cultivation of sea cucumber and avoid coming out of the rearing container. Then, sea cucumber samples were placed in floating net cages in a suspended position. The hanging depth of the sea cucumber rearing container is 2m from the surface of the water (Fig. 2).



Fig. 2. a. Rearing container, b. Treatment Composition, and c. Placement of sea cucumber rearing containers using the floating method

2. Collection and identification process of benthic diatom samples

- In the floating net cage box, the sea cucumber (*Holothuria scabra*) rearing container was placed by providing sediment in the rearing container with three treatments namely: treatment A was given seagrass leaves (*Enhalus acroides*) + chicken manure, treatment B was given seagrass leaves (*E. acroides*) + sago waste, and treatment C was given seagrass leaves (*E. acroides*) + chicken manure + sago waste.
- Benthic diatom samples were taken using a sediment core with a diameter of 2cm.
- The sediment sample was then put into a container and 4% formalin which had been hardened with the addition of borax for further analysis in the laboratory.
- Benthic diatoms bound to sediments were separated using a 300 clements brand orbital centrifuge with a rotation of 5000rpm.
- Then using a pipette, the raised/ separated benthic diatoms are taken and separated into containers. Separation by centrifuge was carried out four times, assuming that all benthic diatoms had been removed. Then, the samples were deposited for 24 hours.
- Benthic diatoms that had been precipitated for 24 hours were diluted 10 times the volume of the precipitate. With a 1ml pipette, the sub sample was taken and placed on the haemocytometer to be observed (**Mujiman, 1984; Nontji, 2008**).
- Identification of benthic diatoms was carried out under a NIKON SF type microscope with 400 times magnification and three replicate observations, based on

the outlines of Jorgensen (1905), Van Heurck (1909), Yamaji (1966) and Tomas (1997).

• Identification of diatom species was directly carried out without removing plastids from the diatom part.

3. Data analysis

The composition species of benthic diatom species in sediments was calculated using the formula according to the method of **Fachrul (2007)**, as follows:

Species composition (%) = $\frac{\sum \text{ individual of a species}}{\sum \text{ individuals of all species}} x 100$

The density of benthic diatoms in the sediment was calculated using the formula according to the method of **Khouw (2008)**, as follows:

$$D = \frac{q}{r^2 x t}$$

Where, D = Benthic diatom density (cell/ cm³); q = Number of individual (cell); value constant 3,14; r = Sediment core radius (1cm), and t = Sediment layer thickness (2cm)

RESULTS

1. Composition of benthic diatom species in floating net cages

Two orders, 14 families, 25 genera, and 66 species of benthic diatoms were found in the floating net cage sediments in the rearing containers of sea cucumbers (Table 1). They were dominated by the order Pennales with eight families, 17 genera and 51 species, while the order of Centrales was assessed to consist of six families, eight genera, and 15 species. The large number of order Pennales in sediments is due to the morphological shape deposited easily in sediments.

Onden	Family	Constant of	Photo-	Treatment		
Order		Species	micrographs	Α	B	С
Pennales	Ashnonthosoo	<i>Achnanthes lanceolata</i> Lange-Bertalot, 1993	O	+	+	+
	Acmantiaceae	<i>Achnanthidium microcephalum</i> Kützing, 1844	M.	+	+	+

Table 1. Composition of benthic diatoms found in rearing containers of sea cucumbers

		Achnanthes longipes C.Agardh, 1824	B	-	+	+
		<i>Cocconeis molesta</i> Kützing, 1884		-	-	+
		<i>Cocconeis scutellum</i> Ehrenberg, 1838	0.	-	+	+
		Amphiprora gigantea Grunow, 1860		+	-	+
		Amphora veneta var.subcapitata Kisselew, 1932	D	+	+	+
	Cymbellaceae	<i>Amphora quadrata</i> Brébisson ex Kützing, 1849		+	+	+
		<i>Amphora ovalis</i> Kützing, 1844	0	-	-	+
		<i>Amphora laevis</i> Gregory, 1857	J.	+	+	+
		<i>Epithemia sorex</i> Kützing, 1844		+	+	+
	Fragilariaceae	<i>Epithemia musculus</i> Kütz. 1844	and the second	+	-	+
		<i>Cymbella ehrenbergii</i> Kützing, 1844		-	+	+
		Fragilaria brevistriata Grunow, 1885		-	+	+
		Fragilaria crotonensis Kitton, 1869	B.	+	+	+

	<i>Fragilaria cylindrus</i> Grunow, 1883		+	+	+
Licmophoraceae	<i>Licmophora dalmatica</i> Grunow, 1867		+	-	-
	Diploneis fusca var. pelagi Cleve, 1894	Ø	+	+	+
	Diploneis splendida Cleve, 1894	B.	+	+	+
	<i>Navicula pusilla</i> W.Smith, 1853		+	+	+
	<i>Navicula concellata</i> Donkin, 1872	Q.	+	+	+
	<i>Navicula elegan</i> W. Smith, 1853	(SSE)	+	+	-
	<i>Navicula complanata</i> Grunow, 1880	C.	-	-	+
	<i>Navicula graniii</i> (Jørgensen) Gran, 1908	and the second second	+	+	+
Naviculaceae	<i>Navicula vanhoeffenii</i> Gran, 1897	the second	+	-	-
	<i>Navicula amphisbaena</i> Bory de Saint Vincent,1824	0	-	-	+
	Navicula directa var.subtilis (Gregory) Lceve, 1883		+	+	+
	Navicula transitrans var. derasa Cleve, 1883	0	÷	+	+

	<i>Navicula tuscula</i> Ehrenberg, 1840	(D)	+	-	+
	<i>Navicula delicatula</i> Cleve, 1894	ſ.	+	+	+
	<i>Navicula distans</i> (W.Smith) Ralfs, 1861	S.	+	+	-
	<i>Navicula mutica</i> Kützing, 1844	0	-	+	-
	Pleurosigma elongatum W.Smith, 1852	•	+	+	+
	<i>Pleurosigma rectum</i> Donkin, 1858	0	-	+	+
	<i>Nitzschia navicularis</i> (Brébisson) Grunow, 1880		+	+	+
	<i>Nitzschia constricta</i> Grunow, 1880	(I)	-	-	+
	<i>Nitzschia panduriformis</i> W.Gregory, 1857	(Mar)	+	+	-
Nitzschiaceae	<i>Nitzschia fasciculata</i> Grunow, 1881	A	+	+	+
	<i>Nitzschia dissipata</i> Grunow, 1862	all	-	+	-
	<i>Nitszchia sigma var rigida</i> Grunow ex Van Heurck, 1880	1 de la compañía de	+	+	+

		<i>Nitszchia acicularis</i> W.Smith, 1853	20	+	+	+
		<i>Nitszchia angularis</i> W.Smith, 1853		+	+	-
		<i>Nitszchia obtusa var. nana</i> Grunow, 1885		+	+	+
		<i>Nitzschia linearis var. subtilis</i> (Grunow) Hustedt, 1923	-1	-	-	+
		<i>Nitszchia obtusa</i> W.Smith, 1853	1.	+	-	+
		<i>Nitzschia hungarica</i> Grunow, 1862	P	+	+	-
		<i>Champylodiscus taeniatus</i> Schmidt in Schmidt et al., 1875		+	+	+
	Surirellaceae	<i>Cymatopleura eliptica</i> W.Smith, 1851		+	+	+
		<i>Gramatophora marina</i> Kützing, 1844		-	+	-
	Tabellariceae	Tabellaria flocculosa Kütz, 1844	A.C.	-	-	+
		<i>Tetracyclus rupestris</i> Grunow in Van Heurk, 1885	O.	-	-	+
Centrales	Biddulphiaceae	<i>Biddulphia reticulata</i> Roper, 1859		-	-	+
		Biddulphia sinensis Greville, 1866	1	-	+	-
		Chaetoceros pendulus Karsten, 1905	>	-	+	-
		<i>Chaetoceros skeleton</i> F.Schütt, 1895		-	+	-

		Chaetoceros teres Clee, 1896	C. Sec.	-	-	+
	Coscinodiscacea	<i>Actinocyclus normanii</i> Hustedt, 1957	\bigcirc	+	-	-
		<i>Coscinodiscus denarius</i> A.Schmidt, 1878		+	+	+
		Coscinodiscus centralis Ehrenberg, 1844	8 8 8	+	+	+
		<i>Thalasiossira condensata</i> Cleve, 1900	0	+	+	+
	Corethraceae	<i>Corethron valdiviae</i> Karsten, 1904		-	-	+
		<i>Melosira nummuloides</i> C.Agardh, 1824		+	+	+
	Malacinagaa	<i>Melosira arenaria</i> Moore ex Ralfs, 1843	8	+	+	+
	Melosiraceae	<i>Melosira borreri</i> Greville, 1833	Ô	+	+	+
		<i>Melosira roeseana</i> Rabenhorst, 1854	0	-	+	+
	Skeletonemataceae	Skeletonem mediterraneum var. punctifera Brun, 1891		-	+	-

Source: Primary Data Analysis (2016)

Treatment: A = seagrass leaves + chicken manure,

 $B = seagrass \ leaves + sago \ waste,$

 $C = seagrass \ leaves + chicken \ manure + sago \ waste.$

(+) = Found, (-) = Not Found.

Figs. (2, 3, 4) show the composition of the benthic diatom species found in the three treatments. A high composition of the genera Navicula was found in the sediments of sea cucumber (*Holothoria scabra*) rearing containers.



Fig. 2. Composition of benthic diatoms in treatment A = seagrass leaves (*E. acoroides*) + chicken manure



Fig. 3. Composition of benthic diatoms in treatment B = seagrass leaves (*E. acoroides*) + sago waste



Fig. 4. Composition of benthic diatoms in treatment C = seagrass leaves (*E. acoroides*) + chicken manure + sago waste

2. Benthic diatom density in sediments

The density of benthic diatoms in the sediment of sea cucumber (*Holothoria scabra*) rearing containers with different compost applications using the floating net cage cultivation method (Figs. 5, 6, 7).



Fig. 5. Benthic diatom density in treatment A = seagrass leaves + chicken manure



Fig. 6. Benthic diatom density in the treatment of seagrass leaves + sago waste



Fig. 7. Benthic diatom density in treatment of seagrass leaves + sago waste + chicken manure

DISCUSSION

The benthic diatoms family with the highest number of genera and species is Naviculaceae, which includes 3 genera and 17 species, and the genera with the highest number of species is Navicula with 13 species (Table 1). The genera Navicula is also commonly found as periphytic plankton in the Kapuas River (Apriliani *et al.*, 2018). The large number of Navicula genera found is attributed to its wide distribution of habitats and the large number of species. According to Kociolek *et al.* (2015), members of the Naviculaceae family can be found in various types of habitats, such as freshwater, estuaries, swamps and marine waters.

The genera that dominated the three treatments was Navicula with 13 species: 10 species in treatment A, 9 species in treatment B, and 9 species in treatment C (Table 1). It turned out that the container with sediment in treatment C recorded a higher number of species, with 51 species of benthic diatoms. The high composition of benthic diatoms in treatment C is possible since the three types of compost given have good nutritional content to support the growth of benthic diatoms, namely chicken manure as manure contains good nutrients (**Utama & Jannah, 2014**), sago waste, which has several important substances for plant growth (**Wahida & Limbingan, 2015; Zaimah & Prihastanti, 2015**), and seagrass leaves (*Enhalus acroides*) with their high nutrition, including 12.11% of protein, 27.23% of ash, 3.30% of carbohydrates, 2.61% of fat, 79.69% of water, and 54.75% of fiber (**Latuconsina** *et al.*, **2021**). In seagrass leaves (*E. acroides*), there are also numerous epiphytic benthic diatoms (**Devayani** *et al.*, **2019**).

The composition of the benthic diatoms in the three treatment (Figs. 2, 3 & 4) belongs to the same genera, namely the genera Achnanthes, Amphora, Campylodiscus, Coscinodiscus, Cymatopleura, Diploneis, Epithemia, Fragilaria, Navicula, Nitzschia, Pleurosigma, and Thalassiosira. While, the genera that were only present in each treatment were the genera Actinocyclus and Licmophora (treatment A), the genera Grammatophora and Skeletonema (treatment B), and the genera Corethron, Tablelaria and Tetracylus (treatment C). Navicula and Nitzschia are benthic diatom genera which are always dominant (**Obal** *et al.*, **1989; Rusmiati** *et al.*, **2019**). The genera found in each treatment had fewer species and fewer cells than the genera found in all treatments. It is assumed that the three treatments have the same effect on the composition genera of the benthic diatom, and are able to adapt well to the aquatic environment, even though there are differences in compost content affecting the nutrient content in the rearing containers.

The adaptability of benthic diatoms to the aquatic environment is very large, the class Bacillariophyceae is abundant in the waters due to its ability to adapt to the environment as a cosmopolitan being resistant to extreme conditions & sensitive to environmental changes, therefore it can be used as a bioindicator of the water quality resulting in a high reproductivity (Hwang *et al.*, 2011; Ariana *et al.*, 2014; Teittinen, 2016; Afiah *et al.*, 2021). Nybakken (2005), in his study revealed that, the phytoplanktons from the class Bacillariophyceae have a very fast response to the addition of nutrients and are able to adapt to the environment in which they live compared to genera from other classes. In this context, Aprilliani *et al.* (2018) suggested that living diatoms attach to a substrate and depend on environmental conditions in the aquatic ecosystems.

Benthic diatoms as benthic phytoplankton are an important component of primary productivity in the benthic environment, apart from cyanobacteria and muticellular algae (Cahoon & Safi, 2002; Suwartimah *et al.*, 2011). The growth of sea cucumbers in floating net cage cultivation can be influenced by benthic diatoms, which are important benthic organisms. As reported by **Padang and Subiyanto** (2019), who obtained the feed composition of sea cucumbers reared in floating net cages with three feed treatments, it turned out to be dominated by the diatom or Baccilariophyceae of phytoplankton (113 species), in addition to the classes Chlorophyceae, Cyanophyceae, and Dinophyceae. The utilization of benthic diatoms by sea cucumbers reared in floating net cages with the treatment of seagrass leaves, sago waste and chicken manure is quite large at 95.09-99.38% (Padang & Subiyanto, 2019). Benthic diatoms in the stomachs of sea cucumbers that live free in nature represent 56% (Padang *et al.*, 2014a), while those reared in solitary confinement are 89% (Padang *et al.*, 2016).

Sea cucumbers (Holothoridae) reared in floating net cages use benthic diatoms as food, as well as sea cucumbers reared in pen culture (**Padang** *et al.*, **2016**). For the composition of food species, Bacillariophyceae from the diatom family is the type of food that has the highest value compared to other types of food for sea cucumbers (**Agusta** *et al.*, **2012**). Benthic diatoms are used as food for aquatic biota, especially benthic biota not only found in sea cucumbers, but also in the rearing of other benthic diatom species in collectors and abalone leg muscles of 98% (**Israwati** *et al.*, **2018**). Furthermore, **Setyono** (**2005**) obtained juvenil larvae of abalone (*Haliotis asinina*) which were fed diatoms of the Nitzchia spp., which recorded a shell growth of about 0.5mm in 2 weeks and 1.5mm in 2 months. The benthic diatom types of Navicula sp. and Nitzschia sp. represent a type of diatom that is proven to be able to support the survival and growth of juveniles of several species of abalone (**Hamid** *et al.*, **2017**). Benthic diatoms of the type Navicula sp., also have a good effect on growth and immunity for the juvenile phase of the sea cucumber *Apostichopus japonicas* (**Xie** *et al.*, **2016**).

Benthic diatoms, generally of the order Pennales, have a wide distribution, and apart from being in the benthic environment, they are also abundant in the pelagic environment. Diatoms have a wide distribution worldwide and respond quickly to changes in the physical and chemical properties of water (Kashima, 2008). Bracher *et al.* (2008) reported that Cyanobacteria and diatoms dominate the composition of phytoplankton in marine waters, affecting about 40% of primary productivity in marine waters and making diatoms the microalgae with the highest abundance. Diatoms are the dominant microalgae in all aquatic ecosystems, and they play an significant role in the carbon cycle, food webs and natural primary productivity (Sabanci, 2012; Soeprobowati *et al.*, 2012).

Diatoms are ecologically fundamental for detecting environmental conditions and water quality (Solak, 2011; Oguz *et al.*, 2020). Diatoms can respond to changes in the environment since they are well stored in sediments (Reid & Ogden, 2009; Soeprobowati *et al.*, 2016; 2018). Moreover, diatoms can be used as bioindicators of pollution since they respond to physical and chemical changes in the environment (Suwartimah *et al.*, 2011; Tan, 2017; Ananingtyas *et al.*, 2018; Chonova, 2018; Ario *et al.*, 2019).

In addition to giving sago waste, seagrass leaves (*Enhalus acroides*) were also given to the seagrass container, given that benthic diatoms were found living as epiphytes on seagrass leaves (**Padang, 2011b; Ario** *et al.,* **2019**). In this respect, **Akbar** *et al.* (**2020**) detected an epiphytic biota that lives in association with seagrass by attaching to seagrass rhizomes, stems and leaves. According to **Suwartimah** *et al.* (**2011**), Bacillariophyceae play an basic role in waters, dominating in terms of both number and type of other phytoplankton. Given the study of **Ario** *et al.* (2019), Bacillariophyceae or better known as diatom is the most common type of microalgae periphyton found with one cell although some of them are in the form of colonies.

On the other hand, seagrass leaf litter enriches the nutrient content in the sediment, stimulating the growth of benthic diatoms. According to **Dewi** *et al.* (2016), seagrass litter contains various nutrients that are useful for plant growth and has the potential to be used as a liquid organic fertilizer since it contains elements of nitrogen, phosphorus and potassium. Naturally, the concentration of nutrients in water varies for each form of compound, including nitrate and phosphate. **Tampubolon** *et al.* (2020) stated that, seagrass plays an important role in the contribution of nutrients (phosphate and nitrate) in a suspended or dissolved form in the waters from the decomposition of seagrass litter by decomposing microorganisms. While, **Prasad** *et al.* (2018) found a large contribution from the decomposition of seagrass litter in producing high organic matter, serving as a nutrient supplier for coastal waters.

Irawan (2017) suggested that seagrass leaf litter will be used by aquatic biota as food or settles into the substrate. Litter that settles in the substrate can be utilized by benthic organisms and benthic diatoms. **Prakoso** *et al.* (2015) stated that epiphytes utilize seagrass leaves as a habitat and also utilize nutrients from seagrass litter as food, thus epiphytes are closely related to seagrass as a place to shelter, find food, grow and develop.

Figs. (5, 6 & 7) show the different densities of benthic diatoms between treatments. However, the highest density of benthic diatoms included the genera of Navicula which had the highest density found in the treatment of seagrass leaves + sago waste + chicken manure. The high density of benthic diatoms in the treatment of seagrass leaves + sago waste + chicken manure is thought to be closely related to the higher nutrient content contained in seagrass leaves, sago waste and chicken manure. Liu et al. (2018) found that the leaf litter of *Enhalus acoroides* seagrass produced an average of 22% of carbon (C), 70% of nitrogen (N), and 38% of phosphorus (P). The average dissolved organic carbon (DOC), dissolved organic nitrogen (DON), and dissolved organic phosphorus (DOP) contributed 55, 95, and 65% of the total C, N and P. This means that seagrass leaf litter produces nutrients that contribute to support the growth of benthic diatoms. According to Astiani et al. (2016), the nutrient content found in chicken manure is 3.77mg/1 (PO₄) and 0.67 (NO₃). Sago waste contains 0.23% of N, 0.04% of P, and 0.52% of K (Tando & Assad, 2018). Sago waste contains carbon (C), nitrogen (N), phosphorus (P) and potassium (K) to support plant growth, aiming to be generally used as an organic fertilizer (Wahida & Limbingan, 2015; Zaimah & Prihastanti, 2015). According to Riaza (2019), sago waste has a crude fiber content of 30.14 and 4.37% of crude protein. Additionally, sago waste also contains 20 and 21% cellulose and lignin, respectively (Kiat, 2006).

The genera of Navicula and Nitzschia, which were dominant in the three treatments, were benthic diatoms from the order Pennales which are widely distributed in waters. **Kilinc and Sivaci (2001)** found Navicula and Nitzschia as the dominant genera in the waters. **Sullivan and Currin (2000)** and **Padang** *et al.* **(2014b)** found that genera of diatoms in the waters with high salinity and in sediments, were those of Navicula, Nitzschia, and Amphora. The density of benthic diatoms is more dominated by the order Pennales or Pennate diatoms than the order Centrales or Centric diatoms. **Mitbavkar and Anil (2002)** and **Suwartimah** *et al.* **(2011)** stated that diatoms of Pennales are generally

found more frequently in the sediments and are benthic, whereas diatoms of Centrales are generally found floating in the water column. The order Pennales is a benthic diatom which has an important role in the productivity in the waters of the benthic environment.

Wibowo *et al.* (2004), Suwartimah *et al.* (2011) and Padang *et al.* (2021) stated that the Pennales order is the primary producer in the life of benthic organisms since it is one of the microalgae that contains chlorophyll.

The genus Nitzschia has a solitary life and is widely distributed in estuarine, fresh waters, marine, and has a motile nature in the substrate and all waters (sublittoral to littoral) with a wide distribution and is able to adapt to high environmental changes (**Suwartimah** *et al.*, **2011**). The habitat of the Navicula genus is widespread or cosmopolitan in all waters, it is also commonly found in benthic waters and has a high tolerance for changes in water quality (**Kurnia & Panjaitan, 2020**). Navicula and Amphora are genera that permanently exist in sediments (**Mitbavkar & Anil, 2002**).

In addition to the order Pennales, order Centrales was also found with a high number of densities, namely the genus Coscinodiscus. This is made possible due to the movement of water and the position of the rearing containers which are placed in floating net cages by hanging them in the middle of the sea. This situation is reinforced by the statement of **Suwartimah** *et al.* (2011) that order Centrales is found in the distribution of benthic plankton, which is thought to be caused by the turbulence of water and tidal currents. Similarly, **Cahoon and Safi** (2002) explained that the distribution of diatoms is influenced by the turbulence of water and tides attributed to providing opportunities for phytoplankton to live freely in benthic communities. The influence of tides affects the distribution of benthic diatoms, as explained by **Cahoon and Safi** (2002) that some planktonic diatoms will move to the surface of the substrate when conditions recede.

The sea cucumber (*Holothuria scabra*) as a benthic organism can utilize the availability of benthic diatoms deposited in sediments as food (**Sembiring et al., 2015**; **Nursid, 2019; Padang et al., 2021**). Sea cucumbers are organisms that are non-selective, meaning they don't choose the type of food they eat (**Padang et al., 2021**). Several studies have found that the stomach contents of sea cucumbers have varying selectivity values, thus showing that sea cucumbers, by means of deposit feeding, filter organic particles stored in sediments, namely plankton. Agusta et al. (2012) discovered that, in addition to plankton, the digestive tracts of sea cucumbers *H. hilla, H. leucospilota,* and *H. impatiens* contained sand, which constituted a significant percentage. Sea cucumbers do not consume sand but utilize bacteria associated with organic detritus extracted from various substrates. The sand serves only as a medium for the entry of the food contained within.

Fig. (7) shows the high diversity and density of benthic diatom cells in sediment treated with seagrass leaves, chicken manure and sago waste. This indicates that the high nutrient content will contribute to the high diversity and density of benthic diatoms. **Virta** *et al.* (2019) found that the diversity of benthic diatom communities was mainly caused by variations in the organic matter content in the sediments. **Dalu** *et al.* (2015) found that the diatom communities were determined by a variety of factors, including nutrient concentrations (ammonia, nitrate), hydrology (eg, water depth and flow) and pH. Furthermore, chicken manure is generally used as compost since it has a better nutrient content compared to other livestock manure (**Utama & Jannah, 2014**).

The results of this study found that treatment with a more diverse compost composition was able to support the high number and density of benthic diatoms since it was suspected that they had a higher nutrient content.

CONCLUSION

A total of two orders, 14 families, 25 genera, and 66 species of benthic diatoms were found in the sediments of sea cucumber (*Holothoria scabra*) rearing containers with different compost applications using the floating net cage cultivation method. Different feed compositions contributed to the density and composition and number of benthic diatom species, with genera composition and higher diatom densities in the treatment of seagrass leaves + sago waste + chicken manure. The benthic diatoms found are dominated by the order Pennales, and particularly the different species of the genus Navicula. The results of this study provide important information to utilize sago waste + chicken manure + seagrass leaves as an alternative compost to stimulate the growth of benthic diatoms as a natural feed for sea cucumber (Holothuridae) cultivation. Further research is needed regarding the survival and growth rate of sea cucumbers and their relationship with the physicochemical parameters of sediment with the addition of seagrass leaves and chicken manure as natural compost. This information could be valuable for optimizing the cultivation of sea cucumbers (Holothuridae) using the floating net cage method, particularly in polyculture systems alongside marine fish cultivation.

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