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Application of Shelter as Feeding Plate with Different Combination of Diatom Species in the Rearing of Juvenile Sea Cucumber, *Stichopus horrens*

(Penggunaan Tempat Perlindungan sebagai Pinggan Suapan dengan Gabungan Berbeza Spesies Diatom dalam Penternakan Juvenil Gamat, *Stichopus horrens*)

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ABSTRACT

The cultivation of *Stichopus horrens,* a sea cucumber with high economic value, requires further development. This study investigates the effect of incorporating shelters that function as feeding plates on the growth and survival of juvenile sea cucumbers by enhancing food availability and accessibility. The study was conducted for two months using 12 rectangular polycarbonate tanks in a completely randomized design with four treatments and three replications. The experiment consisted of the treatments: (A) feeding plate containing *Chaetoceros muelleri* and *Navicula* sp.; (B) feeding plate containing *C. muelleri* and *Nitzschia* sp.; (C) feeding plate containing *C. muelleri, Navicula* sp., and *Nitzschia* sp.; and (D) no feeding plate, but all three diatom species were added directly to the rearing tank. Each tank was stocked with 100 juveniles with an average length and weight of 0.64 ± 0.14 cm and 0.06 ± 0.01 g, respectively. Parameters observed included growth, survival, grazing on the feeding plate, and digestive enzyme activity. The growth of juveniles in treatments A and C was higher and significantly different from treatments B and D ($p<0.05$). The highest survival was obtained in treatment C of 67.0 ± 7.02%, significantly different from treatments B ($p < 0.05$). The results of the present study indicate that the growth of sea cucumber juveniles is enhanced under rearing conditions with the addition of shelters. Furthermore, the presence of *Navicula* sp. significantly accelerates both growth and survival rates in juveniles.

Keywords: Biofilms; clear zone; enzyme activity; growth; survival

ABSTRAK

Kultivasi *Stichopus horrens* iaitu gamat yang mempunyai nilai ekonomi tinggi memerlukan pembangunan lanjut. Penyelidikan ini mengkaji kesan penggabungan tempat perlindungan yang berfungsi sebagai pinggan suapan ke atas tumbesaran dan kemandirian gamat juvenil dengan meningkatkan ketersediaan dan kebolehcapaian makanan. Penyelidikan telah dijalankan selama dua bulan menggunakan 12 tangki polikarbonat segi empat tepat dalam reka bentuk rawak sepenuhnya dengan empat rawatan dan tiga replikasi. Uji kaji ini terdiri daripada rawatan: (A) pinggan suapan yang mengandungi *Chaetoceros muelleri* dan *Navicula* sp.; (B) pinggan suapan yang mengandungi *C. muelleri* dan *Nitzschia* sp.; (C) pinggan suapan yang mengandungi *C. muelleri, Navicula* sp. dan *Nitzschia* sp.; serta (D) tiada pinggan suapan, tetapi ketiga-tiga spesies diatom telah ditambah terus ke tangki pemeliharaan. Setiap tangki diisi dengan 100 juvenil dengan purata panjang dan berat masing-masing 0.64 ± 0.14 cm dan 0.06 ± 0.01 g. Parameter yang diperhatikan termasuk tumbesaran, kemandirian, ragut pinggan suapan dan aktiviti enzim pencernaan. Tumbesaran juvenil dalam rawatan A dan C adalah lebih tinggi dan berbeza secara signifikan daripada rawatan B dan D ($p<0.05$). Kemandirian tertinggi diperoleh dalam rawatan C sebanyak 67.0 ± 7.02%, berbeza dengan ketara daripada rawatan B ($p < 0.05$). Keputusan kajian ini menunjukkan bahawa tumbesaran juvenil gamat dipertingkatkan dalam keadaan pemeliharaan dengan penambahan tempat perlindungan. Tambahan pula, kehadiran *Navicula* sp. dengan ketara mempercepatkan kedua-dua kadar tumbesaran dan kelangsungan hidup juvenil.

Kata kunci: Aktiviti enzim; biofilem; kelangsungan hidup; tumbesaran; zon jelas

INTRODUCTION

The high demand for sea cucumber products in the international market has led to the overexploitation of many economically important species. One of the high-value tropical sea cucumber species exploited is *Stichopus horrens* (Ibañez et al. 2023). It is widely distributed in the Western Indian and Pacific Oceans, including the Galapagos and Hawaii, USA (Hearn & Pinillos 2006; Purcell, Samyn & Conand 2012).

Stichopus horrens is a species of sea cucumber that has been widely used as a raw material in the nutraceutical and pharmaceutical industries (Bordbar, Anwar & Saari 2011; Eriksson et al. 2007). The bioactivity of sea cucumbers has also been tested and utilized in the medical field (Masre et al. 2010; Sarhadizadeh, Afkhami & Ehsanpour 2014; Setianingsih, Putri & Mutiadesi 2020). The demand for sea cucumbers as raw materials for medicines continues to increase, causing natural overexploitation (Pringgenies, Siti & Ervia 2017). Therefore, sea cucumber cultivation must be initiated to overcome overexploitation and maintain its population in nature. The availability of juvenile sea cucumbers from hatcheries strongly influences the sustainability of aquaculture. Research on sea cucumber hatcheries has begun to be carried out and provides hope for success to continue to be developed (Hu et al. 2013). However, some obstacles persist, particularly during the initial stage of juvenile rearing in controlled tanks. The survival rate obtained at this stage is very low (0.05-0.13%), and the growth rate varies greatly (Sembiring et al. 2025).

Juveniles at this stage live attached to the walls of the rearing tanks and eat organisms/plankton attached to the walls. A potential approach to enhance juvenile survival during this developmental stage is the implementation of shelters that concurrently serve as attachment substrates and feeding surfaces for sea cucumber juveniles. Juvenile sea cucumbers in early developmental stages feed on benthic diatoms attached to shelters or feeding plates (Hu et al. 2013), which are considered their primary food source (Xie et al. 2017). With the addition of shelters in the rearing tanks, a wider area will be available as a place to attach sea cucumber juveniles apart from the walls of the rearing tanks. Reeves et al. (2020) and Wang et al. (2022) stated that shelter plays an important role in the adaptation stage of marine organisms in nature. Furthermore, Weiss, Lozano-Alvarez and Briones-Fourzan (2008) and Yanagisawa (1998) reported that abundant shelter availability is conducive to increasing the survival of marine organisms. Other studies indicate that the incorporation of shelters in the rearing systems of sea cucumber *Apostichopus japonicus* significantly influences their survival rates and feeding behavior (Tian et al. 2023; Xia et al. 2012; Yu et al. 2022). The presence of biofilm on shelter surfaces can induce settlement in sea cucumber larvae (Pearce & Scheibling 1990). Juvenile *A. japonicus* preferentially settles on biofilm-coated surfaces (Dong et al. 2010), which also serves as a food source (Liu et al. 2017; Zhang et al. 2015).

The current study used plastic sheets as a feeding plate for the nursery rearing of *S. horrens*. A mixture of benthic microalgae, including *Chaetoceros muelleri, Nitzschia* sp., and *Navicula* sp., was provided as a food source. This research aims to increase the survival and growth of sea cucumber juveniles by adding shelters as an optimum food supply.

MATERIALS AND METHODS

JUVENILE SEA CUCUMBER

The sea cucumber juveniles used in this study were hatched and reared at the Marine Biota Scientific Conservation Area (KKI) hatchery, Gondol-Bali. The juveniles used were the result of spawning and were 45 days old. A total of 1200 juveniles were used with a total length of 0.64 ± 0.14 cm and an average weight of 0.06 ± 0.01 g.

FEEDING PLATE PREPARATION

The feeding plates were made of wavy plastic and measured 30 cm in height and 29 cm in length (Figure 1). A total of 21 feeding plates were used in this experiment. Three species of diatoms, *C. muelleri, Nitzschia* sp., and *Navicula* sp., were used as natural food for juvenile sea cucumbers. Diatoms were cultured in 30 L plastic jars in a controlled manner in the laboratory. Before being placed in the diatom culture container, the feeding plate was sterilized by washing it with detergent, rinsing it with fresh water, and drying it. After the water for the diatom culture was prepared, fertilizer was applied, and inoculants of each diatom species were added. Next, the shelter was inserted in the culture container so the diatoms could attach to the feeding plate. Five days after the culture, the feeding plate was removed and placed in the juvenile rearing tank (Figure 2). The density of diatoms on the feeding plate was calculated using the transect method before being placed in the juvenile rearing tanks. The feeding plate is prepared every 3 days so that the change of the feeding plate in the juvenile rearing tank can be done 2 times a week.

EXPERIMENTAL DESIGN

The treatments in this study were as follows: A) feeding plate with *C. muelleri* and *Navicula* sp.; B) feeding plate with *C. muelleri* and *Nitzschia* sp.; C) feeding plate with *C. muelleri, Navicula* sp., and *Nitzschia* sp.; and D) without a feeding plate. In addition to the diatoms provided on the feeding plates, the same species of diatom was also introduced into their corresponding rearing tanks. The addition was carried out until each diatom species reached a final density of 10,000 cells/mL in the rearing tanks. Each treatment consisted of 3 replicates. The number of feeding plates in each rearing tank was 2 for treatments A and B,

FIGURE 1. The feeding plate is transparent wavy plastic measuring (H × L: 30 × 29 cm) and arranged vertically

FIGURE 2. Feeding plates were put into each diatom culture, and on the fifth day of culture, the feeding plates were removed and were ready to be transferred to the juvenile rearing tank

and 3 for treatment C. In contrast, treatment D received a daily addition of diatoms containing *C. muelleri*, *Navicula* sp., and *Nitzschia* sp., with each species adjusted to a final density of 10,000 cells/mL. No feeding plate was provided in this treatment.

REARING OF *S. horrens* JUVENILE

Juvenile sea cucumber rearing was conducted in the KKI Biota Laut-Gondol, Bali-Indonesia hatchery. The rearing tanks were 12 rectangular plastic tanks with dimensions of 59×47×37 cm with black inner tank walls. The average initial length and weight of sea cucumber juveniles used were 0.64 ± 0.14 cm and 0.06 ± 0.01 g, respectively. Juveniles were randomly assigned to rearing tanks and placed directly at the bottom of each tank at a density of 100 individuals per tank. Each rearing tank was equipped with three aeration points with moderate flow, and water quality was maintained through a static system with a daily manual exchange of 50% of the water volume. The bottom of the tank was cleaned to remove feces and dirt every 2 days.

OBSERVATION OF GROWTH AND SURVIVAL OF JUVENILES

Total length and body weight of sea cucumber juveniles were measured every two weeks during the two-month study. Twenty percent of the individuals at the initial juvenile stocking were taken for total length and body weight measurements. At the end of the study, juvenile survival was calculated.

OBSERVATION OF JUVENILES ATTACHED TO THE FEEDING PLATE

The percentage of juveniles attached to the feeding plate was calculated based on the number of juveniles attached divided by the total number of juveniles multiplied by 100%. Counting was done every 3 days following the installation of the new feeding plate (Figure 3). After counting, juveniles were carefully returned to their respective tanks, and the shelters were replaced with newly prepared ones pre-inoculated with diatoms. The total number of juveniles in each tank was recorded at 14-day intervals during length and weight measurements. To assess food consumption and juvenile preference for specific diatom types, both the number of attached juveniles and clear zone measurements on the feeding plate were recorded.

ENZYME ACTIVITY ANALYSIS

The intestine of sea cucumber juveniles was collected at the end of the study to determine the enzyme activity. A total of 3 individual sea cucumber juveniles were taken from 2 replicates in each treatment for enzyme activity analysis. Sea cucumber juveniles were unfed for 24 h in a separate tank to empty their intestinal contents. Before dissecting, sea cucumber juveniles were first anesthetized through immersion in seawater and given $MgCl_2$ at a dose of 5%. Five minutes after being anaesthetized, juvenile sea cucumbers were placed in an ice container and then dissected to remove the intestine.

To prepare the sample for measuring enzyme activity, the intestine was firstly weighed and mixed with a Tris buffer solution that is 10% of the sample's weight. Then, the sample was crushed using a grinding tool and centrifuged for 10 min at 12,000 rpm at 4 °C. The crude enzyme extract filtrate was then taken for protease, lipase, and amylase activity tests.

Protease activity was calculated using 20 mg/mL casein as substrate dissolved in 0.05 M phosphate buffer of pH 7 and 5 mmol/L tyrosine as standard, then, measuring the enzyme activity on a spectrophotometer at a wavelength of 578 nm. One unit of activity is the amount of enzyme that makes 1 μmol of tyrosine in one minute (Bergmeyer & Grassi 1983). One unit of activity was defined as the amount of enzyme that can produce 1 μmol of tyrosine product per minute (Bergmeyer & Grassi 1983). Amylase activity was measured using 1% starch as substrate in a 20 mM sodium phosphate buffer, pH 6.9, and containing 6.0 mM NaCl, following Worthington's method (1993). The amount of sugar released was measured spectrophotometrically at 540 nm following the 3,5-dinitrosalicylic acid (DNS) method. One unit of enzyme activity was defined as the amount of enzyme that produces 1 μmol glucose/min. Furthermore, lipase activity was calculated using olive oil emulsion as a substrate and Tris-HCl as a buffer according to Borlongan's method (1990). One unit of lipase activity was defined as the volume of 0.01 N NaOH required to neutralize the fatty acids released during 6 h of substrate incubation after being corrected with a blank.

WATER QUALITY ANALYSIS

Water quality monitoring throughout the study included temperature, pH, dissolved oxygen (DO), and light intensity. Temperature and pH values were recorded daily, whereas DO and light intensity were measured weekly. Light intensity measurements were taken at the water surface of the rearing tanks at 08:00 and 16:00 on each sampling day.

DATA ANALYSIS

The survival data, length and weight, specific growth rate, and enzyme activity of sea cucumbers were analyzed using multivariate fingerprint analysis (ANOVA) with Statistical Package for Social Science (SPSS) software (ver. 20). The normality test was carried out using the Kolmogorov-Smirnov non-parametric one-sample test. In contrast, the homogeneity test was used with Levene tests. The normally distributed and homogeneous data was then tested with one-way ANOVA and followed with the post hoc Tukey test. Each test uses a significant value of $p<0.05$.

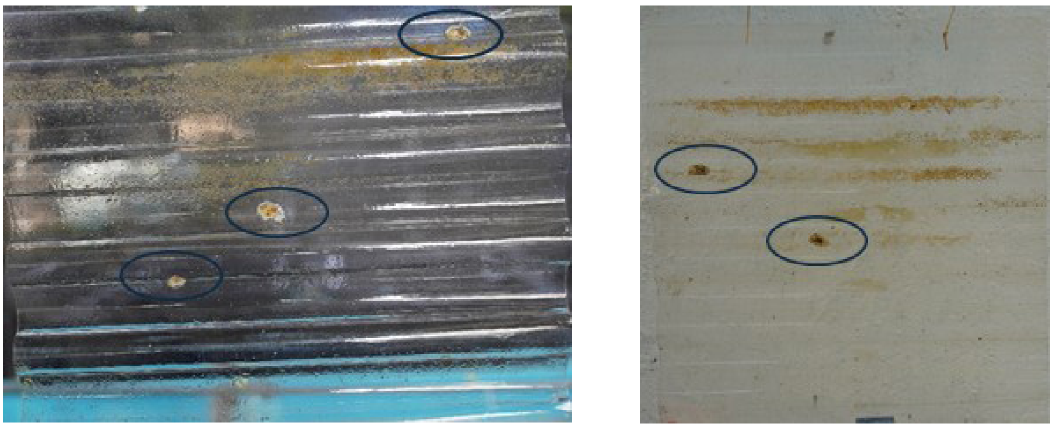

FIGURE 3. The clear zone area on the feeding plate indicates grazing (circles)

RESULTS

GROWTH AND SURVIVAL

The average total length of juveniles at the end of the experiment in treatments A and C was higher and significantly different from treatments B and D ($p<0.05$). Mean final weight and specific growth rate values in treatments A and C were also higher and significantly different from treatment B ($p<0.05$) but not different from treatment D ($p>0.05$) (Table 1). The highest survival at the end of the study was obtained in treatment C at 67.0 ± 7.02%, significantly different from the other treatments ($p< 0.05$), followed by treatments D, A, and B, which ranged from 22.0 to 37.0%.

JUVENILES ATTACHED TO THE FEEDING PLATE

The percentage of juveniles attached to the feeding plate was influenced by the availability of preferred food (Figure 4). The number of juveniles attached was significantly greater on the feeding plate with *Navicula* sp. In treatment C, the proportion of juveniles that successfully attached reached 4.5%, which was higher than in treatment B, where only *C. muelleri* and *Nitzschia* sp. were present. The percentage of juveniles attached to treatment B was 2.0%. The percentage of juveniles attached to the *C. muelleri* feeding plate in treatments A, B, and C was relatively low, ranging from 1.4% to 1.5%.

The results of this study also showed that the number of sea cucumber juveniles attached to the feeding plate was directly proportional to the size of the clear zone area on the feeding plate. The more juveniles attached to the feeding plate, the wider the clear zone size seen on the feeding plate. In treatment C, the average size of the clear zone on the *Navicula* sp. feeding plate reached 58.1 ± 28.5 mm^2, higher when compared to the clear zone on the *Nitzchia* sp. and *C. muelleri* feeding plates, which amounted to 28.5 ± 19.5 mm^2 and 0.90 ± 0.02 mm^2, respectively.

ENZYME ACTIVITY

The observation of protease activity in the intestine of specimens from treatment A was significantly different from treatment B ($p<0.05$) but not significantly different from treatments C and D ($p>0.05$). Amylase activity in treatments A, C, and D significantly differed from treatment B ($p<0.05$), with the highest activity detected in treatment A at 0.789 IU/mL, followed by treatments C and B at 0.686 and 0.654 IU/mL, respectively. The same results were also seen in lipase activity, where treatments A, C, and D were significantly different from treatment B ($p< 0.05$), with the highest activity value of 1.052 IU/mL in treatment C and the lowest in treatment B (0.664 IU/mL) (Figure 5).

During the study, monitoring of water quality parameters, including DO, temperature, and pH, was still within the range suitable for rearing sea cucumber juveniles. Light intensity was monitored twice daily at 7-day intervals for all treatment groups and replicates. Recorded values range from 134.7 to 137.3 lux in the morning and from 312.3 to 418.7 lux in the afternoon (Table 2).

DISCUSSION

Using a shelter that also serves as a feeding plate affected the growth of juvenile sea cucumbers during the two-month experiment. This pattern is in accordance with the behavior of sea cucumbers, which tend to stick to the substrate. Because of this behavior, the use of shelters has been widely practiced in the cultivation of the sea cucumber *Apostichopus japonicus* (Hu et al. 2021). The growth data of sea cucumber juveniles obtained in the present study indicate that *Navicula* sp. diatoms play an important role in promoting the growth of sea cucumber juveniles. The growth of sea cucumber juveniles also tends to be higher in rearing with the addition of shelters containing *Navicula* sp. compared to the direct provision of *Navicula* sp. in the juvenile rearing tank.

The growth of aquatic animals is influenced by genetic and environmental factors, and the nutritional content of the food consumed. In general, diatoms contain nutrients that are excellent for aquatic animals. De La Rosa et al. (2024) reported that *Navicula* sp. diatoms have a high protein content of 44.2% with a lipid content of 10.9%, carbohydrates of 9.2% and ash of 33.1%. Previously, Nijjar, Grimmelt and Brown (1991) even reported that the protein content of *Navicula incerta* reached 46.5-51.7%. However, Gómez-Ramírez et al. (2019) reported that the protein content of *Navicula* sp. was only 24.2%, with a lipid content of 12.38% and carbohydrates of 36.7%. Meanwhile, the diatom *Nitzschia* sp. has a relatively lower protein content compared to *Navicula* sp., namely 13.6% with 3.5% lipids content (de Vicose et al. 2012), 32.3% with 21.3% lipids (Lu et al. 2024), and 15.3% with 7.8% lipids (Grubisi et al. 2024). Compared to *Navicula* sp., the protein content of *Chaetoceros* sp. was also lower at 27.8% with 23.8% lipids (Rodríguez-Núñez & Toledo-Agüero 2017), 26.9% with 14.5% lipids (De La Rosa et al. 2024), and 18.1% with 27.3% lipids (Martínez-Córdova et al. 2012). The nutrient content of each diatom species is quite variable, which could be influenced by its growth phase and culture environment conditions such as temperature, salinity, and light intensity (Fimbres-Olivarría et al. 2015). Based on some of the reported diatom nutrient content data, the nutrient content of *Navicula* sp. was higher than that of *Nitzschia* sp. and *Chaetoceros* sp. Thus, the high growth rate of juvenile sea cucumbers observed in the present study was closely associated with the nutrient content of *Navicula* sp. diatoms, whether they adhered to the feeding plates (treatment C) or the walls of the rearing tanks (treatment D). In addition to its nutritional content, *Navicula* sp. can also adhere well to form biofilms on shelters that function as feeding plates (Gómez-Ramírez et

TABLE 1. Growth and survival of juvenile sea cucumber *Stichopus horrens* with the addition of different feeding plates

Parameter	Treatment			
	A	B	C	D
Initial weight (g)	0.056 ± 0.026	0.067 ± 0.013	0.047 ± 0.021	0.064 ± 0.016
Final weight (g)	0.66 ± 0.05[a]	0.22 ± 0.07[b]	0.76 ± 0.02[a]	0.47 ± 0.06[ab]
Initial length (cm)	0.68 ± 0.18	0.60 ± 0.26	0.61 ± 0.15	0.70 ± 0.12
Final length (cm)	1.08 ± 0.1[a]	0.73 ± 0.7[b]	1.07 ± 0.6[a]	0.87 ± 0.2[b]
Specific growth rate (%/day)	5.24 ± 0.08[a]	3.30 ± 0.06[b]	5.62 ± 0.06[a]	4.54 ± 0.03[ab]
Survival (%)	22.7 ± 3.51[a]	22.0 ± 4.16[a]	67.0 ± 7.02[b]	37.0 ± 6.24[c]

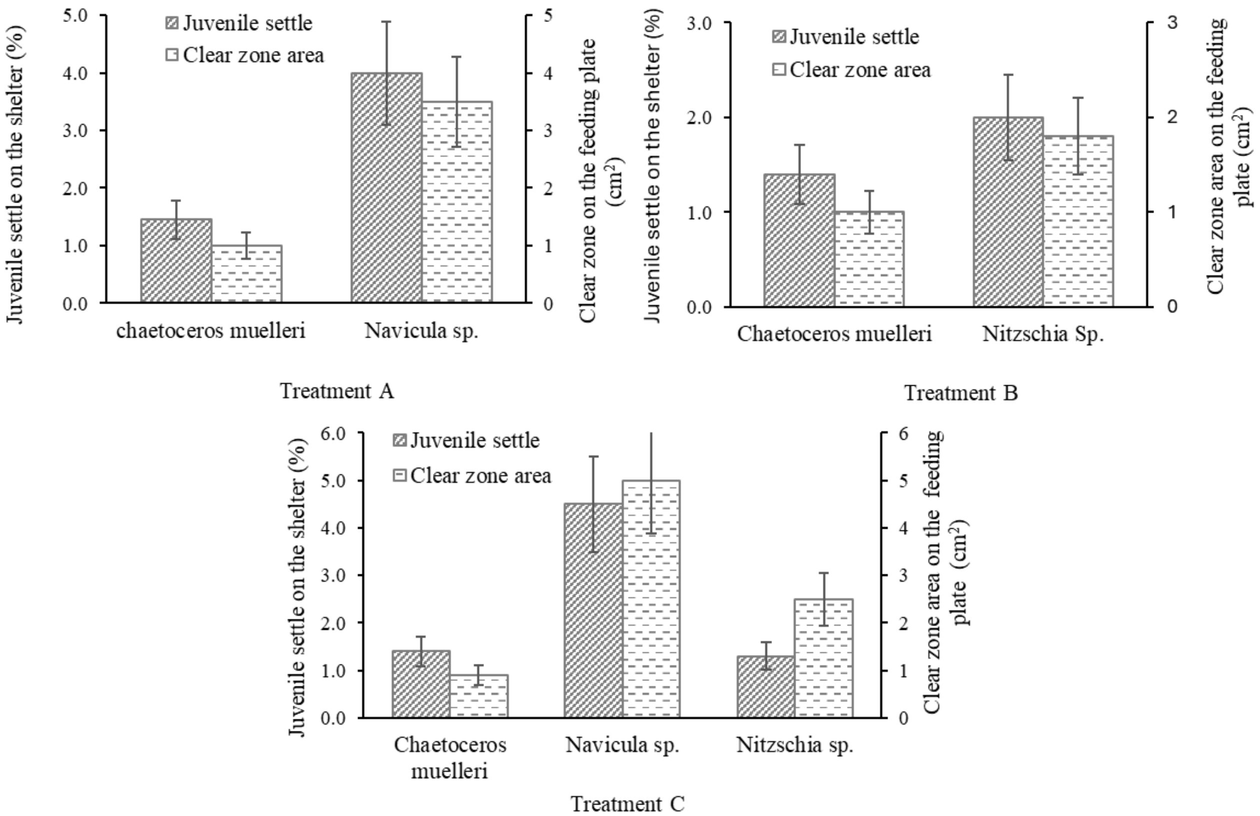

FIGURE 4. Percentage of juveniles attached to the shelter (treatments: A) *C. muelleri* and *Navicula* sp. feeding plate; B) *C. muelleri* and *Nitzschia* sp. feeding plate; C) *C. muelleri*, *Navicula* sp., and *Nitzschia* sp. feeding plate; and D) without feeding plate). The data presented in this figure is the average ± sd. of 20 observations during the experiment

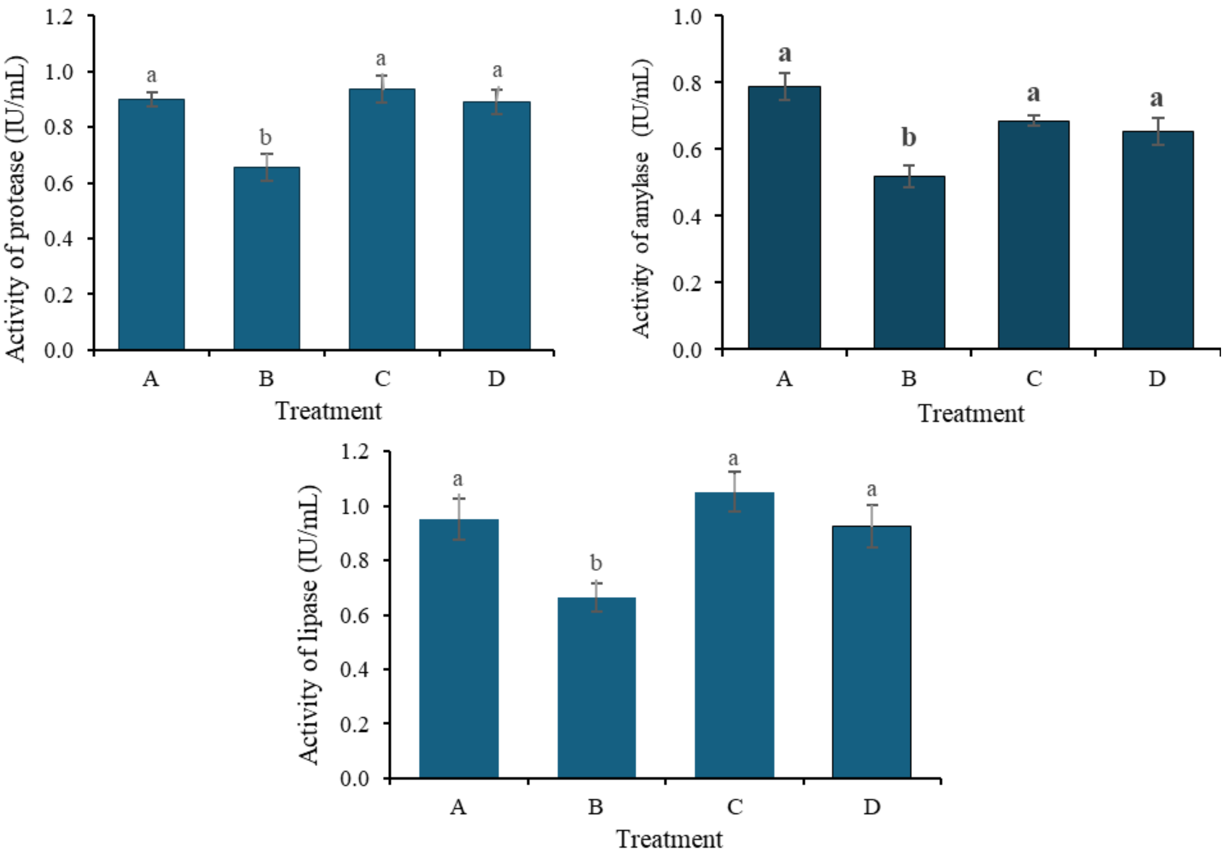

FIGURE 5. Protease, amylase, and lipase enzyme activities in the gut of juveniles at the end of the study

TABLE 2. Water quality parameter values during the experiment

Parameters	Treatment			
	A	B	C	D
DO (mg/L)	5.09 ± 0.03	5.09 ± 0.04	5.08 ± 0.04	5.10 ± 0.03
Temperature (°C)	30.00 ± 0.10	29.98 ± 0.17	29.97 ± 0.32	29.98 ± 0.12
pH	8.04 ± 0.04	8.07 ± 0.06	8.02 ± 0.03	8.05 ± 0.06
Light intensity (Lux)				
Morning	137.33 ± 16.60	134.67 ± 23.34	137.00 ± 12.03	135.67 ± 4.92
Afternoon	312.33 ± 16.76	314.00 ± 12.33	318.67 ± 14.64	318.00 ± 9.93

al. 2019). It was further explained that the type of shelter material used greatly influences the thickness of biofilms formed by *Navicula* sp. Hu et al. (2013) used corallite pipe material and polyvinyl chloride as shelters to facilitate the attachment of juvenile sea cucumbers *S. horrens* reared in nets. The study by Romero-Gallardo et al. (2024) showed that the sea cucumber *Isostichopus badionotus* preferred shelters with biofilm on the surface, especially for small-sized sea cucumbers. Biofilms on surfaces of shelter can induce larvae to settle (Pearce & Scheibling 1990), and *A. japonicus* juveniles actively prefer refugia with biofilms on their surfaces (Dong et al. 2010), which can be a food source (Liu et al. 2017; Zhang et al. 2015). Rioja, Palomar-Abesamis and Juinio-Meñeza (2020) reported that the growth of juvenile *Stichopus cf. horrens* increased significantly with the addition of shelter or dark conditions in the rearing tank, as it can increase food accessibility. The shelter can provide additional grazing surface for juveniles, thus reducing competition for food.

The combination of *C. muelleri, Nitzschia* sp., and *Navicula* sp. yielded high survival rates in both the presence (Treatment C) and absence (Treatment D) of shelter, outperforming the two-species combinations of *C. muelleri* with either *Nitzschia* sp. or *Navicula* sp. The findings indicate that the combination of three diatom species provides a more balanced and complete nutritional profile, which significantly enhances the survival of juvenile sea cucumbers. Hu et al. (2013) reported that from oosperm to settled juvenile, the survival rate of sea cucumber *S. horrens* was 1.0%-6.83% when enough live microalgae were supplied, and the survival rate reached about 23.3%-50.5% during 80 days of culture after settlement. The presence of shelters with diatom biofilms in the rearing tanks can provide a wider area for sea cucumber juveniles to attach. Mercier, Battaglene and Hamel (2000) stated that phytoplankton substrates are suggested as favorable substrates for sea cucumber *H. scabra* larval settlement, serving as a transition stage to the sandy seafloor.

The shelter's capacity as a feeding plate with more food availability significantly increased growth and survival compared to feeding plates with less food availability. In this study, the percentage of juveniles attached to the feeding plate with a combination of the three diatom feeds was higher than in the treatment with only two species of diatom feed. This may be due to the lack of density of *C. muelleri* and *Nitzschia* sp. diatoms attached to the feeding plate (Figure 2), causing a limited food source for juveniles. Slater and Jefs (2010) stated that the availability of phytobenthos plays an important role in the development of sea cucumber juveniles.

Juveniles reared on the feeding plate treatment with a combination of three diatom species tend to form a clean circle (clear zone) on the feeding plate, and they will remain in the area until the available food runs out. This can be seen from the appearance of a clear zone area on each feeding plate used (Figure 2), and this indicates that feeding plates can increase the effectiveness of providing food for sea cucumber juveniles.

Increased growth and survival of juveniles are the most important goals during juvenile production, which is to increase production efficiency. The type of feed given and the ability of the sea cucumber to digest the feed greatly affect the effectiveness of the feed in supporting the growth of sea cucumber juveniles. The activity of digestive enzymes, including protease, lipase, and amylase, can illustrate the ability of sea cucumbers to digest the feed consumed. Enzyme activity in the gut of juvenile sea cucumbers contributes to changes in the digestive capacity and absorption of feed nutrients, which can further support sea cucumber growth (Gao et al. 2015, 2014; Tang et al. 2015). In this study, the activities of the enzyme protease, lipase, and amylase were similar in both groups with and without feeding plates. However, it is noteworthy that the activity levels of these three enzymes were low in treatment B, which included a feeding plate but no *Navicula* sp. This result is in line with the results of research by Xie et al. (2017), who reported that juvenile sea cucumbers *A. japonicus* fed with *Navicula* sp. showed higher amylase, protease, and lipase enzyme activities. These results indicate that the benthic diatom *Navicula* sp. has a positive effect on the growth of sea cucumber juveniles because it can increase the activity of digestive enzymes. This data aligns with the better growth response of juvenile sea cucumbers in the three treatments fed with *Navicula* sp. diatoms in this study.

CONCLUSION

The growth performance of juvenile sea cucumber *Stichopus horrens* is enhanced by the inclusion of shelter in the rearing tanks, particularly when the shelter is colonized by the benthic diatom *Navicula* sp. The results of this study also suggest that the diatom *Navicula* sp. plays a significant role in promoting the growth of juvenile sea cucumbers.

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