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#### Research Article

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# The Effect of Stocking Density on Survival and Growth of Tiger Shrimp (*Penaeus monodon*) Cultivated with Seaweed (*Gracilaria* sp.) in Traditional Pond

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#### **ABSTRACT**

In order to overcome the decline of pond carrying capacity due to intensification, polyculture of *Penaeus monodon* with *Gracilaria* sp. can be implemented to allow sustainability. This study aimed to evaluate the effect of shrimp densities in a polyculture system with *Gracilaria* sp. on the shrimp's survival rate and growth. The research design used in this study was Randomized Block Design (RBD) in triplicate with 3 treatments of stocking density (2, 3, and 4 ind/m²). This research was carried out for 90 days in extensive ponds with 0% use of artificial feed. At the end of the study, the results showed that different stocking density applied had no significant effect (p>0,05) on growth performance. Nonetheless, shrimp at a stocking density of 2 ind/m² had the highest survival rate (30,30±1,48%) compared to other treatments. Higher shrimp density causes competition for space, food, and oxygen which leads to stress resulting in suppressed growth of shrimp. However, *Gracilaria* integration in shrimp polyculture effectively reduces nutrient loads and stabilize water quality (temperature, pH, salinity, and DO), leading to improved shrimp health and a decreased need for chemical inputs, offering a practical, eco-friendly solution for maintaining shrimp growth in such a denser environment.

Keywords: algae, aquaculture, bioremediation, coastal, integrated

# **ABSTRAK**

Dalam rangka mengatasi penurunan daya dukung lingkungan akibat intensifikasi budidaya udang, sistem polikultur udang windu dengan rumput laut Gracilaria sp. dapat diimplementasikan sebagai pendekatan dalam pengembangan budidaya yang berkelanjutan. Penelitian ini bertujuan untuk mengetahui pengaruh dari padat tebar udang windu yang berbeda terhadap sintasan dan pertumbuhannya yang dibudidayakan dengan Gracilaria sp.. Rancangan penelitian yang digunakan dalam penelitian ini adalah Rancangan Acak Kelompok (RAK) dengan 3 perlakuan padat tebar (2, 3, dan 4 ekor/m²) dan 3 kali ulangan. Penelitian dilaksanakan selama 90 hari pada tambak tradisional dengan pemanfaatan 0% pakan buatan. Hasil penelitian menunjukkan bahwa padat tebar udang tidak berpengaruh nyata (p>0,05) terhadap performa pertumbuhan. Meskipun demikian, perlakuan kepadatan 2 ekor/m² mendapati nilai sintasan tertinggi (30,30±1,48%) dibanding dengan perlakuan yang lainnya. Peningkatan padat tebar menyebabkan adanya kompetisi ruang, pakan, dan oksigen yang mana menimbulkan stres sehingga terjadi penekanan laju pertumbuhan pada udang budidaya. Namun, budidaya udang terintegrasi dengan Gracilaria secara efektif menurunkan kandungan nutrien anorganik (amonia) dan menstabilkan kualitas air (suhu, pH, salinitas, dan DO), sehingga meningkatkan kesehatan udang dan mengurangi kebutuhan penggunaan bahan kimia, yang mana merupakan solusi praktis dan ramah lingkungan untuk menjaga petumbuhan udang di lingkungan kepadatan tinggi.

Kata kunci: alga, akuakultur, bioremediasi, pesisir, integrasi

#### 1. Introduction

Indonesia's aquaculture production is currently ranked second after China with a total production of 14.84 million tons out of 122.57 million tons of global aquaculture production (FAO, 2022). One of the leading export products from Indonesia in the aquaculture sector is shrimp, one of which is tiger shrimp (Penaeus monodon). With its high sales value, the tiger shrimp business is quite promising. However, the main problem with tiger shrimp cultivation in the last few decades is low production. This is caused by a wide range of environmental impacts such as mangrove degradation, a decrease in water quality, inland salinization, and disease outbreaks (Sivaraman et al., 2018). Under these conditions, the pond is not ideal for shrimp farming.

Low shrimp production due to decreased carrying capacity and disease infections has encouraged the government to maintain shrimp production usina traditional (extensive) systems. Traditional aquaculture is mostly environmentally friendly because it uses waste and by-products available such as crop residues, animal or human waste, or natural food in open water as nutritional input for the cultivated aquatic organisms (Edwards, 2015). Farmers usually grow tiger shrimp with a low stocking density. i.e. between 1-5 ind/m<sup>2</sup> (BBPBAP, 2017). However, increasing the production of tiger shrimp still needs to be accomplished. Increasing the density of shrimp fry is an approach commonly used to increase the production and efficiency of ponds in shrimp cultivation (Sun et al., 2016). According to Anh et al. (2022), seed density is an important aspect that influences growth and productivity in tiger shrimp cultivation. Therefore, the utilization of correct cultivation systems is very necessary, one of which is a polyculture system alongside seaweed production.

Randusanga Wetan Village, Brebes Regency is a center for red seaweed (Gracilaria). So far, many farmers have carried out polyculture methods between Gracilaria and several other commodities such as milkfish and shrimps. Biologically, seaweed can be added to tiger shrimp ponds to serves and improve the quality of the environment for its growth (Widowati et al., 2020). According to Rejeki et al. (2016), Gracilaria sp. has the capability to absorb excess organic nutrients and also toxic pollutants in water. Seaweed utilizes nutrients (N and P) as a food source for its growth and development. While nitrogen acts as a constituent of amino acids, phosphorus could present as energy for photosynthesis (Khatimah et al., 2023). Additionally, the seaweed polyculture provides higher profit value (Diatin et al. 2020), which could help farmers get faster economic returns by having two different commodities cultured. Thus, it can be inferred that this system creates such an optimal condition, which is benefitting both the environment and economic outcome.

Previous research on tiger shrimp density was carried out by Aftabuddin et al. (2020), who culture tiger shrimps at different densities in concrete tanks. Based on the research results of Amalia et al. (2022), cultivating tiger shrimps combined with seaweed and blood cockle is the most effective way to increase shrimp growth rate. Where the daily growth rate of shrimp (SGR) increased from 5.62±0.04%/day (monoculture) to 5.75±0.03%/day (polyculture). Other research related to tiger shrimp culture with Gracilaria sp. by Anh et al. (2022), in traditional plus ponds on the Mekong River, Vietnam, found that polyculture shrimp with a 50% artificial feed ratio had comparable growth to monoculture shrimp with a 100% artificial feed ratio. Therefore, this research was conducted to find out the growth performance of tiger shrimps and the optimal stocking density for use in polyculture systems in traditional ponds.

### 2. Materials and methods

This research applies experimental methods and was carried out in Randusanga Wetan Village, Brebes Regency, Central Java from December 2022 to February 2023.

## 2.1. Research materials

Penaeus monodon post larvae were purchased from a nearby hatchery around Randusanga Wetan Village, Brebes. The tiger shrimp used are shrimp that have entered the juvenile stage (PL 25) with an initial body weight of 0.23±0.05 g/ind. The red seaweed Gracilaria was collected from a seaweed farm in Tangerang, Banten province before being sent to the culture site.

# 2.2. Research procedure

research was carried out in Randusanga Wetan village, Brebes District, Brebes, Central Java. Three traditional ponds with an area of around 3000-10.000 m<sup>2</sup> are used during this experiment (Figure 1). Each pond is divided into 3 parts with partitions made of bamboo and fish nets so that each part has 300m<sup>2</sup> in size (Figure 2). The preparation of the ponds includes cleaning the pond. The pond was cleaned of pests by applying saponin and leaving it for a week. After cleaning, the seaweed is added into the pond. Seaweed is sown a week earlier before the shrimp are stocked. Seaweed is stocked using the broadcast method with a density of 100 g/m<sup>2</sup>, which commonly used by local farmers. The shrimp and seaweed are maintained until the end of the experiment whilst the data needed is collected every month.



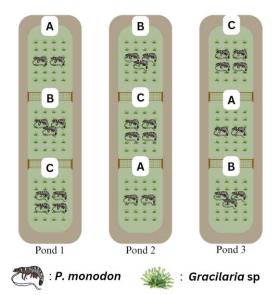
Figure 1. Satelite Image of The Research Pond Location

# 2.3. Pond Maintenance

The Regular water exchange is essential for maintaining water quality and balancing salinity during research period. In traditional brackish water ponds, tidal flow is commonly used to ensure water quality by facilitating the natural movement of water in and out of the pond. This natural movement of water helps maintain optimal oxygen levels, crucial for the health of cultivated. Additionally, planting vegetation like mangrove along the pond dikes plays an important role especially to prevent erosion. The combination of tidal flow management, and adding vegetation along the dikes help create sustained ecosystem without using too much artificial product.

# 2.4. Research design

This research was designed with a Randomized Block Design where there were 3 stocking density treatments. Each treatment was tested with 3 replications in different groups so that created 9 different enclosures (Figure 2). The treatment provided includes A (Shrimp density of 2 ind/m²), B (Shrimp density of 3 ind/m²), and C (Shrimp density of 4 ind/m²), which was modified from previous research from Ahn *et al.* (2022) where the trials consisted of three different stocking densities (2, 4, and 6 ind/m²) which fed with 100% and 50% ratio of commercial feed.



**Figure 2** Model/Research Design with 3 different treatment namely, A (2 ind/m²), B (3 ind/m²), and C (4 ind/m²)

# 2.5. Data collection

The shrimp growth is monitored once a month by taking 10 individuals at random from each enclosure to be used as samples. An electronic weighing scale from *Camry* with a precision of 0.1 g was used to perform the weighing. At the end of the rearing period, all the shrimp harvested are counted to determine the survival rate. Shrimp growth performance is measured by several variables, namely weight gain and Specific Growth Rate (SGR).

Water quality was managed by adding clean water through the inlet at least once a week. The parameters, such as temperature, pH, DO and salinity in the cultivation media were measured every morning during the 90-days of study. Water temperature and salinity were measured with a WQC (Water Quality Checker) Thermo Scientific Orion Star a322. The value of pH was measured using a pH meter EZDO® 5011 with an accuracy of 0.01. The DO was recorded using the YSI@Pro DO meter (read in 0.1 mg/L). Ammonia and nitrate measurements were carried out once a month. Water samples for ammonia and nitate analysis were collected from each pond and immediately preserved with ice in a cooler box to preserve the samples.

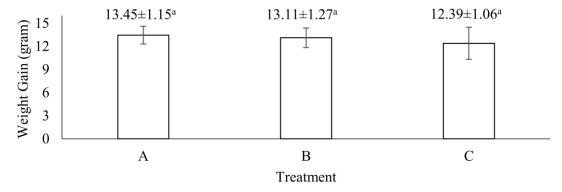
Samples then transported within 6 hours to the laboratory in the Faculty of Fisheries and Marine Science, Universitas Diponegoro to be analyzed using spectrophotometer.

# 2.5. Data analysis

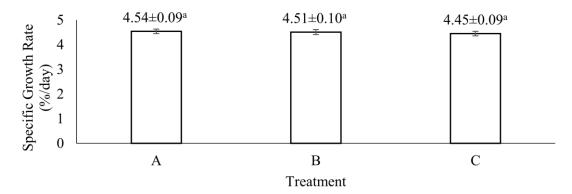
Shrimp weight gain, SGR, and SR data were tested using analysis of variance (ANOVA). The ANOVA was used to determine significant treatment interactions. Data was tested if and only if the data was normally distributed and homogeneous. The normality test was carried out using the Shapiro-Wilk test, while the homogeneity value was tested using the Levene test. Data were analyzed with a confidence level (F test) of 95%. If there was a significant difference, Duncan's multiple area test is carried out to determine the difference in the mean values of the treatments, so that the best treatment results can be obtained.

#### 3. Results and Discussion

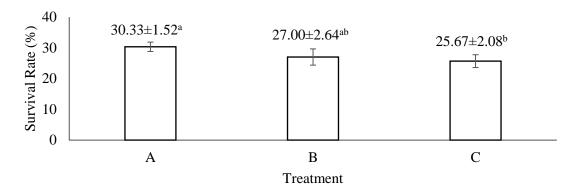
Results of statistical analysis of the overall data showed that stocking density significantly affected the survival of tiger shrimps but had no effect on the growth. The shrimp growth performance can be seen in Figure 3, 4, and 5.



**Figure 3.** Weight Gain of *P.monodon* in polyculture with *Gracilaria* sp. at various stocking densities over 90 days of culture



**Figure 4.** Specific Growth Rate (SGR) of *P.monodon* in polyculture with *Gracilaria* sp. at various stocking densities over 90 days of culture



**Figure 5.** Survival Rate (SR) of *P.monodon* in polyculture with *Gracilaria* sp. at various stocking densities over 90 days of culture

Note: Values are mean  $\pm$  SD, mean values with different superscripts on the same row represent significantly different results (p<0.05)

The weight gain during 90 days of culture was in the range of 12.39–13.45 g (Figure 3). The three treatments showed no significant differences in values (P > 0.05). Same thing was also recorded on the SGR value, as no significant differences were found between the three treatments (P > 0.05). The less pronounced growth rate differences between stocking densities of all treatments suggest that at these densities, resource availability may still be within an acceptable range, preventing intense competition for food and space. The SR obtained in this study showed that the lowest stocking density (2 ind/m²) had the highest shrimp survival rate (30.30±1.48%.) and statistically showed significant difference on values (P<0.05) from stocking densities of 4

The data obtained on water physics and chemistry are presented in **Table 1**. The water quality for each parameter experienced fluctuations during the research period. The temperature recorded in the three ponds varied from 29.7 to 29.91 °C. The variation in values that occurred was not much different even though in pond 2 the temperature value was

slightly warmer than in the other two ponds. The pH of the three ponds has almost the same value with an average of 7.8. Meanwhile, DO varies in each pond with a value range of 3.49-5.21 mg/L, where pond 2 has the highest average DO value (5.21 mg/L). The salinity value also differs in each pond from 25.29 to 28.37 ppt. During the maintenance period, the nitrate values obtained ranged from 0.92-1.2 mg/L, and the ammonia values ranged from 0.04-0.05 mg/L. During this experiment, all the parameters are still in the recommended range, despite the different treatments performed.

Based on the results of several previous studies, it is known that stocking density can directly influence the growth and survival of shrimp, as well as the production efficiency of each pond unit (Anh et al., 2020). A study by Aftabuddin et al. (2020) also obtained results where different stocking densities (400, 450, and 500 PL/m³) would produce different growth with the lowest stocking density producing the highest growth. However, in this study, all treatments don't really show a distinct difference in the growth performance. It can be due to the pond water condition that is still in the

Table 1. Water Quality Parameters in The Culture Pond

Parameter	Pond 1	Pond 2	Pond 3	Recommended values	
				P. monodon	Gracilaria sp.
Temperature (°C)	$29.70 \pm 1.12$	$29.91 \pm 1.09$	$29.89 \pm 1.33$	29 – 32 (*)	20 - 34 (a)
pН	$7.81 \pm 0.27$	$7.82 \pm 0.36$	$7.84 \pm 0.29$	7.6 - 8.8 (*)	6.8 - 8.2 (b)
DO (mg/L)	$3.49 \pm 0.96$	$5.21 \pm 1.84$	$3.81 \pm 0.85$	>3 (*)	> 3 - 8 (c)
Salinity (ppt)	$26.03 \pm 3.31$	$25.29 \pm 3.52$	$28.37 \pm 2.94$	$5 - 40^{\ (*)}$	$15 - 30^{(b)}$
Nitrate (mg/L)	$0.92 \pm 0.05$	$0.97 \pm 0.05$	$1.2 \pm 0.24$	<10 (**)	$0-1.45^{\ (a)}$
Ammonia (mg/L)	$0.04 \pm 0.04$	$0.052 \pm 0.03$	$0.037 \pm 0.04$	<0.1 (***)	< 1 <sup>(d)</sup>

Note:

<sup>(\*)</sup> WWF (2014); (\*\*) PPRI (2001); (\*\*\*) Tsai and Chen (2002), (a) Rejeki et al. (2018), (b) SNI (2010),

<sup>(</sup>c) Ariyati et al. (2007), (d) Pescod (1973)

favor of shrimp growth and the highest density of 4 ind/m<sup>2</sup> remained within the pond's carrying capacity especially with the help of the red seaweed, Gracilaria. This findings are in line with the study from Arnold et al. (2009) that recorded insignificant result from different densities of P. monodon with the presence of substrates. In their study, the mean weight of shrimp was 0.33±0.03 g at a density of 2,500 m<sup>3</sup> and 0.30±0.04 g at a density of 5,000 m<sup>3</sup>, which statistically not significant. Additionally, Anh and Ngan (2017), demonstrated that the co-culture of black tiger shrimp (Penaeus monodon) with seaweed improved water quality in co-culture treatments compared to monoculture treatments.

The results of this research have a lower shrimp weight value compared to the national standard for tiger shrimp rearing (SNI, 2021), where the shrimp weight reared for 90 days has an average weight of 17.5 g. This could happen since each pond contains a different amount of food resources. Without any supplements and fully reliant on natural food, it is possible for shrimp to not grow according to the estimated weight. However, the ability of red seaweed (Gracilaria sp.) that acts as a substrate and natural food source for shrimp, as well as maintaining water quality (Roleda and Hurd 2019; Diatin et al. 2020; Anh et al. 2024) help the shrimp with different treatments to grow without contrast weight gain between each treatments.

Shrimp growth rate is directly related to environmental conditions. Traditional cultivation is also a cultivation technique whose results are very dependent on natural conditions because the system does not utilize additional feed other than natural feed from ponds. The results of this study have a lower SGR value (4.45-4.54%/day, Figure 4) compared to previous research by Amalia et al. (2022), with an SGR value of more than 5.6%/day. The SGR value in this research is a sufficient value for tiger shrimp cultivation. However, the decline in the SGR value is thought to have occurred because the condition of the ponds (water and soil quality) worsened. The carrying capacity of water will decrease if ponds are used continuously without proper management. During the growout period, nutrients and organic residues tend to accumulate at the bottom of the pond and excessive accumulation can result in damage to the pond system (Lemmonier et al., 2004). According to Choeronawati et al. (2019), the outcome of fish farming production is related to biotechnical aspects which include selection, pond construction, infrastructure and the cultivation system used. Similar to the results of research by Prihadi and Brata (2019) which found that ponds resulting from remediation produced 357.15 kg/ha of tiger shrimps and ponds without remediation produced 50-100 kg/ha.

In traditional brackish water ponds, other than technical aspects, availability of food is also crucial. Plankton is one of the main natural food sources for shrimp, so increasing the productivity of pond cultivation is primarily dependent on the availability of plankton in the pond (Hendrajat and Andi, 2018). The amount of natural food stock available in ponds will decrease as stocking density increases. A study by Abu Hena et al. (2006) found that 64.72-91.87% of macrobenthos experienced a decrease in the total number of species sampled in ponds during the cultivation period, indicating that shrimp can intensively prey on benthic fauna in a pond. In addition, high stocking densities produce higher waste excretion which harms the growth of tiger shrimps. Anh et al. (2020) also found that differences in tiger shrimp density from 1,000-3,000 PL/m<sup>3</sup> resulted in different TAN contents, where a shrimp stocking density of 3,000 PL/m<sup>3</sup> produced the highest amount of TAN, namely 0.63 mg/L, which is harmful for the growth of tiger shrimps. Nevertheless, adding Gracilaria in a shrimp pond is such a good strategy for farmers to maintain shrimp growth while applying higher density of shrimp.

Seaweeds are ecosystem engineers which could alter coastal environments by creating complex habitats on previously bare mudflats. Seaweed such as Gracilaria can also be functional as a place for micro macrozoobenthos to live and reproduce (Matinez-Curci et al. 2023), which are crucial natural food for shrimp, crab, fish, and other aquatic organisms in water bodies (Abualreesh 2021). In this trial, no significant differences in shrimp growth were observed, likely because the shrimp were cultivated in a healthy area with optimal conditions, minimizing stress and resource competition. The abundance of food sources could be one of the reasons for the insignificant distinction between treatments. since the pond's bottom was not dried during the pond's preparation. According to Ahn et al. (2024), the dried pond coupled with liming could result in the mortality of most zoobenthos species. However, P. monodon shrimp could also ingest both the natural food sources (zooplankton and zoobenthos) and red seaweed available in the culture pond, contributing to shrimp growth under non-feed supplement conditions.

Until the end of the research period, the SR value of tiger shrimps had decreased from the initial number of shrimps released in the pond. The SR value for the 3 ind/m<sup>2</sup> treatment was not significantly different from both the 2 ind/m<sup>2</sup> and 4 ind/m<sup>2</sup> treatments, but the 2 ind/m<sup>2</sup> treatment showed a significant difference compared to the 4 ind/m<sup>2</sup> treatment, with the highest SR value recorded at 30.3±1.48% for 2 ind/m<sup>2</sup> (Figure 5). Mortality which increases with high stocking density can occur due to lack of nutritional intake and physiological decline in shrimp due to stress. When shrimp are exposed to less than optimal environmental conditions (ex. crowding stress, poor water quality, or nutritional imbalance), the shrimp's immune function is reduced and they are susceptible to disease (Boyd and Phu 2018). According to research by Abdussamad and Thampy (1994), cannibalism during molting can be the main cause of death of P. monodon in the post-larval and juvenile stages because they are more susceptible to cannibalism at higher densities. There are numerous biotic and abiotic factors that contribute to the success in production including product quality (Varghese et al., 2020). Where abiotic factors are physical and chemical factors of water (water quality) while biotic factors include predators, food, and the presence of other biota or competitors.

The SR of this study is lower than the results of the study by Anh et al. (2022) who obtained the SR value for tiger shrimps from polyculture with Gracilaria tinuistipitata which varies from 67.01-91.67%. However, the results of this study are in line with research by Taslihan et al, (2015) which found that tiger shrimps cultured in traditional ponds without biosecurity and surrounded by other shrimp ponds, had a survival rate of 22.6±15.8%. The limited amount of natural food certainly has a big influence on meeting the energy needs and health of shrimp. However, the presence of Gracilaria cultivated with shrimp is able to satabilize water conditions so that the media is still in good condition. This is supported by the water quality conditions (Table 1) during the experiment. Research by Widowati et al. (2020), also showed that seaweed densities of 150 and 100 g/m<sup>2</sup> were more effective in reducing ammonia content compared to a density of 50 g/m<sup>2</sup>. Therefore, there was no significant difference in survival values between the stocking density of 3 ind/m<sup>2</sup> towards stocking density of 4 ind/m<sup>2</sup> or 2 ind/m<sup>2</sup>. Nonetheless, the integration of seaweed, Gracilaria, is a great combination for improving the carrying capacity of aquaculture ponds by enhancing and sustaining water quality whilst providing additional habitats within the system.

During 90 days of culture, water quality parameters experienced continuous fluctuations. Based on the research data, it is known that the values of water quality parameters including temperature, pH, DO, salinity, nitrate, and ammonia are still within the acceptable range for suitability for tiger shrimp cultivation waters. The water quality conditions observed in this study, respectively, were not significantly different from those reported in a similar study by Anh et al. 2022 with an average temperature ranging from 27.4 to 30.3 °C (minimum 26.2 °C and maximum 32.2 °C), and pH levels of 7.9-8.5. The temperature in each pond is not much different (varies from 29,71-29,91 °C) because the geographical conditions are not too far away so the intensity of light entering the water is also not much different. The pH content is also greatly influenced by weather and microbial activity in water. Due to the absence of rain storms during the dry season, pH showed lower values than during the rainy season (Rodrigues et al., 2017). Longterm pH stress on shrimp can reduce growth, whereas low or high pH can alter the composition and function of shrimp gut microbiota (Yu et al., 2020). Meanwhile, during the rainy season when fresh water enters the ponds, it affects water conditions so that salinity decreases. The rapid fluctuations in salt levels can cause physical strain in farmed animals, leading to changes in their susceptibility to diseases (Millard et al., 2020). However, in this study, it was noted that the salinity value was still within the threshold for suitability for tiger shrimp.

The three ponds still have suitable DO values according to WWF (2014) eligibility standards, namely >3 mg/L. The low DO values are thought to be because data collection was carried out in the morning so that photosynthetic microorganisms activity of such phytoplankton did not occur. In the morning, the water temperature is cooler and the light intensity is not as much as during the day so the number of phytoplankton also decreases. Apart from elements like nutrient availability, light, and grazing pressure, various types phytoplankton react differently to varying temperature conditions (Schabhhüttl et al., 2012). Dissolved oxygen has an important role in influencing feed consumption and tiger shrimp maturation. Lack of dissolved oxygen can be directly harmful to shrimp, especially because it causes a substantial increase in toxic metabolites (Gunalan, 2010).

The nitrate and ammonia content during the cultivation period is known to still be within the feasibility standards for cultivating tiger shrimps and *Gracilaria* sp. During the culture period, the nitrate values obtained ranged from 0.92-1.2 mg/L, and the ammonia values ranged from 0.04-0.05 mg/L. An increase in nitrate content occurs when nitrifying bacteria work by oxidizing nitrite to nitrate. The availability of nitrate in ponds functions as a necessary nutrient for the seaweed *Gracilaria* sp. to grow. Nitrate values in ponds between 0 and 1.45 mg/L are sufficient concentration for the growth of red seaweed *Gracilaria* sp. (Rejeki *et al.*, 2018).

Increasing stocking density from 2 to 4 ind/m<sup>2</sup> in this study did not cause ammonia values that exceeded the standard limit (<0.1mg/L). Low ammonia content indicates that ammonia decomposition has occurred by the Gracilaria sp. The mechanism of seaweed as a biofilter is to take in nutrients such as ammonia and nitrate during photosynthesis. The decreasing ammonia concentration indicates that Gracilaria starts absorbing the ammonia in wastewater after the 10th day through a process called ammonification which is used for its growth (Trianti and Ratih, 2020). The research conducted by Anh et al. (2020) reported that the quality of the waters used for cultivating tiger prawns with G. tenuistipitata seaweed do not differ significantly between shrimp stocking density treatments (1,000; 2,000 and 3,000 PL/m³). Anand et al. (2019) also mentioned that stocking density does not affect cultivation water quality parameters. Furthermore, Anh et al. (2022) observed that the use of Gracilaria seaweed significantly reduces nitrogen and phosphate content in waters, which shows that seaweed utilizes the nutrients obtained from shrimp waste for the growth of the seaweed itself. By using shrimp waste for growth, seaweed acts as a biofilter and pond water quality improves and can be maintained (Andayani et al., 2016).

This study was done in relatively limited space. It may create issues in scaling the findings to other settings of large scale farming. Conducting studies of a larger scale will help substantiate these findings thus rendering the recommendations appropriate for aquaculture systems. In addition, this relatively short duration of the study prevented the assessment of certain long-term consequences on growth, survival or system sustainability due changes in stocking densities. Supplementing the duration may prove beneficial in examining seasonal effects and the extent to which sustained densities have on shrimp's health, yield reliability and Gracilaria's condition in the polyculture system.

In terms of future directions, it would be preferable to investigate a number of other variables that may be significant such as the biomass of *Gracilaria*, nutrients, and water quality management measures in relation to improving shrimp growth and survival. Additional studies could assess the pollution reduction and water clearing in *Gracilaria* and the possible synergism in polyculture systems to evaluate the ecological advantages of the polyculture systems approach. In this way, the future studies could make better stocking density recommendations by broadening the scope of these variables.

## 4. Conclusion

Implementing various shrimp densities (2, 3, and 4 ind/m<sup>2</sup>) on an extensive polyculture system had a significant influence on shrimp survival but not on its growth performance (absolute weight and SGR). Based on these findings, farmers can adopt all the three different densities applied (2, 3, and 4 ind/m2). However, it is recommended that farmers utilize 2 ind/m<sup>2</sup> of stocking density for optimal yielding a weight gain outcomes, approximately 13.45±1.15 g, SGR of 4.54±0.09 %/day, and SR of 30.30±1.48% in traditional polyculture systems with Gracilaria. Further studies are still needed to understand more about the effect of seaweed's types and how the density of *Gracilaria* could enhance the growth of tiger shrimp.

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