

# Studying How to Minimize Nutrient Waste while Maximizing Macroalgae Biomass; A Vertical Method Study on An Integrated Aquaculture Recirculating System

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## Abstract

Integrated multi-trophic aquaculture (IMTA) has been highlighted in terms of the placement of species in nutrient distribution areas. Similarly, the feeding rate in monoculture system can be complex and threaten aquaculture sustainability. A study was conducted with the different feeding rates (5%, 7.5%, and 10% day<sup>-1</sup>) on the rabbitfish *Siganus guttatus* in an IMTA system. The system included the green mussel *Perna viridis*, sea urchin *Diadema setosum*, and seaweed *Kappaphycus alvarezii*, which was grown vertically. The aim was to evaluate the ability of *K. alvarezii* to remove inorganic nutrients produced from feed waste at each feeding rate. The 42-day experiment showed that *K. alvarezii* removed total ammonia nitrogen (TAN), dissolved inorganic nitrogen (DIN), and dissolved inorganic phosphorus (DIP) nutrients, above 95% for all feeding rates. There was no significant difference in nutrient removal efficiency between the feeding rate levels ( $P > 0.05$ ). However, the biomass of *K. alvarezii* increased with higher feeding rates with a significant difference ( $P < 0.05$ ). The capacity of vertically reared *K. alvarezii* optimally to reduce the concentration of nitrogen and phosphorus compounds. This shows that this algae cultivation method can effectively manage fish feed waste in a multi-trophic cultivation system.

## Introduction

Aquaculture is considered a crucial sector for ensuring food security for the world's growing population (FAO, 2022). However, numerous studies have found that the environmental impact of aquaculture activities, particularly in intensive systems that use artificial feed to promote species growth, is on the rise (Boyd et al., 2020; Dauda et al., 2019). The expansion of intensive monoculture systems has led to an increase in nutrient waste from feed residue, feces, excretion, and respiration products (Amoussou et al., 2022; Nederlof et al., 2021). Many studies have focused

on quantifying nutrient waste loads in aquaculture activities. For example, in a monoculture system, 36-63% of nitrogen (N) and 18-30% of phosphorus (P) from feed are released into the water Nederlof et al. (2021). This waste poses a threat to the sustainability of aquaculture (Heriansah et al., 2024) and food security.

The integrated multi-trophic aquaculture (IMTA) system is considered a sustainable solution for aquaculture development (Dauda et al., 2019; Nederlof et al., 2021) and offers greater benefits compared to monoculture systems (Thomas et al., 2021). In an IMTA system, fed species such as fish or shrimp are integrated with lower trophic species, including organic extractive

species (e.g., shellfish, sea cucumbers, and sea urchins) and inorganic extractive species (e.g., macroalgae, commonly known as seaweed). This creates a nutrient flow scenario where suspended leftover feed and feces from fish or shrimp can be filtered by shellfish as suspension feeders, settled waste can be consumed by sea cucumbers or sea urchins as deposit feeders, and dissolved waste can be absorbed by seaweed (Chopin et al., 2012). Therefore, increasing the number of trophically different species in IMTA systems can promote ecosystem stability (Campanati et al., 2022; Nederlof et al., 2021) and generate additional income through the production of economically valuable species (Knowler et al., 2020).

One of the main reasons for establishing an IMTA system is to make use of the nutrient waste from the species being fed. This approach aims to increase the productivity of the system per unit of feed, which would otherwise go to waste in monoculture systems (Ramli et al., 2020). Numerous studies have shown that IMTA systems are highly efficient in utilizing feed nutrient waste. For example, Nederlof et al. (2021) found that the closed model IMTA system absorbed around 65-75% of nitrogen (N) and 65% of phosphorus (P), which is higher than the open model with absorption levels of 50% N and 40% P. However, there are still challenges and needs associated with this integrated aquaculture system. These include economic species diversity and implementation methods (Felaco et al., 2020), the placement of species in nutrient distribution areas (Nederlof et al., 2021), and the temporal and spatial availability of nutrients (Camelo-Guarín et al., 2021). These challenges have led to the development of various IMTA system models to enhance our understanding of more effective IMTA practices.

This study integrated four functional species with significant economic value. The selection criteria for these species are primarily based on positive interactions between species from different trophic levels, namely the rabbitfish *Siganus guttatus* as a fed species, the green mussel *Perna viridis* as a suspension feeder, the sea urchin *Diadema setosum* as a deposit feeder, and the macroalga *Kappaphycus alvarezii* as a species that extracts inorganic nutrients. The potential nutrient waste from the three aquatic species includes leftover feed from rabbitfish, as well as feces and metabolic products of each species, which can spread throughout the aquaculture area. This waste ultimately ends up in the form of inorganic compounds, such as ammonia ( $\text{NH}_3\text{-N}$ ), ammonium ( $\text{NH}_4\text{-N}$ ), nitrite ( $\text{NO}_2\text{-N}$ ), nitrate ( $\text{NO}_3\text{-N}$ ), orthophosphate ( $\text{PO}_4\text{-P}$ ), and several other compounds (Abakari et al., 2021). Accumulated waste can trigger eutrophication, negatively impacting organisms and water quality (Boyd et al., 2020; Campanati et al., 2022). Therefore, the presence and performance of seaweed species as the main extractive species of inorganic compounds is crucial for assimilating as much of the dispersed nutrient waste as possible. Although it is known that water contains

microalgae and other microorganisms that can also contribute to nutrient removal (Abdelfattah et al., 2023; Shelknanloymilan et al., 2012), this experiment focuses on nutrient removal by seaweed.

In an IMTA system, the availability of nutrients that can be assimilated by seaweed as an extractive species depends on the feeding rate of the fed species. The relationship between feeding rate, growth, and aquaculture waste is complex, especially in monoculture systems (Kim et al., 2021; Niu et al., 2016). However, it is important to understand how this complexity translates to an IMTA system. Therefore, this study aimed to clarify the consistency of the complexity of feeding rate when practiced in an IMTA system. The study assessed several rabbitfish feeding rates to determine the ability of *K. alvarezii* to remove inorganic nutrients and the resulting biomass production in an integrated multi-trophic recirculating aquaculture system. While the horizontal method of seaweed cultivation has been widely investigated in the IMTA system and proven to absorb nutrients horizontally (Kang et al., 2021; Neori et al., 2004; Zhang et al., 2019), this study reared seaweeds vertically to maximize nutrient assimilation both horizontally and vertically. This is the first study to establish such a system and its goals. The findings of this study will be useful for expanding knowledge in exploring the potential for optimizing good practices in the IMTA system.

## Materials and Methods

### Experimental Design

The study was conducted at the Balik Diwa Institute of Maritime Technology and Business Laboratory, in Makassar City, Indonesia. The experimental setup consisted of four 120 L plastic tanks arranged in a way that allowed water to circulate by gravity from the top tank (rabbitfish), to the bottom tank (green mussel, sea urchin, seaweed). A ball valve was used to connect the tanks, and a water pump was specifically used to recirculate water from the seaweed tank to the rabbitfish tank.

In this experiment, a total of 36 tanks were used, with four tanks assigned to each experimental unit. The tanks for the green mussel were equipped with collector baskets, and coral fragments were added to the bottom of the tanks as a substrate, similar to the sea urchin tanks. Seaweed tanks were designed for vertical rearing, with two diagonal stretches for four hanging ropes. The seaweed was tied to each hanging rope in three columns with a vertical spacing of 10 cm. Figure 1 provides illustrations and pictures of the experimental design.

### Rearing Management

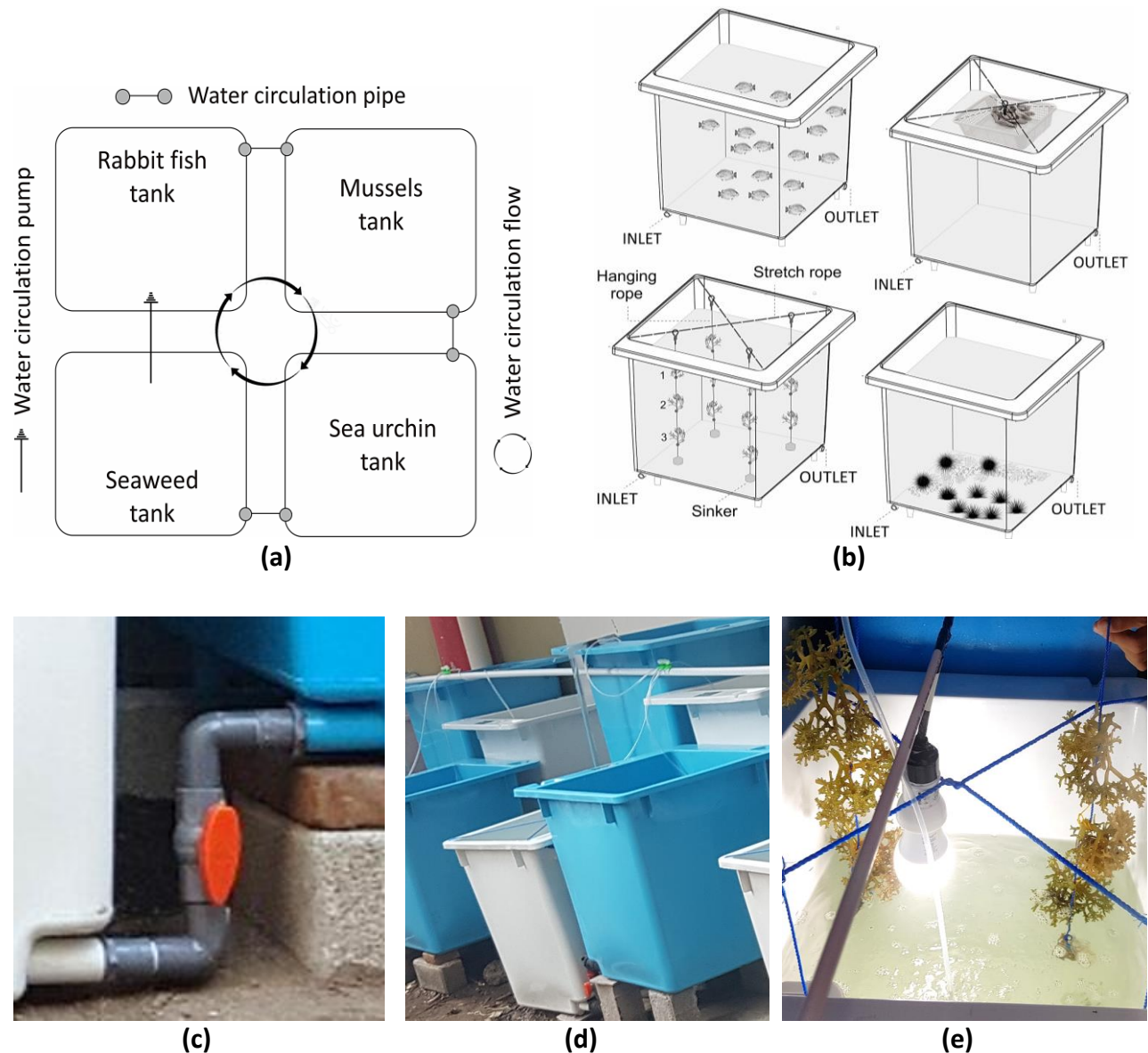
Juveniles of rabbitfish, with an initial weight of  $2.3 \pm 0.2$  g per fish, were commercially obtained from local hatcheries. Green mussels and sea urchins, with

initial weights of  $6.8 \pm 0.2$  g and  $30.7 \pm 0.3$  g per individual, respectively, as well as *K. alvarezii* seeds, were obtained locally from farmers. Each species was first adapted to the media and fed for 5 days before being stocked. In each tank with a volume of 80 L of water, 16 individual rabbitfish, 20 individual green mussels, 10 individual sea urchins, and  $20 \pm 0.2$  g per point of seaweed were randomly stocked. Green mussels, sea urchins, and seaweed were stocked after seven days of stocking of rabbitfish to allow the availability of nutrients in their rearing tanks.

The feeding rate levels for rabbitfish were divided into three groups: 5%, 7.5%, and 10% of biomass weight, with three replications. The fish were fed four times a day (8 am, 12 pm, 4 pm, and 8 pm). Despite being herbivorous with a protein requirement of 29–34% (Li et al., 2018), the commercial feed used in this study had a

protein content of 40% to provide organic and inorganic nutrients for other species. The weight of the rabbitfish was measured every week to adjust the amount of feed given.

Additionally, every two hours after feeding, the ball valve (0.5-inch pipe) in each tank is opened for two hours (10 am to 12 pm, 2 pm to 4 pm, 6 pm to 8 pm, and 10 pm to 12 am) to circulate the water by gravity. Simultaneously, a submersible pump (23 watts) was used to transfer water from the *K. alvarezii* tank to the rabbitfish tank. Dissolved oxygen was provided through continuous aeration from an LP60 blower with an aeration stone placed in the center of each tank. Light-Emitting Diodes (LED) Premium Lighting 1120-1260 Lumens were used to provide light in the *K. alvarezii* tanks.



**Figure 1.** Illustration of the experimental design: (a) scheme of the circulation flow, (b) set up of tanks, (c) tank connecting valve, (d) unit experimental, (e) seaweed tank.

**Data Collection**

During the study, water quality was carefully maintained within a tolerable range by rabbitfish, green mussels, sea urchins, and seaweed. To ensure this, we monitored various parameters daily in each tank using an AZ 86031 Water Quality Meter instrument. Additionally, we assessed the concentration of ammonia (NH<sub>3</sub>-N), ammonium (NH<sub>4</sub>-N), nitrite (NO<sub>2</sub>-N), nitrate (NO<sub>3</sub>-N), and orthophosphate (PO<sub>4</sub>-P) to measure dissolved inorganic compounds. We collected 100 mL water samples in polypropylene (PP) bottles on days 1, 7, and 49 and analyzed them in a laboratory following the protocols recommended by the American Public Health Association (APHA, 2017).

The organic compounds are further categorized into total ammonia nitrogen (TAN), dissolved inorganic nitrogen (DIN), and dissolved inorganic phosphorus (DIP). TAN is calculated as the sum of NH<sub>3</sub>-N and NH<sub>4</sub>-N (Abakari et al., 2021), DIN is obtained from the sum of NH<sub>4</sub>-N, NO<sub>2</sub>-N, and NO<sub>3</sub>-N (Osti et al., 2020), and DIP is represented by total PO<sub>4</sub>-P (Cotiyane et al., 2019). Furthermore, the nutrient removal efficiency of each group of dissolved inorganic compounds was quantified using the equation from Pham & Bui (2020) as follows:

$$NRE (\%) = \frac{C_0 - C_f}{C_0} \times 100$$

In the above equation, NRE refers to nutrient removal efficiency (%), C<sub>0</sub> is the 7<sup>th</sup> day concentration (mg L<sup>-1</sup>), and C<sub>f</sub> is the 49<sup>th</sup> day concentration (mg L<sup>-1</sup>).

In this study, we calculated the biomass production of *K. alvarezii* over a 42-day period at different feeding rates. It is important to note that the initial weight of the *K. alvarezii* stock was similar (20±0.2 g) and did not differ significantly (P>0.05) between all sampling points. Therefore, biomass production was determined by measuring the total final weight of the seaweed (Irhayyim et al., 2020) using a WH-B28 lectronic balance with an accuracy of 0.01 g. Weight was expressed in grams (g).

**Statistical Analysis**

The normal distribution and homogeneity of data variance were initially assessed using the Shapiro-Wilk test and Levene tests to ensure that the data met

parametric statistical assumptions. The results of the tests indicated that the data were normally distributed and showed homogeneity (P>0.05). Subsequently, one-way ANOVA was employed to examine the impact of feeding rate level on TAN, DIN, and DIP concentrations, NRE of TAN, DIN, and DIP, as well as biomass production of *K. alvarezii*. Significant effects were then compared using the Tukey honestly significant difference (HSD) test at a significance level of 95% (P<0.05). All statistical analyses were conducted using IBM SPSS Statistics Version 25 software.

**Results**

**Water Quality Parameters**

In this study of the recirculating system of IMTA, we summarized four general water quality parameters as average values during 42 days of vertical rearing of *K. alvarezii*. The means for all parameters were not significantly different (P>0.05) among feeding rate levels (Table 1).

**TAN, DIN, and DIP Concentrations**

At the start of the experiment (day 1), the concentrations of TAN, DIP, and DIP in the *K. alvarezii* tank were relatively low and did not differ significantly between feeding rate levels (Figure 2). However, by day 7, the mean concentrations of inorganic compounds for all groups had significantly increased. The concentrations of TAN, DIP, and DIP which increased with higher feeding rate levels, showing significant differences between the three feeding rate levels (P<0.05). By the end of the experiment (day 49), the concentrations of TAN, DIP, and DIP had significantly decreased, with no significant differences among the feeding rate levels (P>0.05).

**Nutrient Removal Efficiency (NRE) of TAN, DIN, and DIP**

In this study of the IMTA recirculation system, the efficiency of nutrient removal by *K. alvarezii*, including TAN, DIN, and DIP as a result of rabbitfish feeding rates, is presented in Table 2. The nutrient removal efficiency of vertically reared *K. alvarezii* was found to be above 95% for each feeding rate level. There was no significant difference (P>0.05) in the means removal percentage means for TAN, DIN, and DIP among feeding rate levels.

**Table 1.** The mean values of water quality parameters in the *Kappaphycus alvarezii* tank for 42-day period at different feeding rate levels

Water parameters	Feeding rate 5%	Feeding rate 7.5%	Feeding rate 10%
Salinity (ppt)	28.2±0.5 <sup>a</sup>	28.3±0.4 <sup>a</sup>	28.3±0.4 <sup>a</sup>
Dissolved oxygen (mg L <sup>-1</sup> )	5.9±0.9 <sup>a</sup>	6.2±1.0 <sup>a</sup>	6.4±1.0 <sup>a</sup>
Temperature (°C)	27.3±1.6 <sup>a</sup>	27.2±1.6 <sup>a</sup>	27.3±1.8 <sup>a</sup>
pH	7.7±0.3 <sup>a</sup>	7.8±0.2 <sup>a</sup>	7.8±0.3 <sup>a</sup>

Data are expressed as mean±standard deviation (SD) (n=3). The same superscript letter indicates no significant difference among feeding rate levels (P>0.05).

**Biomass Production of *K. alvarezii***

In this experiment, *K. alvarezii* was grown vertically, with 12 plants per tank (Figure 1b). Each tank had four hanging ropes, each with three columns and tie points. After 42 days, the final biomass of *K alvarezii* increased with higher feeding rate levels, showing a significant difference ( $P<0.05$ ) among the three feeding rate levels (Table 3). However, the final biomass between the planting columns did not show a significant difference ( $P>0.05$ ) for all feeding rates.

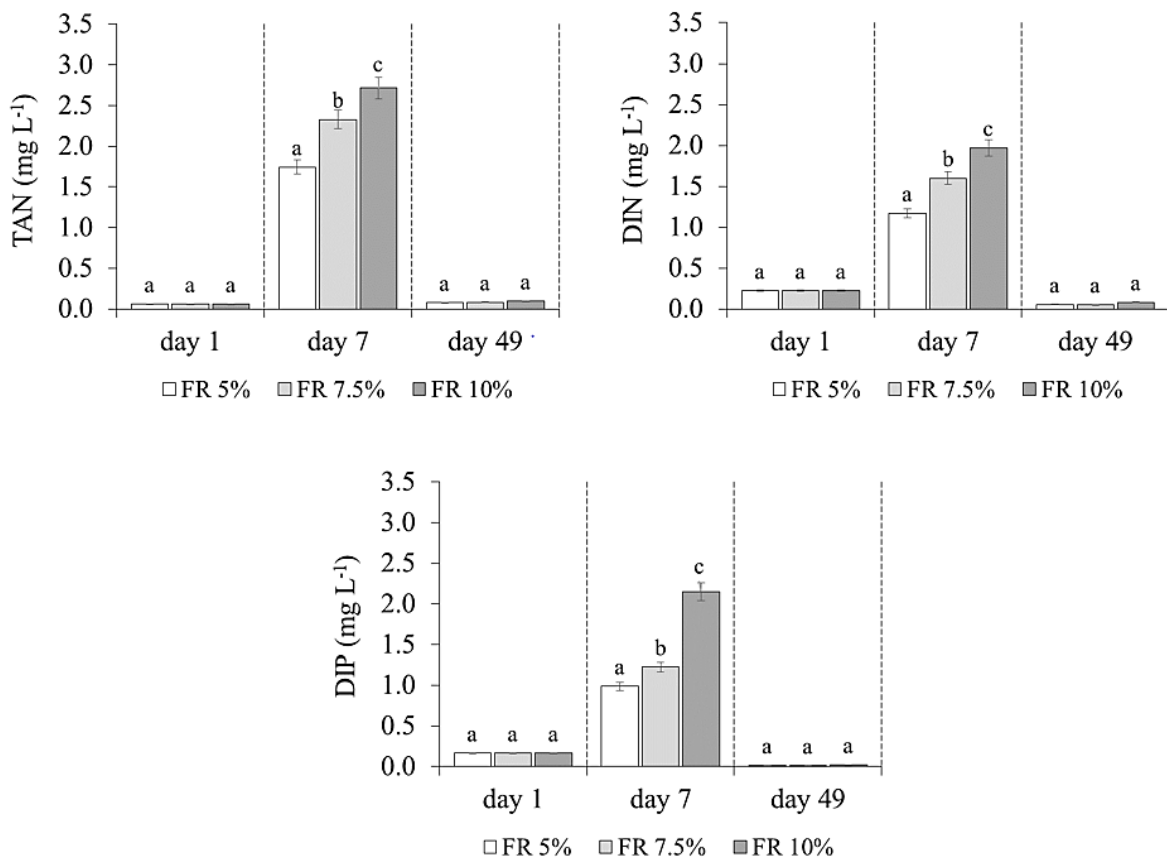
**Discussion**

The use of artificial feed in aquaculture has trade-offs in biological, economic and ecological aspects, making it an interesting focus of research (Heriansah et al., 2023). The amount of nutrients supplied in the diet has a significant impact on organism growth (Barani et al., 2019). However, artificial feed accounts for more than half of total production expenses, affecting profitability (Sanchez-Muros et al., 2020). Additionally, a significant portion of the feed supplied to aquaculture systems is wasted in the form of leftover feed and excretory wastes such as feces and urine (Dauda et al., 2019). Many reports have been published on the amount of nutrient waste generated by aquaculture

activities, primarily from feed. For example, rabbitfish retain only 25% of nitrogen (N) and 11% of phosphorus (P) from feed, with the remaining 52% of nitrogen (N) and 74% of phosphorus (P) being discarded into the water (Prihadi et al., 2020).

Feeding rate is a crucial factor in feeding management as it determines the daily amount of feed given to cultured organisms. Providing excessive feed can lead to increase waste and water quality deterioration, while insufficient feed can result in slow growth (Kim et al., 2021; Niu et al., 2016). In an intensive monoculture system, increasing the amount of feed does not always improve productivity as most of the feed is wasted. In our study, we used an integrated multi-trophic recirculation system to examine the ability of *K. alvarezii* seaweed to remove nutrients based on rabbitfish feeding rates.

In a 42-day laboratory study, the water quality parameters (salinity, dissolved oxygen, temperature, and pH) were found to be suitable for *K. alvarezii* at all feeding rate levels (Table 1). These parameters showed only minor fluctuations during the experiments and were within the recommended ranges for *K. alvarezii*, which include salinity 25–32 ppt, dissolved oxygen  $>4$  mg L<sup>-1</sup>, temperature 25–35°C, and pH 7–9 (Aris & Labenua, 2020). The observed parameters were conducive to optimal performance and nutrient uptake



**Figure 2.** The concentrations of total ammonia nitrogen (TAN), dissolved inorganic nitrogen (DIN), and dissolved inorganic phosphorus (DIP) in the *Kappaphycus alvarezii* tanks reared vertically. Different letters above the error bars indicate significant differences among feeding rate levels ( $P<0.05$ ).

in *K. alvarezii*. It is known that the condition of the aquaculture medium, particularly water quality parameters, is associated with nutrient removal efficiency (NRE).

Physiologically, salinity is related to osmotic pressure which can affect nutrient uptake performance (Choi et al., 2010). The concentration of dissolved oxygen in this study was always more than 5 mg L<sup>-1</sup> due to the constant aeration provided during the experiment and the ability of *K. alvarezii* to photosynthesize (Ajjabi et al., 2018). Additionally, the use of LED lighting in this study can increase photosynthesis activity (Huang et al., 2021). Temperature affects the kinetics of seaweed nutrient uptake, especially if there is no water change (Macchiavello & Bulboa, 2014). Similarly, pH can change the nutrient absorption activity of seaweed (Gao et al., 2018). The suitable conditions for various water quality indicators in this study may be related to the best performance of *K. alvarezii* in nutrient absorption at each feeding.

The most important components of aquaculture wastewater are nitrogen (N) and phosphorus (P), which come from dissolved feed and feces (Campanati et al., 2022; Chiquito-contreras et al., 2022). In this study, dissolved N and P elements are represented by the concentrations of compounds ammonia (NH<sub>3</sub>-N), ammonium (NH<sub>4</sub>-N), nitrite (NO<sub>2</sub>-N), nitrate (NO<sub>3</sub>-N), and orthophosphate (PO<sub>4</sub>-P), which are subsequently classified as TAN, DIN, or DIP concentrations. It was discovered that feeding rate influences the production of these three types of nutrients (Dauda et al., 2019). On day 7 (before green mussels, sea urchin, and seaweed were stocked), TAN, DIN, and DIP concentrations increased rapidly at feeding levels of 5%, 7.5%, and 10%, respectively (Figure 2). Temporally increasing nutrient concentrations are also found in many other recirculation system studies (Effendi et al., 2020; Sri-uam et al., 2016; Sun et al., 2014).

The rabbitfish in this study were fed a diet with 40% protein four times a day, at feeding rates of 5%, 7.5%, and 10%. Large amounts of nitrogen (N) and phosphorus (P) may be released in the form of food waste, feces, and metabolic products (Prihadi et al., 2020). Every two hours after feeding (for a total 8 hours a day), water is circulated successively to tanks for green mussels, sea urchins and seaweed. Therefore, the seaweed tank is the last tank in the water circulation process before it flows back into the rabbitfish tank. The increased concentration of inorganic nutrients in the water is a response to the accumulation of food waste, feces, and metabolic excretions from the rabbitfish (Melendres & Largo, 2021; Wang et al., 2012). This may explain the drastic increase in the concentrations of total ammonia nitrogen (TAN), dissolved inorganic nitrogen (DIN), and dissolved inorganic phosphorus (DIP) in the seaweed tanks on day 7, which also increased linearly based on the level of feeding rate.

On the eighth day of the experiment, each tank was stocked with green mussels, sea urchin, and seaweed to establish the IMTA system. Apart from leftover feed and rabbitfish feces, the presence of organic extractive species such as mussels and sea urchin can lead to an increase in nutrient waste due to the feces of these species (Grosso et al., 2021; Srisunont & Babel, 2015). However, the results indicated a significant decrease in the concentrations of TAN, DIN, and DIP at the end of the experiment in the seaweed tanks across all feeding rate levels. These findings are consistent with previous research on the ability of *K. alvarezii* to act as a waste bioremediator (Chiquito-contreras et al., 2022; Kang et al., 2021; Ramli et al., 2020).

Nutrient measurements were not taken every week in this study, but daily changes are possible following the stocking of extractive species (Effendi et al., 2020; Largo et al., 2016; Li et al., 2019). Despite an increase in the feeding rate, nutrient concentrations at

**Table 2.** The mean values of nutrient removal efficiency (NRE) by vertically reared *Kappaphycus alvarezii* for each nutrient group and feeding rate level

Level of feeding rate	Nutrient removal efficiency (NRE)		
	TAN	DIN	DIP
5%	96.1±0.7 <sup>a</sup>	95.2±0.9 <sup>a</sup>	98.5±1.0 <sup>a</sup>
7.5%	96.5±0.4 <sup>a</sup>	96.6±0.6 <sup>a</sup>	99.0±0.6 <sup>a</sup>
10%	96.7±0.4 <sup>b</sup>	96.8±0.4 <sup>a</sup>	99.0±0.5 <sup>a</sup>

TAN=total ammonia nitrogen, DIN=dissolved inorganic nitrogen, DIP=dissolved inorganic phosphorus (DIP). Data are expressed as mean±standard deviation (SD) (n=3). The same superscript letter indicates no significant difference among feeding rate levels (P>0.05).

**Table 3.** The mean final biomass of *Kappaphycus alvarezii* reared for 42 days vertically at different feeding rate levels

Biomass (g)	Feeding rate 5%	Feeding rate 7.5%	Feeding rate 10%
Column 1	235.2±3.8 <sup>a</sup>	256.8±2.4 <sup>b</sup>	278.0±3.2 <sup>c</sup>
Column 2	233.1±4.8 <sup>a</sup>	256.1±1.8 <sup>b</sup>	276.0±1.1 <sup>c</sup>
Column 3	232.4±5.5 <sup>a</sup>	248.9±3.4 <sup>b</sup>	279.7±0.8 <sup>c</sup>
Total (all column)	698.7±1.7 <sup>a</sup>	761.9±2.0 <sup>b</sup>	833.7±1.7 <sup>c</sup>

Data are expressed as mean±standard deviation (SD) (n=3). The different superscript letter indicates significant difference among feeding rate levels (P>0.05).

day 49 were much lower than at day 7. In a monoculture system, raising the feeding rate results in increased waste in the form of residual feed and excrement (Kim et al., 2021; Niu et al., 2016). However, the current study findings suggest that the consequences of this feeding rate are not an issue in multi-trophic cultivation systems, as the presence of green mussels and sea urchin likely contributed to the reduction in TAN, DIN, and DIP. However, several studies and reviews show that the main consumer of inorganic nutrients is seaweed (Arumugam et al., 2018).

The interesting findings in this study found that at the end of the trial, all feeding rates had statistically equal amounts of TAN, DIN, and DIP. This suggests that *K. alvarezii* can accumulate nutrients in tissues when concentrations are high in the water, which can then be used to meet metabolic needs if the water has a nutritional deficit (Rustam et al., 2017). Seaweed uptake rates have been shown to increase linearly with nutrient availability (Harrison & Hurd, 2001; Wang et al., 2023). Therefore, it is reasonable to assume that whatever nutrients are supplied at the feeding rate level will be fully digested by the seaweed if it can reach them. These findings highlight the efficiency with which seaweed absorbs nutrients.

The previous assumptions were confirmed by the average nutrient removal efficiency values for vertically reared *K. alvarezii*, which were all greater than 95% for each nutrient group and feeding rate (Table 2). Previous studies found that *K. alvarezii* reared horizontally absorbed 61.6% and 59.5% of total nitrogen and 3.4% and 5.5% of total phosphorus in IMTA recirculation system tanks and flow-through system cultures, respectively (Azad et al., 2017). A recent study found that horizontally grown *K. alvarezii* in tanks absorbed 81.6%  $\text{NH}_3\text{-N}$ , 94.6%  $\text{NO}_2\text{-N}$ , 94.5%  $\text{NO}_3\text{-N}$ , and 95.4%  $\text{PO}_4\text{-P}$  from intensive shrimp pond waste (Syahrir, 2024). *K. alvarezii* grown horizontally in fish cages has been reported to remove 30%  $\text{PO}_4\text{-P}$ , an inorganic nutrient (Kambey et al., 2020). The variations in findings between the prior study and the current study could be attributed to different system designs.

This study found that rearing *K. alvarezii* vertically can significantly reduce TAN, DIN, and DIP concentrations at all feeding rates. Nutrient waste from rabbitfish, green mussels, and sea urchins accumulated on the tank's surfaces, columns, and bottom (Azad et al., 2017; Bouwman et al., 2013). The tank set-up and vertical stocking method allows *K. alvarezii* to efficiently photosynthesize and assimilate scattered nutrients (Nursidi et al., 2017). Vertical distribution also increases stocking numbers, leading to more efficient nutrient absorption (Pong-Masak & Sarira, 2020). Additionally, the closed recirculation system ensures that nutrients in the form of feed waste, feces, and metabolic products are available for assimilation by *K. alvarezii*. Water flow, as reported by previous studies, also plays a crucial role in the efficiency of *K. alvarezii* nutrient uptake for biomass production (Azad et al., 2017).

The final total biomass of *K. alvarezii* in this study was influenced by the feeding rate level, with higher feeding rates resulting in higher biomass production, as shown in Table 3. The quantity of nutrients from different feeding rates is the main factor influencing these results. Previous studies have shown that seaweed growth increases linearly with nutrient availability (Harrison & Hurd, 2001; Wang et al., 2023). Interestingly, there was no consistent trend in terms of which column produced the highest biomass at each feeding rate level. This suggests that *K. alvarezii* can effectively assimilate nutrients from both the surface and columns of the rearing media for growth (Nursidi et al., 2017; Pong-Masak & Sarira, 2020). With the application of appropriate technology and design, vertical systems can be easily expanded to larger areas without the need for additional horizontal space, making them a method with promising scalability potential. This scalability provides opportunities for implementation in various locations with diverse aquatic conditions, including coastal areas, open seas, and even controlled aquaculture facilities, for both commercial production and sustainable aquaculture management.

The findings of this study offer valuable insights into the potential for developing seaweed aquaculture using a vertical approach in a multitrophic system. The application of this approach can increase production and productivity, as more seaweed can be cultivated in the same area in different columns, resulting in higher biomass production (Nursidi et al., 2017; Pong-Masak & Sarira, 2020), leading to increased yields. In addition, by maximizing space and resource efficiency, this method can reduce operational costs. This is a potential economic implication of the study findings, especially increased profits.

Additionally, the utilization of nutrients in the water column could help reduce aquaculture waste (Nursidi et al., 2017; Pong-Masak & Sarira, 2020), including waste generated from other species in the system. These findings may also address the challenges associated with the placement of species in nutrient distribution areas in Integrated Multi-Trophic Aquaculture (IMTA) systems (Nederlof et al., 2021), especially in the water column area. Thus, the vertical seaweed cultivation method can reduce costs associated with ecosystem restoration and environmental impact mitigation.

Furthermore, it is important to note that the IMTA system increases the number of species, thereby strengthening the economy and food production system resistance to sustainable seaweed aquaculture (Chopin et al., 2012; Zhang et al., 2019). A recent study found that IMTA-based seaweed is characterized as a source of improved biochemical composition, bioactive compounds, protein profiles, and techno-functional components (Machado et al., 2020). This is strategic for seaweed as a product that has economic value for food and non-food ingredients, as well as an important

ecosystem services role (Chopin & Tacon, 2021).

This study only measured nutrient removal efficiency in water and did not assess the extent to which nutrients are absorbed by seaweed tissue and other species involved. Additionally, the contribution of microalgae, which are also known to remove nutrients (Abdelfattah et al., 2023; Shelknanloymilan et al., 2012) should be considered. This highlights the importance of considering various factors that could contribute to the observed changes in this experiment. Therefore, further research is required to understand these aspect better. Additionally, this study was conducted at a laboratory scale, thus more laboratory experiments, and even field experiments, are necessary to ensure that this system performs effectively at a larger scale. Information from future research in this direction will be valuable and could provide comprehensive insights into how to minimize aquaculture waste, which is currently a critical issue in sustainable aquaculture (Amoussou et al., 2022; Nederlof et al., 2021).

## Conclusion

Even though the feeding rate given to rabbitfish as a fed species is higher, the nutrient waste in the rearing medium for *Kappaphycus alvarezii* is lower. Apart from the role of shellfish and sea urchins in the IMTA system, *K. alvarezii* cultivated using the vertical method can efficiently remove inorganic nutrient waste. These results demonstrate that this seaweed cultivation method can effectively optimize fish feed waste management in multitrophic cultivation systems, leading to sustainable aquaculture. However, we recommend conducting more experimental to ensure that this system works well on a field scale.

## Ethical Statement

All experimental protocols complied with the research guidelines and regulations at the Research Institute of the Balik Diwa Institute of Maritime Technology and Business. After the experiment was completed, the animals and macroalgae were handled by the Aquaculture Unit of the Balik Diwa Institute of Maritime Technology and Business for care and utilization.

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## Author Contribution

HH: Conceptualization, methodology, technical works, writing—original draft, reviewing and editing

manuscript. RS, NN, and SS: Conceptualization, methodology, supervision, reviewing and editing manuscript.

## Conflict of Interest

The authors declare that there are no conflicts of interest associated with this publication.

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