

Research Article

Bioremediation Potential of Brown Seaweed *Sargassum crassifolium* for Treating Wastewater in Lobster Culture

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Abstract

Wastewater generation from lobster culture systems has emerged as a serious environmental concern. *Sargassum crassifolium*, a naturally abundant yet underutilized brown seaweed species in the Northern Province of Sri Lanka, was evaluated in this study for its potential in wastewater bioremediation. This represents the first investigation in Sri Lanka employing a brown seaweed species for such a purpose. The research aimed to assess the efficiency of *S. crassifolium* biomass in removing nutrients from lobster culture effluents. Biomass optimization experiments were carried out using three different biomass densities (20, 30, and 40 g L⁻¹), where 20 g L⁻¹ was identified as the optimal biomass level for nutrient removal. Initial analyses included key physicochemical parameters of the wastewater ammonia, nitrite, nitrate, and phosphate concentrations. The nutrient removal experiment was performed in six glass tanks (60 × 30 × 28 cm) using the same volume of wastewater (20 L), comprising three replicates of the treatment and one control without seaweed. Experimental duration was 28 days and water quality measurements were taken at three-day intervals and once a week over 28 days. The results indicated that *S. crassifolium* effectively removed 87.59% of ammonia, 92.86% of nitrite, 62.5% of nitrate, and 50.42% of phosphate. Nutrient removal in the seaweed-treated tanks was significantly higher than in the control (P<0.05). Furthermore, seaweed biomass increased by 43.21% during the study period. Overall, the findings suggest that *S. crassifolium* serves as an efficient and environmentally sustainable approach for nitrogen and phosphorus removal from lobster culture wastewater, underscoring its potential use in eco-friendly aquaculture wastewater management systems.

Keywords: Bioremediation; Lobster aquaculture; Nutrient removal; *Sargassum crassifolium*; Wastewater treatment



1. Introduction

Around the world, communities have historically depended on capture fisheries as a primary source of protein. As a result of capture fishery production has been stagnant and unable to fulfill the increasing demand, aquaculture has become one of the most rapidly expanding food-producing industries globally, playing a vital role in fulfilling the increasing need for fish-based protein. A wide range of species, including shrimp, lobsters, crabs, sea cucumbers, seaweeds, tilapia, and carps, are cultured across various regions (Verdegem *et al.*, 2023).

Among these, mariculture, particularly the culturing of lobsters, has experienced notable expansion in recent decades. However, aquaculture effluents often contain high concentrations of nitrogen (N) and phosphorus (P), primarily derived from uneaten feed, fertilizers, fecal matter, and other metabolic wastes (Iber and Kasan 2021). The accumulation of these nutrients in receiving waters can lead to harmful algal blooms, eutrophication, oxygen depletion, and overall deterioration of water quality (Turcios and Papenbrock, 2014; Bambaranda *et al.*, 2019). Therefore, aquaculture wastewater treatment is a major part of the sustainable aquaculture (Liu *et al.*, 2019; Ojewole *et al.*, 2024).

In Sri Lanka, aquaculture is a growing economic activity supported by 2,905 km² of inland water resources and 190.5 km² of brackish water resources, primarily used for prawn and shrimp farming while lobster culturing has been identified as a potential aquaculture industry to promote rural income from sustainable aquaculture (Jayantha *et al.*, 2010; Stephen *et al.*, 2017; Basnayake and De Silva, 2025). However, inadequate wastewater handling in aquaculture operations has resulted in nutrient enrichment and various environmental issues, posing risks to sustainable development. In Sri Lanka, lobster culturing is an emerging sector within the aquaculture industry (Basnayake and De Silva, 2025). Lobster culturing, in particular, generates wastewater containing substantial amounts of organic and inorganic pollutants, which pose serious risks to adjacent coastal ecosystems especially in the Northern Province, where most lobster culturing activities are located (Devi *et al.*, 2007; Stephen *et al.*, 2017; Farag *et al.*, 2023; Basnayake and De Silva, 2025). The release of nutrient-rich effluents into coastal waters can alter local water chemistry, promote algal blooms, and threaten sensitive marine habitats such as seagrass beds and coral reefs.

Conventional wastewater treatment methods, chemical, physical and biological, have been employed to mitigate these impacts; however, the complex composition of aquaculture effluents often limits the effectiveness of any single approach (Ahmed and Thompson, 2019). Among these, biological treatment has gained increasing attention as an eco-friendly, cost-effective, and sustainable alternative for managing aquaculture wastewater (Ansari and Khan, 2010; Abonyi *et al.*, 2024).

Phycoremediation, the use of algae including macro algae such as seaweeds to remove dissolved nutrients and pollutants, has been recognized as an efficient, low-cost, and environmentally sustainable approach for aquaculture wastewater treatment (Mugg *et al.*, 2000; Guo *et al.*, 2015; Sultana *et al.*, 2024). Seaweeds, particularly brown macroalgae species are highly effective in assimilating inorganic nutrients such as ammonia, nitrite, nitrate, and phosphate, thereby improving overall water quality (Arumugam *et al.*, 2018; Sultana *et al.*, 2024). Among them, species of *Sargassum* have demonstrated strong potential for removing excess nutrients from aquaculture effluents and mitigating eutrophication risks (Mai *et al.*, 2010; Yu *et al.*, 2014). In Integrated Multitrophic Aquaculture (IMTA) systems, species of *Sargassum* help improve water quality while promoting more environmentally sustainable and balanced aquaculture practices (Zhu *et al.*, 2025).

The coastal waters of Sri Lanka are rich in seaweed diversity, with numerous species distributed across different coastal regions (Premarathna *et al.*, 2020). Abdulwahid *et al.* (2024) have reported that the dry powder of *Sargassum polycystum* can absorb nutrients from wastewater, especially nitrate, effectively. Among the native macroalgae, *S. crassifolium* is widely distributed along the Sri Lankan coast and shows great promise for use in phycoremediation because of its high capacity for nutrient absorption. The efficiency of nutrient removal is largely influenced by the optimization of seaweed biomass density (Rawat *et al.*, 2011; Kaewduang *et al.*, 2024). Given the limited research on wastewater treatment in lobster aquaculture, this study was conducted to assess *S. crassifolium*'s ability to remove nitrogen and phosphorus from lobster culture effluents and to identify the most effective biomass density for maximum nutrient removal.

2. Materials and Methods

2.1 Seaweed Collection and Preparation

S. crassifolium samples were collected from the Valaipadu coastal region in the Northern Province of Sri Lanka. Species identification was conducted based on morphological characteristics (Coppejans *et al.*, 2009). The harvested *S. crassifolium* specimens were carefully washed with seawater to eliminate epiphytes, debris, and other contaminants. The cleaned samples were then packed in High-Density Polyethylene (HDPE) bags filled with seawater and promptly transported to the laboratory at the National Aquaculture Development Authority (NAQDA) Regional Center in Kilinochchi, Sri Lanka, for further processing and experimental use.

2.2 Wastewater Collection

The study was carried out at a lobster culture system located in Point Pedro, Sri Lanka. Wastewater from the chosen culture system was collected in a 500 L high-density plastic container. At the time of collection, temperature, dissolved oxygen (DO), pH and salinity were measured using an EcoSense DO200A dissolved oxygen meter (YSI Inc., Yellow Springs, OH, USA), EcoTestr pH meter (Eutech Instruments, Singapore) and

MASTER-S/MillM salinity refractometer (ATAGO Co., Ltd., Tokyo, Japan). Ammonia, nitrite, nitrate, and phosphate were measured following standard procedures using Digital Aqua test kits (Sera), DR900 Multiparameter Portable Colorimeter (Hach Company, Loveland, CO, USA) and DR6000 UV-Vis Spectrophotometer (Hach Company, Loveland, CO, USA). The Winkler method was used to analyze Biological Oxygen Demand (BOD).

2.3 Optimization of Seaweed Biomass Density

The experiment was performed using twelve glass tanks (60 × 30 × 28 cm) set up in a Completely Randomized Design (CRD) with three replicates for each treatment. Good quality *S. crassifolium* thalli without physical damage were carefully chosen and introduced at three different biomass densities: 20, 30, and 40 g L⁻¹. Control tank maintained without seaweed. Each tank was filled with 20 L of the collected wastewater, and baseline water quality parameters, including temperature, pH, salinity, Dissolved Oxygen (DO), ammonia, nitrite, nitrate, and phosphate, were recorded before introducing the seaweeds. Water quality was monitored twice daily, in the morning and evening.

2.4 Nutrient Removal Efficiency

Six glass tanks (60 × 30 × 28 cm) were set up in a fully randomized design for the 28-day nutrient-removal experiment. *S. crassifolium* (20 g L⁻¹) was added to three tanks as the treatment tanks, while three tanks without seaweed were used as controls. Each tank held 20 liters of wastewater from lobster culture system. All of the tanks had constant aeration for the course of the experiment. Nutrient concentrations (ammonia, nitrite, nitrate and phosphate) were measured every three days and once a week using a digital colorimeter (DR900) following standard methods (Devi and Gowri, 2007).

Nutrient concentrations, including ammonia, nitrite, nitrate, and phosphate, were measured in each treatment and control tank on Days 0, 7, 14, 21, and 28 of the experiment. Before sampling, the wastewater in each tank was gently mixed to ensure homogeneity. Water samples were collected from the middle water column using clean sampling bottles and analysed immediately using a DR900 multiparameter portable colorimeter (Hach Company, Loveland, CO, USA) following the manufacturer's instructions and standard analytical procedures described by Devi and Gowri (2007). Nutrient concentrations were expressed as mg L⁻¹. Three replicate tanks were maintained for each treatment, and the mean values were used for subsequent statistical analysis.

2.5 Seaweed Growth Measurement

The initial (W1) and final (W2) biomass of *S. crassifolium* was recorded.

Biomass increase percentage was calculated using the following formula (Yong *et al.*, 2013).

$$\text{Biomass increase (\%)} = \frac{(W2 - W1)}{W1} \times 100$$

where:

W1= Initial wet seaweed biomass (g)

W2 = Final wet seaweed biomass (g)

Specific Growth Rate (SGR) was calculated using the following formula (Yong *et al.*, 2013).

$$SGR = \frac{\ln W_2 - \ln W_1}{T} \times 100$$

where:

W1= Initial wet seaweed biomass (g)

W2 = Final wet seaweed biomass (g)

T = Culture duration (days)

During the experimental period, evaporation losses were compensated for by adding distilled water to maintain consistent salinity levels across all treatments.

3. Results

3.1 Optimization of Seaweed Biomass Density

The results of the optimization trial reported clear variations in nutrient removal efficiency among the three tested biomass densities (20, 30, and 40 g L⁻¹) of *S. crassifolium* grown in 20 L of lobster culture wastewater every three days and every seven days for 28 days. The highest nutrient removal efficiency was recorded at the lowest density (20 g L⁻¹), particularly for ammonia and nitrite, compared to the 30 g L⁻¹ and 40 g L⁻¹ treatments (Table 1). Statistical analysis confirmed that nutrient removal efficiency at 20 g L⁻¹ was significantly higher than that of the other two densities ($p < 0.05$). Conversely, the lowest performance was observed at the 40 g L⁻¹ biomass, suggesting that higher stocking densities may reduce nutrient absorption efficiency due to self-shading or mass transfer limitations (Bambaranda *et al.*, 2019; Kaewduang *et al.*, 2024).

Table 1. Final nutrient concentrations in lobster aquaculture wastewater after three days of treatment with different biomass densities of *S. crassifolium*

Biomass (g L⁻¹)	Ammonia (mg L⁻¹)	Nitrite (mg L⁻¹)	Nitrate (mg L⁻¹)	Phosphate (mg L⁻¹)
Control	1.80 ± 0.01 ^a	0.23 ± 0.00 ^a	0.08 ± 0.01 ^a	3.90 ± 0.11 ^a
20	0.04 ± 0.01 ^b	0.01 ± 0.01 ^b	0.15 ± 0.00 ^b	3.90 ± 0.11 ^b
30	1.43 ± 0.00 ^c	0.17 ± 0.00 ^c	0.10 ± 0.00 ^c	3.03 ± 0.19 ^c
40	1.27 ± 0.01 ^d	0.14 ± 0.00 ^d	0.12 ± 0.00 ^d	2.25 ± 0.14 ^d

Values are means ± SD, (n = 3). Means with different superscript letters (a,b,c,d) within a column are significantly different at p < 0.05.

According to the observed trend, the percentage of nutrients removed reduced as seaweed stocking density increased. Similar findings were reported by a study evaluated *Ulva lactuca* biomasses of 2, 4, and 6 kg in 600 L of marine fishpond effluent (Cohen and Neori, 1991). He discovered that increasing biomass did not increase the effectiveness of nutrient removal. In Sailfin molly effluent, Kaewduang *et al.* (2024) found that the optimal stocking density for *Caulerpa lentillifera*, *Ulva rigida*, and *Gracilaria fisheri* was 30 g L⁻¹, after which nutrient removal decreased. These results collectively demonstrate that the optimal threshold differs among seaweed species and nutrient types, and that nutrient uptake efficiency is not directly correlated with biomass density.

The biomass-density trial showed that the 20 g L⁻¹ treatment produced markedly lower final ammonia and nitrite concentrations than the 30 and 40 g L⁻¹ treatments (Table 1), indicating that nutrient removal efficiency for these two nitrogen compounds declined at higher seaweed densities. One possible explanation is self-shading within densely stocked thalli, which may have reduced light penetration and photosynthetic activity, thereby limiting the energy available for nutrient assimilation (Bambaranda *et al.*, 2019; Kaewduang *et al.*, 2024). In addition, greater biomass density may have restricted water movement around the thalli and reduced nutrient diffusion to algal surfaces, resulting in less efficient uptake.

However, these mechanisms were not directly measured in the present study; therefore, they should be interpreted as possible explanations rather than confirmed causes. Moreover, nitrate and phosphate did not show the same consistent density-related pattern observed for ammonia and nitrite. This indicates that the influence of biomass density may vary according to nutrient type and highlights the need for future studies to measure light availability, dissolved oxygen, pH, water circulation, and nutrient uptake rates under different seaweed stocking densities.

Additionally, the thalli's surface-to-volume ratio and growth stage can affect its ability to absorb nutrients (Nayana *et al.*, 2023). The absorption efficiency of younger, fast-growing tissues is generally higher than that of mature biomass. Therefore, the observed nutrient removal performance may be explained by the imbalance between nutrient demand and

inadequate nutrient availability at greater stocking densities. All of these findings highlight how important it is to maintain an ideal biomass density in order to maximize seaweeds' capacity for biofiltration in wastewater treatment systems.

3.2 The Effectiveness of Nutrient Removal During the Course of the 28-day Treatment

The results indicate that all evaluated nutrient concentrations in the wastewater collected from the lobster culture system had significantly ($p < 0.05$) decreased after 28 days of treatment (Table 2). Over the course of the experiment, the biomass of *S. crassifolium* increased by 43.21%, indicating active nutrient uptake for growth. The efficiency of seaweeds as nutrient biofilters is supported by the same nutrient uptake behavior seen in *Gracilaria* sp. and *Ulva* sp. (Devi *et al.*, 2007).

The removal efficiency of nitrite was the highest at 92.86%, followed by ammonia 87.59%, nitrate (62.50%) and phosphate (50.42%). The substantial decreases ($p < 0.05$) attest to efficient absorption of dissolved nutrients of *S. crassifolium*. Since ammonium may be directly integrated into the synthesis of amino acids and proteins, it is a favored nitrogen source for many macroalgae (Kumar and Bera, 2020). The near-total elimination of nitrite further illustrates the strong biofiltration ability of *S. crassifolium* (Badraeni *et al.*, 2020). On the other hand, nitrate uptake was decreased, most likely because nitrate reduction to ammonium before assimilation requires more energy (Torres *et al.*, 2021).

Table 2. Nutrient removal efficiency of *S. crassifolium* under 20 g L⁻¹ biomass density after 28 days of wastewater treatment.

Parameter	Initial Concentration (mg L ⁻¹)	Final Concentration (mg L ⁻¹)	Removed (mg L ⁻¹)	Removal Efficiency (%)	p Value
Ammonia	1.37 ± 0.01 ^a	0.17 ± 0.01 ^b	1.19 ± 0.34	87.59 %	0.000
Nitrite	0.14 ± 0.00 ^a	0.01 ± 0.00 ^b	0.12 ± 0.03	92.86 %	0.000
Nitrate	0.24 ± 0.01 ^a	0.09 ± 0.00 ^b	0.14 ± 0.04	62.50 %	0.000
Phosphate	4.76 ± 0.02 ^a	2.36 ± 0.02 ^b	2.40 ± 0.72	50.42 %	0.001

Values are means ± SD (n = 3). Within each row, means with different superscript letters (a, b) are significantly different at $p < 0.05$.

The lowest removal efficiency (50.42%) was seen for phosphate, which might be explained by microbial competition, physiological restrictions in phosphorus storage, or phosphate release from senescing algal tissue during the experiment (Badraeni *et al.*, 2020). Co-cultivation with other macroalgae that have a higher affinity for phosphorus or optimizing water circulation to lessen re-release effects may increase the efficiency of phosphate uptake.

Figures 1, 2, 3 and 4 show the changes in ammonia, nitrite, nitrate and phosphate concentrations over time during treatment with 20 g L⁻¹ of seaweed and in the control. The horizontal axis indicates the treatment days and the vertical axis indicates the nutrient concentrations in discharged waste water. According to the graphs the reduction of ammonia, nitrite, nitrate and phosphate concentrations in the treated tanks with the seaweed indicated a rapid decline at the beginning and then it continued progressively up to 28th date. The present results revealed that *S. crassifolium* has made a significant contribution to the rapid decline of ammonia concentration in the waste water.

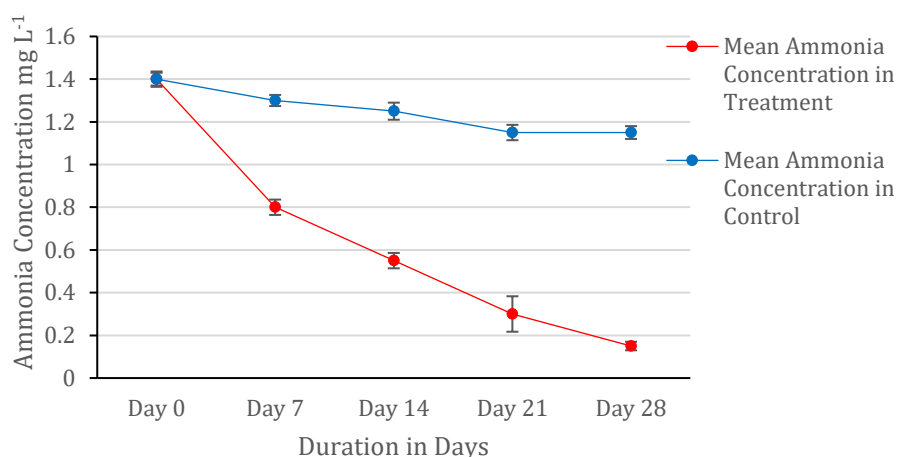


Figure 1. Changes in Ammonia concentration over time during treatment with 20 g L⁻¹ of seaweed and in the control

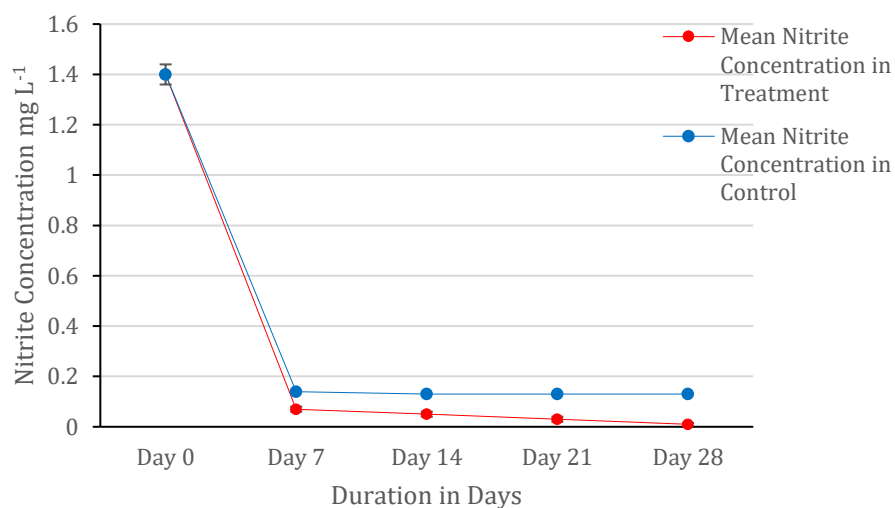


Figure 2. Changes in Nitrite concentration over time during treatment with 20 g L⁻¹ of seaweed and in the control

Notably, nitrite concentration also declined substantially in the control tanks, particularly during the first seven days of the experiment. This indicates that part of the reduction in nitrite concentration may have resulted from processes independent of the seaweed treatment. Since all tanks contained the same lobster-culture wastewater and received continuous aeration, natural microbial activity, including the conversion of nitrite to other nitrogen forms, may have contributed to the observed decline in the control. However, after Day 7, nitrite concentrations in the seaweed-treated tanks continued to decrease gradually, reaching nearly zero by Day 28, whereas concentrations in the control tanks remained relatively stable. This pattern suggests that the presence of *S. crassifolium* may have contributed to additional nitrite removal or supported conditions favourable for nutrient transformation. Nevertheless, because microbial activity and other water-quality variables were not measured, the specific mechanism cannot be confirmed. Therefore, the results should be interpreted as showing lower residual nitrite concentrations in the seaweed-treated tanks rather than attributing the entire reduction solely to seaweed uptake.

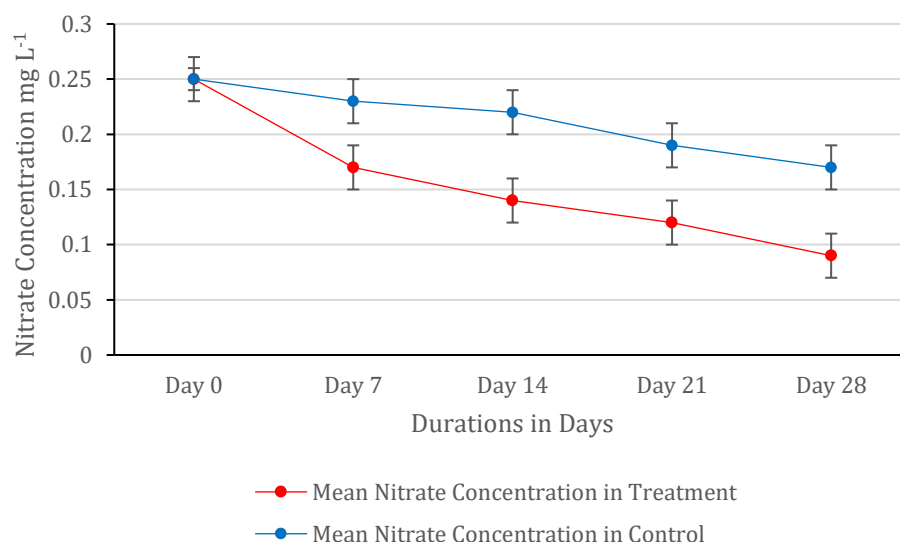


Figure 3. Changes in Nitrate concentration over time during treatment with 20 g L⁻¹ of seaweed and in the control

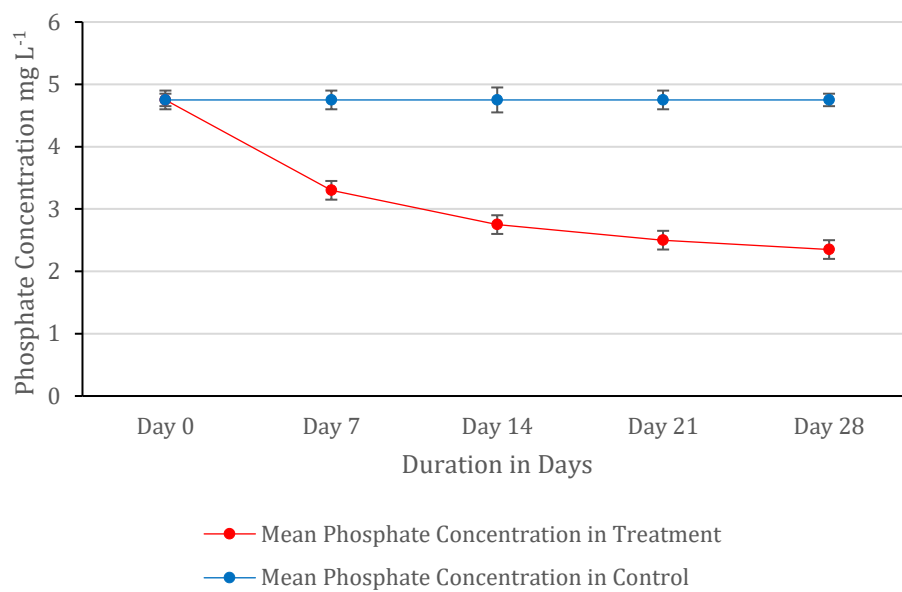


Figure 4. Changes in Phosphate concentration over time during treatment with 20 g L⁻¹ of seaweed and in the control

3.3 Factors Affecting the Absorption of Nutrients

The physiological and ecological traits of *S. crassifolium* can be used to explain the observed patterns of nutrient removal. Brown seaweeds absorb nitrogen mostly through active transport systems that include nitrate and ammonium transporters. The glutamine synthetase–glutamate synthase (GS–GOGAT) pathway then facilitates absorption (Liu *et al.*, 2019). The availability of light, photosynthetic efficiency, and thallus health all affect how well these processes work. As previously mentioned, high stocking densities can lower these characteristics through competition and shadowing.

Furthermore, nitrification, the process by which ammonia is converted to nitrite and nitrate, which the seaweed can then absorb, may be indirectly facilitated by the biofilm and microbial communities connected to the surfaces of *S. crassifolium*. These synergistic interactions between macroalgae and microbial populations improve the overall efficiency of nutrient removal in integrated systems (Mao *et al.*, 2025).

3.4 Comparative Analysis and Real-World Consequences

The current study's nitrogen removal efficiencies, when compared to earlier research, fall within the range documented for other seaweeds utilized in the treatment of aquaculture wastewater. For example, during 20 days, *Gracilaria verrucosa* eliminated up to 94.5% nitrite and 99.3% ammonia from aquaculture effluent (Devi *et al.*, 2007). In fish culture systems, *Ulva rigida* also removed 75% of the nitrogen (Kaewduang *et al.*, 2024). The findings imply that *S. crassifolium* has a great chance of being used as a natural biofilter in integrated multi-trophic aquaculture (IMTA) systems.

4. Conclusions

This study showed that wastewater nutrient concentrations declined in both the seaweed-treated and control tanks during the 28-day experiment. Therefore, the observed reductions cannot be attributed solely to direct uptake by *S. crassifolium*, as aeration, microbial activity, and other physicochemical processes may also have contributed to nutrient transformations. However, tanks containing *S. crassifolium* at 20 g L⁻¹ showed lower final concentrations of ammonia, nitrite, nitrate, and phosphate than the control tanks, indicating that the seaweed treatment was associated with enhanced nutrient reduction.

Among the biomass densities tested, 20 g L⁻¹ produced the most favourable nutrient-removal performance. In the seaweed-treated tanks, the calculated reductions were 87.59% for ammonia, 92.86% for nitrite, 62.50% for nitrate, and 50.42% for phosphate, while seaweed biomass increased by 43.21%. These findings indicate that *S. crassifolium* has potential as a supplementary biofilter in lobster-culture wastewater treatment systems. Future studies should include measurements of microbial activity, dissolved oxygen, pH, and nutrient transformations in both treatment and control tanks, and should evaluate the performance of *S. crassifolium* at pilot scale under practical aquaculture conditions.

Author Contributions

Conceptualization, R.M.G.N.R., A.C., and B.V.A.S.M.B; software, W.R.A.M.F; validation, W.R.A.M.F., and B.V.A.S.M.B; formal analysis, K.P., and A.A.D.O.H; investigation, K.P., and B.N; data curation, K.P; writing—original draft preparation, K.P; writing, review and editing, K.P., A.A.D.O.H., B.N., R.M.G.N.R., A.C., B.V.A.S.M.B; visualization, K.P., and A.A.D.O.H; supervision, B.N., R.M.G.N.R., A.C., B.V.A.S.M.B; project administration, B.N., and B.V.A.S.M.B; funding acquisition, K.P. All authors have read and agreed to the published version of the manuscript.

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