

Growth Mechanism and Carbon Sequestration of *Gracilaria verrucosa*

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Abstract

Gracilaria verrucosa, a commercially valuable red macroalga, plays a pivotal role in coastal aquaculture and carbon sequestration. This study investigates the effects of varying initial seedling weights (50 gram, 75 gram, 100 gram, and 125 gram/tie) on biomass yield, specific growth rate (SGR), and carbon sequestration over a 49-day cultivation period. Conducted in a floating raft system under controlled coastal conditions in South Sulawesi, Indonesia, the experiment followed a completely randomized design (CRD) with three replications per treatment. Biomass was harvested at seven-day intervals and assessed for dry weight, growth rate, and carbon composition using standardized methods and elemental analysis protocols. The results indicate that the 125 grams/tie treatment yielded the highest biomass, suggesting that larger initial weights enhance growth through enhanced nutrient uptake. However, the highest SGR and carbon sequestration were recorded in the 50 grams/tie treatment, highlighting the impact of planting density on physiological performance and carbon sequestration. The 100 grams/tie treatment presented the most balanced profile, maintaining high biomass while avoiding significant declines in SGR or carbon sequestration. These findings reveal a critical trade-off between economic and ecological outcomes, with implications for seaweed-based climate mitigation strategies and sustainable aquaculture design. Optimizing planting density can improve both yield and ecosystem services, aligning cultivation strategies with environmental sustainability objectives.

Keywords: *Gracilaria verrucosa*; biomass; carbon sequestration; growth rate; planting density

INTRODUCTION

Seaweeds have garnered increasing attention in recent years due to their multifaceted contributions to ecological sustainability and economic productivity, particularly in coastal aquaculture systems. Among the various macroalgal species, *Gracilaria verrucosa* has emerged as a key aquaculture commodity in Southeast Asia, especially Indonesia, due to its high economic value as a primary source of agar, widely used in the food, cosmetic, and pharmaceutical industries (Mouedden *et al.*, 2024; Purwanti *et al.*, 2024). Beyond its industrial utility, *G. verrucosa* serves vital ecological functions in marine ecosystems, such as improving water quality through nutrient absorption and providing habitat for marine organisms (Radulovich *et al.*, 2015; Chai *et al.*, 2018; Martínez-Curci *et al.*, 2023). Macroalgae, including *Gracilaria* species, are significant in the global carbon cycle, particularly through carbon sequestration in coastal ecosystems. Their high photosynthetic capacity enables them to capture substantial amounts of CO₂, contributing to blue carbon storage and climate change mitigation (Wang *et al.*, 2021; Pessarrodona *et al.*, 2023; Friess *et al.*, 2022). Studies have shown that nutrient-rich environments enhance both growth rates and carbon absorption efficiency in seaweeds (Radulovich *et al.*, 2015). Additionally, seaweeds help maintain ecological balance by assimilating dissolved inorganic nitrogen and phosphorus, mitigating the adverse effects of eutrophication from aquaculture and land-based runoff (Rejeki *et al.*, 2016; Papageorgiou *et al.*, 2023; Zhou *et al.*, 2022; García-Poza *et al.*, 2022).

Despite these benefits, the cultivation efficiency of *G. verrucosa* is influenced by environmental and physiological factors. The initial weight of seaweed seedlings has been identified as a key factor affecting photosynthetic efficiency, growth performance, and carbon assimilation. Seedling density, impacting intra-species competition for light and nutrients, is another critical factor. High densities can lead to shading and reduced photosynthesis (Ross *et al.*, 2024), while low densities may result in underutilization of space and suboptimal biomass yield. Therefore, identifying the optimal seedling weight to balance growth efficiency and carbon sequestration is essential. Gaps in the existing literature include limited studies on specific cultivation conditions, long-term effects on ecosystem

health, and the potential of polyculture systems to enhance resilience and productivity (Rahim *et al.*, 2023; Mantri *et al.*, 2022).

Although environmental factors like light, salinity, and nutrients have been studied for macroalgal growth (Ashkenazi *et al.*, 2022; Frieder *et al.*, 2022; Bermejo *et al.*, 2023; Wang *et al.*, 2023), limited data exist on how planting density affects the physiological performance of *G. verrucosa*. This gap restricts efforts to optimize cultivation for both economic gain and ecosystem services. Moreover, initial seedling weights and intra-species competition directly affect photosynthetic capacity, growth efficiency, and physiological stress in red macroalgae (Alamoudi *et al.*, 2022). Planting density significantly influences biomass yield and carbon sequestration, but thresholds exist beyond which competition may reduce growth rates (Rahim *et al.*, 2023). This study investigates the impact of initial seedling weights (50, 75, 100, and 125 grams) on biomass yield, specific growth rate (SGR), and carbon sequestration in a coastal aquaculture setting in South Sulawesi, Indonesia. By analyzing both growth and carbon metrics, the study aims to determine the optimal planting density that balances biomass production and carbon sequestration. The novelty of this research lies in its dual focus considering planting density as both a production and ecosystem service variable, highlighting its importance for sustainable, productive, and climate-resilient aquaculture systems.

MATERIALS AND METHODS

This study was conducted at the Maranak Aquaculture Research Facility in Takalar Regency, South Sulawesi, Indonesia, characterized by a coastal environment with sandy-mud substrates and proximity to coral reef ecosystems, favorable for macroalgal growth due to nutrient availability, moderate hydrodynamics, and stable physicochemical conditions. The sea surface temperature (SST) ranged from 28 to 30°C, optimal for *G. verrucosa* growth (Radulovich *et al.*, 2015; Ross *et al.*, 2024). Salinity ranged from 32 to 34 ppt, supporting osmoregulation in *Gracilaria* species (Nguyen *et al.*, 2017). Light intensity varied from 1,500 to 2,500 lux, affecting photosynthesis. The site was selected based on minimal wave disturbance, easy access, and successful cultivation history.

A Completely Randomized Design (CRD) was employed in this study due to its simplicity and effectiveness in minimizing allocation bias in macroalgal growth experiments (Martínez-Curci *et al.*, 2023; Rahim *et al.*, 2023). CRD is ideal for settings with homogeneous environmental conditions, enabling independent assessment of treatment variables. Four initial planting densities were tested: 50 grams/tie, 75 grams/tie, 100 grams/tie, and 125 grams/tie, labeled as treatments A through D. Each treatment was replicated three times, resulting in twelve experimental units. Seaweed cultivation was performed using a floating raft system with polyethylene ropes, which is commonly used in tropical aquaculture for its adaptability and ability to optimize nutrient uptake and reduce sediment interference (Wu *et al.*, 2023). Biomass measurements were taken at seven-day intervals over 49 days, capturing growth trends and carbon assimilation.

Biomass yield was quantified at each harvest point by weighing the freshly harvested samples. These samples were then sun-dried for three consecutive days and oven-dried at 60°C for 24 hours to achieve a constant dry weight. This method ensures consistency and eliminates moisture-related variability (Caroca-Valencia *et al.*, 2023). For carbon sequestration analysis, dried samples were pulverized using a mechanical grinder to produce a fine powder, which was analyzed using a CHN elemental analyzer. This combustion-based technique accurately quantifies the carbon, hydrogen, and nitrogen content of organic materials, widely recognized for its precision and reliability in evaluating the carbon sequestration capacity of macroalgal biomass (Ilac *et al.*, 2024).

The specific growth rate (SGR) was calculated at each harvest point using the logarithmic growth formula:

$$\text{LPH} = \frac{\ln\left(\frac{W_t}{W_0}\right)}{t} \times 100$$

Note: W_t = final dry weight (g), W_0 = initial dry weight (g), and t = the number of days between measurements; this formula measures relative growth and reflects growth efficiency over time intervals, a commonly used method in marine plant research (Rahim *et al.*, 2023).

Data Analysis

All data were analyzed using inferential statistics to test treatment effects. A one-way ANOVA assessed the influence of initial seedling weight on biomass yield, SGR, and carbon sequestration, appropriate for CRD (Martínez-Curci *et al.*, 2023). Tukey's HSD test followed ANOVA to identify significant pairwise differences. Analyses were conducted using SPSS and R at 95% confidence ($p < 0.05$).

RESULTS AND DISCUSSION

Biomass yield increased progressively across all treatments throughout the 49-day cultivation period. As shown in Table 1, larger initial seedling weights consistently promoted greater biomass accumulation, with the highest yield observed in the 125 grams/tie treatment. This trend supports previous findings that higher initial biomass enhances competitive ability and resource capture (Mishra & Singh, 2022).

Table 1. Biomass Yield During the Planting Cycle

Planting Density (grams/tie)	Day 7 (grams/tie)	Day 14 (grams/tie)	Day 21 (grams/tie)	Day 28 (grams/tie)	Day 35 (grams/tie)	Day 42 (grams/tie)	Day 49 (grams/tie)
50 grams/tie	58.53	67.73	79.33	91.00	98.00	107.67	121.00
75 grams/tie	87.70	101.63	119.33	129.33	143.67	159.67	167.67
100 grams/tie	116.40	131.07	145.33	158.67	172.33	186.67	201.67
125 grams/tie	145.47	164.17	180.33	198.00	217.33	231.33	247.33

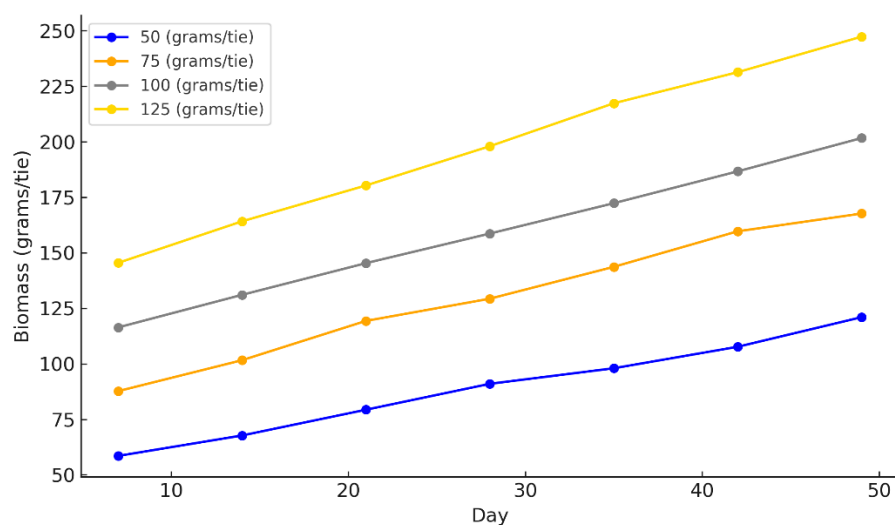


Figure 1. Biomass Yield During the Planting Cycle

The results show a clear trend: the 125 grams/tie treatment yielded the highest biomass at the end of the study, with 247.33 grams/tie. This aligns with the findings of Mulyaningrum *et al.* (2019), which suggested that larger initial seedling weights facilitate faster establishment and increased nutrient uptake. However, while the 125 grams/tie treatment showed the highest biomass, the 100 grams/tie treatment demonstrated a more stable growth pattern, indicating that moderate initial densities can lead to more balanced growth. These findings highlight the trade-off between maximizing biomass yield and sustaining growth efficiency over time, especially under nutrient limitations (Mishra & Singh, 2022). The increased biomass at higher planting densities can be explained by enhanced nutrient uptake, as larger initial biomass enables the seaweed to absorb nutrients more efficiently in the early stages of growth. This also highlights the necessity of balancing planting density to prevent shading and nutrient depletion, which are common challenges in denser systems (Chai *et al.*, 2018; Radulovich *et al.*, 2015).

The SGR results are summarized in Table 2. The data demonstrate that the 50 grams/tie treatment consistently achieved the highest SGR values, starting at 2.25%/day on Day 7 and gradually declining to 1.80%/day by Day 49. This trend indicates that lower planting densities reduce intra-specific competition and enable more efficient resource use, thereby enhancing photosynthetic activity (Purnomo *et al.*, 2022; Chowdhury *et al.*, 2023).

These results align with the findings of Radulovich *et al.* (2015) and Mulyaningrum *et al.* (2019), which indicated that lower initial densities reduce competition for light and nutrients. As biomass accumulates, however, competition for limited resources intensifies, leading to a decline in SGR across all treatments. The superior SGR in the 50 grams/tie treatment can be attributed to better access to light and nutrients. Interestingly, moderate densities such as 75 grams/tie showed a more stable growth pattern, suggesting that intermediate planting densities may optimize both growth and yield (Liu & Iersel, 2023; Tarsa *et al.*, 2022).

Table 2. Specific Growth Rate (SGR)

Planting Density (grams/tie)	Day 7 (%/day)	Day 14 (%/day)	Day 21 (%/day)	Day 28 (%/day)	Day 35 (%/day)	Day 42 (%/day)	Day 49 (%/day)
50 grams/tie	2.25	2.17	2.20	2.14	1.92	1.82	1.80
75 grams/tie	2.24	2.17	2.21	1.95	1.85	1.80	1.64
100 grams/tie	2.17	1.93	1.78	1.65	1.56	1.49	1.43
125 grams/tie	2.17	1.95	1.75	1.64	1.58	1.46	1.39

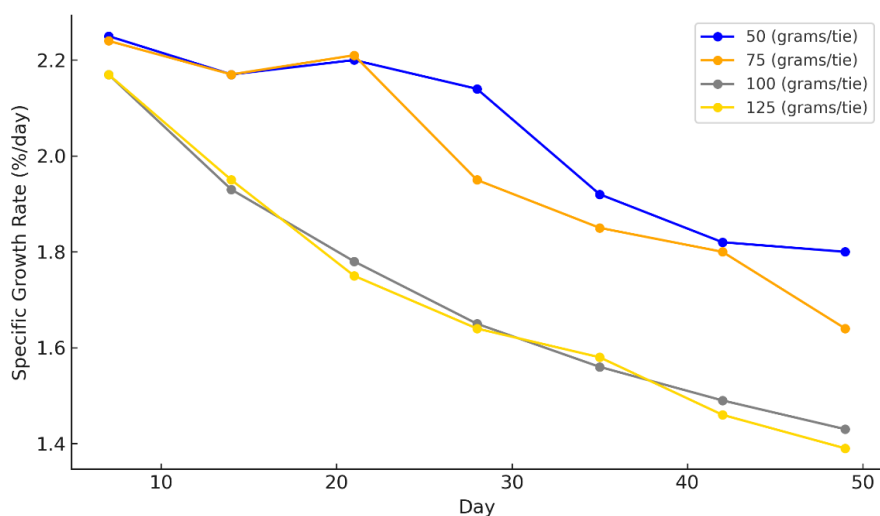


Figure 2. Specific Growth Rate (SGR)

The analysis revealed an inverse relationship between planting density and carbon sequestration, as shown in Table 3. The 50 grams/tie treatment achieved the highest sequestration rate (26.95%), highlighting how lower competition enhances carbon assimilation capacity (Arcanjo *et al.*, 2023; Chowdhury *et al.*, 2023).

This result is consistent with Radulovich *et al.* (2015) and Caroca-Valencia *et al.* (2023), which showed that lower planting densities promote higher photosynthetic efficiency. In higher-density treatments, shading and nutrient depletion hinder photosynthesis, reducing carbon sequestration potential (Le & Wei, 2023). The interaction between shading, nutrient uptake, and intra-specific competition plays a crucial role, creating thresholds where further increases in density no longer improve yield but decrease sequestration (Cai *et al.*, 2022; Zhu *et al.*, 2023). These findings are particularly relevant for blue carbon strategies, where optimizing efficiency is a key objective (Bennett *et al.*, 2023; Fakhraei *et al.*, 2020).

The statistical summary (Table 4) supports observed trends in biomass, SGR, and carbon sequestration. The 125 grams/tie treatment had the highest biomass, while the 50 grams/tie treatment showed the best SGR and carbon sequestration. ANOVA confirmed significant differences ($p < 0.05$), with Tukey's HSD showing pairwise significance between 50 and 125 g/tie treatments. These outcomes illustrate the inherent trade-offs between yield and efficiency, underscoring the importance of balanced cultivation strategies in sustainable aquaculture (Rahim *et al.*, 2023; Le & Ying, 2023).

Table 3. Carbon Sequestration Summary

Planting Density (grams/tie)	Day 49 (%C)
50 grams/tie	26.95
75 grams/tie	26.51
100 grams/tie	24.57
125 grams/tie	22.59

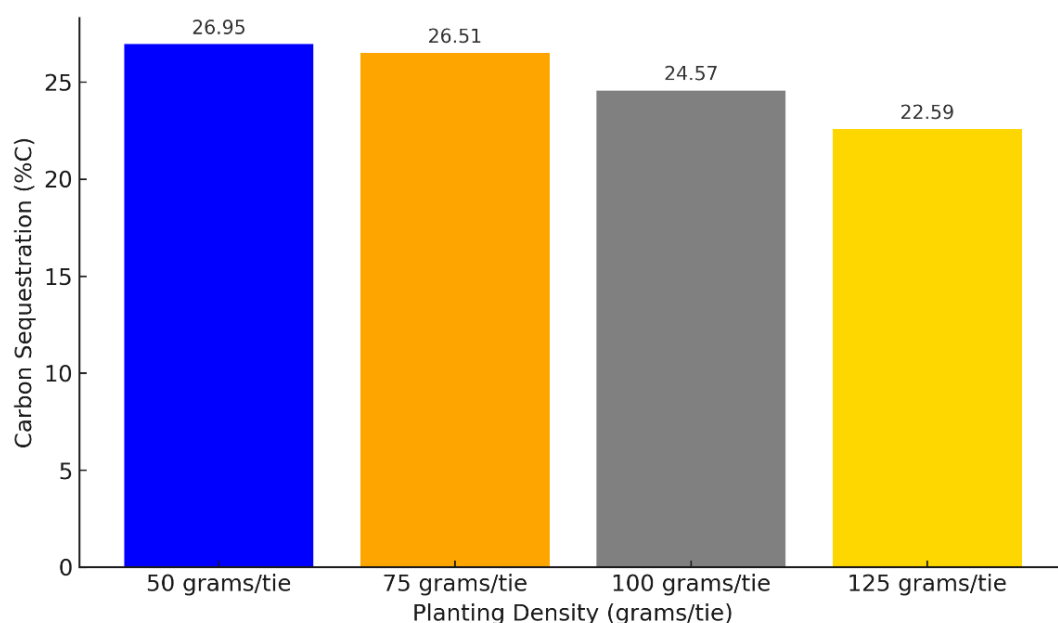


Figure 3. Carbon Sequestration

Table 4. Statistical Summary (Mean $\pm$ SD) after 45 Days

Initial Seeding	50 grams/tie	75 grams/tie	100 grams/tie	125 grams/tie
Biomass (grams/tie)	71 $\pm$ 1.00	93 $\pm$ 3.22	102 $\pm$ 3.79	122 $\pm$ 1.53
SGR (%/day)	2.04 $\pm$ 0.01528	1.98 $\pm$ 0.01528	1.72 $\pm$ 0.01528	1.70 $\pm$ 0.01528
Carbon sequestration (%C)	26.95 $\pm$ 1.64	26.51 $\pm$ 0.33	24.57 $\pm$ 1.77	22.59 $\pm$ 0.85

CONCLUSION

This study reveals that initial planting density influences *G. verrucosa*'s growth and carbon sequestration, with 125 g/tie maximizing biomass and 50 g/tie enhancing SGR and carbon sequestration. The 100 g/tie treatment offers a balanced yield without compromising carbon efficiency. These findings support seaweed's role in blue carbon strategies and aquaculture optimization through ecological-commercial integration.

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