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Research Article

Biostimulant potential from seaweed (*Kappaphycus alvarezii*) on rice seedling development

La Ode Afa [1,*], Nini Mila Rahni [1], Andi Awaluddin [1], Wa Ode Nuraida [1], La Ode M. Aslan [2], Noor Akbar Al-Ubaidah [1], and Paisal [1]

[1] Department of Agrotechnology, Faculty of Agriculture, Universitas Halu Oleo, Jl. HEA Mokodompit Kampus Hijau Bumi Tridharma Anduonohu, Kendari, Sulawesi Tenggara 93232, INDONESIA
[2] Department of Aquaculture, Faculty of Fisheries and Marine Science, Universitas Halu Oleo, Jl. HEA Mokodompit Kampus Hijau Bumi Tridharma Anduonohu, Kendari, Sulawesi Tenggara 93232, INDONESIA
* Corresponding author (✉ laodeafafpuho@gmail.com)

ABSTRACT

Seaweed (Kappaphycus alvarezii) liquid extract contains hormones, and macro and micronutrients that might benefit plant growth and development. This study aimed to evaluate the effect of a biostimulant liquid of seaweed extract on the vigor of rice seedlings. The research was conducted in the experimental field of the Faculty of Agriculture, Halu Oleo University, Kendari, Southeast Sulawesi. A factorial treatment design was conducted with a randomized block design. The first factor was the seedling method (wet sowing (S1) and dry sowing (S2)), and the second factor was six biostimulant concentrations (no biostimulant (K0), 0.8% (K1), 1.6% (K2), 2.4% (K3), 3.2% (K4), and 4.0% (K5)). The observed variables included seedling height, leaf area, number of tillers, root dry weight, and seedling dry weight. Data were analyzed using ANOVA, and DMRT at α=0.05. The results showed that the wet sowing method produced better seedling vigor than the dry sowing method after 14 days. The wet sowing method combined with a 0.8% seaweed extract resulted in the highest seedling vigor. The present study concluded the biostimulant potential of seaweed extract for rice.

Keywords: Dry sowing, *Oryza sativa*, seedling vigor, sowing method, wet sowing

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INTRODUCTION

Rice (*Oryza sativa* L.) is an important staple food of the world, including Indonesia; it provides 63% of the total energy and 37% of the protein requirements (David et al., 2020). Different rice varieties adapt to various agroecological conditions, such as rainfed, deep-water, tidal, swamp land, dry land, and irrigated fields (Widjajanto et al., 2021).

One key factor in increasing rice productivity across different agroecological zones is the vigor of seeds or seedlings (Afa et al., 2020). Vigorous seeds or seedlings can better adapt to environmental stresses and are crucial for increasing yields by 20%-30% (Adri et al., 2019). Seedling preparation for rice transplanting usually takes 14 to 28 days using dry or wet methods. Dry sowing is a rice planting method that does not require flooding, maintaining soil moisture at field capacity, which promotes better root development. In contrast, wet sowing involves flooding or semi-flooded soil, resulting in poor soil aeration. Transporting seedlings from the nursery to the planting field is generally more difficult with the wet sowing method, often leading to physical damage to the seedlings. The dry sowing method offers a more efficient alternative due to its lighter weight, easier transportation, and improved seedling adaptability (Wang et al., 2024). Proper management during the nursery stage is essential for producing vigorous rice seedlings, and the right technological inputs can contribute to this (Safitri et al., 2018; Afa et al., 2018a; Afa et al., 2018b; Afa et al., 2021).

Many researchers have recently explored the use of biostimulants derived from liquid extracts of seaweed, such as *Kappaphycus* sp. to promote plant growth and production. Biostimulants in seaweed contain bioactive compounds, including growth-regulating hormones like gibberellins, cytokinins, auxins (IAA), abscisic acid, and many other bioactive substances (Trivedi et al., 2018; Noli et al., 2021; Vaghela et al., 2023).

These biostimulants can enhance crop resistance to stress, increase productivity, and improve yield quality when used in small quantities. At a concentration of 0.8%, most of these seaweed compounds are present at physiologically relevant levels, which significantly improve plant growth performance (Vaghela et al., 2023). Layek et al (2018) found that sap from *Kappaphycus alvarezii* at concentrations between 2.5% and 5.0% improved the vigor of rice seeds. Seaweed also contains essential macro- and micronutrients, including potassium (K), sodium (Na), magnesium (Mg), phosphate (P), iodine (I), and iron (Fe) (Cardoso et al., 2015; Kilowasid et al., 2023).

The application of biostimulants during the seedling phase can significantly boost rice productivity in a cost-effective, simple, and environmentally friendly way. Additionally, *K. alvarezii* is found throughout Indonesian waters (Aslan et al., 2021) and is one of the country's key fisheries commodities, as this seaweed has been widely cultivated (Cahyani et al., 2020; Rahim et al., 2019). Therefore, using seaweed could be a viable solution for meeting the biostimulant needs of food crops in Indonesia. This study aimed to evaluate the effect of liquid seaweed extract biostimulant concentration and sowing method on the seedling vigor of rice.

MATERIALS AND METHODS

Research setting and materials

This research was conducted at the experimental field of the Faculty of Agriculture, Halu Oleo University, Kambu Village, Kambu District, Kendari City, Southeast Sulawesi Province. The research is located at coordinates 04° 0' S and 122° 41' E, at an altitude of approximately 13.33 meters above sea level. The study was carried out from September to October 2024.

The materials used in this study included Inpari 32 rice seeds with a germination rate of 98%, sourced from the Plant Seed Inspection and Certification Center (BPSB TP) of Southeast Sulawesi. The liquid extract from *K. alvarezii* was obtained from approximately 35-day-old seaweed. The liquid extract was diluted with distilled water according to the treatment concentration. The seedling was maintained under a plastic house. The rice seed was germinated in plastic boxes (30 cm x 40 cm x 15 cm).

Experimental design and procedure

The research used a factorial treatment with two factors arranged in a randomized block design. The first factor was the seedling method (S), which had two levels: dry seedlings and wet seedlings. The second factor was the concentration of seaweed extract (K), which had six levels: no biostimulant (0%), 0.8%, 1.6%, 2.4%, 3.2%, and 4.0%. The treatment had 12 combinations. Each combination was repeated three times, resulting in a total of 36 experimental units.

The seedling medium consisted of topsoil plus husk charcoal 4:1 (w/w). The soil was dried, grounded through a 2 mm sieve, and then 5 kg of mixed media was placed in a plastic box. Two types of seedling media were prepared according to the treatment: dry seedling and wet seedling. Plastic box for dry seedling medium had holes at the bottom. The mixture was then filled with water and left for 24 hours. The moisture content at field capacity was then measured to determine the volume of water to be applied during maintenance. For the wet seedling, growing media was set to mimic the conditions of rice fields. This condition was maintained throughout the seedling stage.

For preparing seaweed extract, fresh seaweed was cleaned using seawater as needed and then chopped. The chopped seaweed was blended until smooth, and the resulting mixture was filtered through muslin cloth to produce the liquid sap. The liquid sap was stored in a plastic bottle and refrigerated at a temperature of about 5 °C until use. Before application, the liquid of seaweed extract was diluted with distilled water according to the specified concentration.

The volume and application schedule of the liquid were 3 mL per plant at 7 and 14 days after planting (DAP), and 5 mL per plant at 21 and 28 DAP. Premium rice seeds were soaked in, liquid extract for 18 hours before planting. The primed seeds were then aerated to restore their original moisture content.

Each plastic box contained 25 planting holes, with a single seed per planting hole. The planting distance was 6 cm x 3 cm. NPK Yaramila fertilizer was applied at planting, with a dosage of 5 g per plastic box.

Observations were made at 7, 14, 21, and 28 DAP. Observed variables included seedling height, leaf area, number of tillers, root dry weight, and seedling dry weight.

Data analysis

The observation data were analyzed using analysis of variance (ANOVA). If the ANOVA indicated a significant effect (F_calculated > F_critical), a mean difference test was conducted using Duncan's Multiple Range Test (DMRT) at a significance level of α = 0.05.

RESULTS AND DISCUSSION

Seedling height

The results of the independent effects of seedling method and biostimulant concentration on seedling height are presented in Table 1. The results for the interaction between seedling method and biostimulant concentration on seedling height at 21 DAP are shown in Table 2.

Table 1. Independent effects of seeding method and seaweed extract concentration on rice seedling height.

Treatments	Seedling height (cm)			
	7 DAP	14 DAP	21 DAP	28 DAP
Sowing method (S)				
Dry (S1)	15.05a	25.94	30.78b	41.31b
Wet (S2)	11.84b	25.10	35.30a	44.39a
Seaweed extract concentration (K)				
0 % (K0)	9.59b	22.92c	28.41b	38.27b
0.8 % (K1)	13.70a	25.62ab	34.68a	44.27a
1.6 % (K2)	13.90a	24.95b	34.32a	43.66a
2.4 % (K3)	14.83a	27.32a	33.36a	44.15a
3.2 % (K4)	14.59a	26.62ab	33.89a	44.32a
4.0 % (K5)	14.04a	20.64b	33.59a	42.44a

Note: Numbers followed by different letters in the same column are considered significantly different according to DMRT at α = 0.05.

Table 1 shows that the results of the independent effect test on the sowing method treatment indicated that the average seedling height of rice plants at 7 DAP was highest with the dry sowing method (S1), which was significantly different from the wet sowing method (S2). At 14 DAP, however, there was no significant difference in seedling height, although seedlings from the dry sowing method (S1) tended to be taller than those from the wet sowing method (S2). At 21 and 28 DAP, the wet seedling method (S2) resulted in the highest average seedling height, which was significantly different from the dry seedling method (S1). During the initial stages of seedling growth (germination phase), soil aeration and moisture conditions are crucial for sprout development. In dry seedlings, the nursery media maintains field capacity moisture, which creates favorable soil aeration and moisture conditions that support faster growth and emergence. Soil aeration promotes root growth and enhances the rate of photosynthesis and chlorophyll content, which in turn boosts plant growth (Li et al., 2019). As a result, rice seedlings are taller at 7-10 days after sowing when grown using the dry seedling method compared to the wet seedling method. However, after 14 days, wet seedlings show better height growth than dry seedlings. By 14 days after planting, soil aeration no longer poses a barrier to the growth of rice plants, allowing seedlings in the wet seedling method to grow taller at 21 and 28 DAP compared to those in the dry seedling method. Wet or saturated conditions are ideal for the nursery or seeding of paddy rice plants. Proper seeding ensures the production of healthy rice seedlings. In water-saturated soil, oxygen is still available for root development as it enters the plant through the stomata and travels down the stem to the roots. Plant roots need sufficient oxygen to produce ATP from sugar, which provides the energy required for maintaining health and supporting normal physiological activities (Li et al., 2019).

The results of DMRT of the independent effect of biostimulant concentration on the average height of rice seedlings at the age of 7 DAP were highest at a concentration of 2.4% (K3) which was not significantly different from the other treatments but significantly different from the treatment without 0% biostimulant treatment (K0). The effect of treatment at the age of 14 DAP seedlings was highest at a concentration of 1.6% (K2) which was not significantly different from the concentration treatment of 0.8% (K1) and 3.2% (K4), but significantly different from the concentration treatment of 1.6% (K2) and 4% (K5). At 21 DAP, the highest seedling height was observed at a 0.8% concentration (K1), while at 28 DAP, the highest seedling height was found at a 3.2% concentration (K4). These results were not significantly different among the treatments, except for the 0% biostimulant treatment (K0), which produced the lowest average seedling height. Biostimulants in seaweed plants are components of bioactive compounds that affect plant growth hormones in the form of gibberellins, cytokinins, auxins, brassinosteroids, and many other bioactive compounds (Noli et al., 2021; Vaghela et al., 2023). The application of *K. alvarezii* liquid extract up to a concentration of 3.2% increased the seedling height of rice plants. However, above this concentration, a decline was observed, although the effect was not consistent throughout the seedling growth phase, likely due to variations in environmental conditions during this period.

Table 2. Interaction effect of sowing method and biostimulant concentration on the seedling height of rice plants at 21 DAP.

Sowing method (S)	Seedling height (cm)						Average
	0 % (K0)	0.8 % (K1)	1.6 % (K2)	2.4 % (K3)	3.2 % (K4)	4.0 % (K5)	
Dry (S1)	28.03b p	30.22a q	31.52a q	31.35a q	31.16a q	32.38a q	30.78 q
Wet (S2)	28.79c p	39.13a p	37.12ab p	35.36b p	36.61b p	34.81b p	35.30 p
Average	28.41b	34.68a	34.32a	33.36a	33.89a	33.59a	

Note: Numbers followed by different letters in the same column (p, q) or in the same row (a, b, c) are considered significantly different according to DMRT at α = 0.05.

Applying seaweed extract can enhance the height growth of food crops (da Costa et al., 2017; Vasantharaja et al., 2019) and horticultural plants (Arioli et al., 2024). This growth promotion is believed to be due to the biostimulants present in seaweed extracts, which act as plant growth stimulants and accelerate metabolic processes in plant tissues (Trivedi et al., 2018; Thaimei et al., 2024). According to (Layek et al., 2018), sap from *K. alvarezii* at concentrations of 2.5% to 5.0% increased the vigor of rice seeds. Gibberellins play a key role in seed germination as well as stem and root elongation. They promote plant growth by stimulating the lengthening of internodes the segments between leaves by influencing the protein balance at the cellular level (Sasongko et al., 2020). The increase in plant height can be attributed to gibberellins triggering cell division and elongation.

Table 2 shows the interaction between seedling method and biostimulant concentration on the average height of rice seedlings at 21 DAP. In the dry seedling method (S1), the highest average seedling height was observed with a 1.6% biostimulant concentration (K2), which was not significantly different from the other treatments but significantly higher than the no biostimulant treatment (K0). In the wet seedling method (S2), the highest average seedling height was found with a 0.8% biostimulant concentration (K1), which was not significantly different from the 1.6% biostimulant concentration (S2K2) but was significantly higher than the other treatments. The combination of the wet seedling method with a 0.8% biostimulant concentration (S2K1) produced the highest average seedling height compared to the dry seedling method with either a 0.8% (S1K1) or 1.6% biostimulant concentration (S1K2). The lowest average seedling height was recorded in the dry seedling method with no biostimulant treatment (S1K0), which was not significantly different from the wet seedling method with no biostimulant (S2K0). Table 2 shows the DMRT results for the interaction between seedling method and biostimulant concentration on the average height of rice seedlings at 21 DAP. In the dry seedling method (S1), the highest average seedling height was observed with a 1.6% biostimulant concentration (K2), which was not significantly different from the other treatments but significantly higher than the no biostimulant treatment (K0). In the wet seedling method (S2), the highest average seedling height was found with a 0.8% biostimulant concentration (K1), which was not significantly different from the 1.6% biostimulant concentration (S2K2) but was significantly higher than the other treatments. The combination of the wet seedling method with a 0.8% biostimulant concentration (S2K1) produced the highest average seedling height compared to the dry seedling method with either a 0.8% (S1K1) or 1.6% biostimulant concentration (S1K2).

The lowest average seedling height was recorded in the dry seedling method with no biostimulant treatment (S1K0), which was not significantly different from the wet seedling method with no biostimulant (S2K0). Azeredo et al. (2017) stated that hormones affect plant growth at low concentrations. At a concentration of 0.8%, most of the seaweed compounds are present at physiologically relevant levels that enhance plant growth. The *K. alvarezii* seaweed biostimulant contains 27 mg L^{-1} of Indole-3-acetic acid (IAA), 20 mg L^{-1} of Zeatin, and 24 mg L^{-1} of Gibberellin (GA3). At a 0.8% concentration of the seaweed extract, it contains 0.216 mg L^{-1} of IAA, 0.16 mg L^{-1} of Zeatin, and 0.192 mg L^{-1} of Gibberellin (Layek et al., 2015). However, high concentrations of growth hormones can actually inhibit plant growth. Swandari and Faisal (2023) noted that gibberellins stimulate cell division and growth, leading to stem elongation and an increase in the number of plant internodes. Gibberellin application boosts auxin levels by promoting the production of proteolytic enzymes, which help soften plant cell walls (Jaenudin et al., 2021; Rasul et al., 2021; Swandari & Faisal, 2023). Fadholi & Koesriharti (2022), observed that the application of gibberellin hormones can increase the size of stem epidermal cells, plant height, stomatal density on the lower leaf surface, and total chlorophyll content.

Leaf area of rice seedlings

Table 3 shows that, for the independent effect of seedling method, the highest average leaf area of rice seedlings at 14 DAP was observed with the dry seedling method (S1). However, at 21 and 28 DAP, the highest leaf area was obtained with the wet seedling method (S2). The effect of seedling method on leaf area followed a similar pattern to other growth parameters, with rice seedlings grown using the wet seedling method showing a higher leaf area than those grown with the dry seedling method. For the independent effect of biostimulant concentration, the highest average leaf area at 14 and 21 DAP was observed at a concentration of 2.4% (K3), which was not significantly different from the other concentrations (0.8% (K1), 1.6% (K2), 3.2% (K4), and 4.0% (K5), but significantly higher than the no biostimulant treatment (K0). At 28 DAP, the highest leaf area was obtained at a concentration of 3.2% (K4), which was not significantly different from the other treatments (0.8% (K1), 1.6% (K2), 2.4% (K3), and 4.0% (K5), but significantly different from the no biostimulant treatment (K0). Higher biostimulant concentrations (4%) tended to reduce the leaf area of rice seedlings.

Table 3. Independent effects of sowing method and biostimulant concentration on the leaf area of rice seedlings.

Treatments	Leaf area (cm^2)		
	14 DAP	21 DAP	28 DAP
Sowing method (S)			
Dry (S1)	6.36a	9.34b	16.26a
Wet (S2)	5.66b	11.04a	17.11a
Seaweed extract concentration (K)			
0 % (K0)	4.95b	8.16b	13.09b
0.8 % (K1)	5.96a	10.80a	17.42a
1.6 % (K2)	6.07a	10.78a	17.26a
2.4 % (K3)	6.54a	10.84a	18.05a
3.2 % (K4)	6.49a	10.39a	18.09a
4.0 % (K5)	6.08a	10.18a	16.20a

Note: Numbers followed by different letters in the same column are considered significantly different according to DMRT at α = 0.05.

Seaweed extract, when applied at specific concentrations, can promote plant growth. The addition of auxin, one of the key components in seaweed liquid extract, stimulates plant growth by encouraging cell division and enlargement, which ultimately leads to increased leaf expansion (Jaenudin et al., 2021; Salsabila & Nihayati, 2024). Biostimulants at optimal concentrations can enhance plant growth and development, resulting in larger leaves. Growth hormones found in seaweed liquid extract, such as gibberellins, auxins, and cytokinins, work together to stimulate an increase in leaf area (both width and length). The physiological effects of these hormones cause stem elongation and promote the growth of flowers and leaves. These hormones regulate the growth rates of various tissues and coordinate their development to form a complete plant (Bakti et al., 2018; Jaenudin et al., 2021).

The tillers number of the rice seedlings

The DMRT results for the independent effect of the seeding method on the average number of tillers in rice seedlings at 28 DAP were highest with the dry sowing method (S1), which was significantly different from the wet sowing method (S2) (Table 4). This suggests that using the dry sowing method is more effective in promoting tiller formation in rice than the wet sowing method. The DMRT results for the independent effect of biostimulant concentration on the average number of tillers in rice seedlings at 21 and 28 DAP showed that concentrations of 0.8% (K1) and 4.0% (K5) resulted in the highest number of tillers, which were not significantly different from the other treatments but were significantly higher than the control group (K0), which had the lowest number of tillers. The effect of biostimulants on tiller formation was most effective at lower concentrations, with noticeable results starting at 21 DAP. A 0.8% concentration of seaweed liquid extract as a biostimulant can trigger biochemical, morphological, and physiological responses that enhance the growth process (Saban et al., 2018; Arnanto et al., 2024), leading to taller rice seedlings.

The interaction between seeding method and biostimulant concentration on the average number of tillers in rice seedlings at 28 DAP was shown in Table 5. In the dry sowing method (S1), the highest average number of tillers was observed in the 0.8% biostimulant concentration treatment (K1). This treatment was not significantly different from the other biostimulant concentrations, except for the no biostimulant treatment (K0), which showed a significant difference. In the wet sowing method (S2), the highest average number of tillers was found with a 4.0% biostimulant concentration (K4), which was not significantly different from the treatments with biostimulant concentrations of

2.4% (S2K3) and 3.2% (S2K4). However, it was significantly higher than the treatments with 0.8% (S2K1), 1.6% (S2K2), and no biostimulant (S2K0). The dry sowing method with the 0.8% biostimulant concentration (S1K1) resulted in the highest average number of tillers compared to the wet sowing method with a 4.0% biostimulant concentration (S2K5). Meanwhile, the lowest average number of tillers was found in the wet sowing method with no biostimulant (S2K0). The optimal biostimulant concentration for promoting tiller formation varies depending on the seeding method.

Table 4. Mean comparisons the independent effects of sowing method and biostimulant concentration on the number of tillers of rice seedlings.

Treatments	Number of tillers		
	14 DAP	21 DAP	28 DAP
Sowing method (S)			
Dry (S1)	1.08	2.60	4.11a
Wet (S2)	1.06	2.60	3.61b
Seaweed extract concentration (K)			
0 % (K0)	0.73	2.07b	2.63b
0.8 % (K1)	1.73	2.83a	4.07a
1.6 % (K2)	1.03	2.63a	3.90a
2.4 % (K3)	1.23	2.73a	4.20a
3.2 % (K4)	1.00	2.57a	4.13a
4.0 % (K5)	1.03	2.67a	4.23a

Note: Numbers followed by different letters in the same column are considered significantly different according to DMRT at α = 0.05.

Table 5. Interaction effect of sowing method and biostimulant concentration on the number of tillers of rice seedlings at 28 DAP.

Sowing method (S)	Number of tillers						
	0 % (K0)	0.8 % (K1)	1.6 % (K2)	2.4 % (K3)	3.2 % (K4)	4.0 % (K5)	Average
Dry (S1)	2.73c p	4.73a p	4.28a p	4.47a p	4.20a p	4.27a p	4.11 p
Wet (S2)	2.53d p	3.40c q	3.53bc q	3.93ab q	4.07a p	4.20a p	3.61 q
Average	2.63b	4.07a	3.90a	4.20a	4.13a	4.23a	

Note: Numbers followed by different letters in the same column (p, q) or in the same row (a, b, c) are considered significantly different according to DMRT at α = 0.05.

The dry sowing method requires a lower concentration of biostimulants compared to the wet sowing method for producing tillers. The higher concentration used in the wet sowing method may be due to the water in the planting medium, which dilutes the biostimulant. When biostimulants are sprayed onto the leaves, they are not directly absorbed by the plant but instead fall onto the surrounding planting medium near the plant roots. This can reduce the concentration of the biostimulant, which is why the highest number of tillers in the wet sowing method is achieved at a higher biostimulant concentration (4.0%). Hormones play a crucial role in the growth and development of rice plants. Auxin, a component of the biostimulant seaweed extract, can increase the number of tillers (Jin et al., 2016). Auxin accumulates in the tiller buds and stem bases (Yang et al., 2017). Hormones regulate the growth rate of different tissues and coordinate these parts to form the overall structure of the plant. These non-nutritive organic compounds are active at low concentrations and can induce biochemical, morphological, and physiological responses (Arnanto et al., 2024).

Root dry weight

The DMRT results for the independent effect of seedling method on the average dry weight of rice seedling roots at 28 DAP showed that the wet seedling method (S2) produced the highest values, significantly differing from the dry seedling method (S1)

(Table 6). In contrast, the DMRT results for the independent effect of biostimulant concentration on the average dry weight of rice seedling roots at 28 DAP were highest at a concentration of 3.2% (K4). This was not significantly different from the treatments at 0.8% (K1), 1.6% (K2), 2.4% (K3), and 4.0% (K5), but was significantly different from the treatment with no biostimulant (K0). The application of liquid seaweed extract biostimulant at a concentration of 3.2% led to a better response in increasing the dry weight of rice seedling roots. Biostimulants, such as auxin, play a role in promoting the formation of root hairs and lateral shoots (Ramadhani & Karyawati, 2024). Root hairs and lateral roots are essential for optimizing the absorption of water and minerals in plants (Salsabila & Nihayati, 2024). Growth hormones, either individually or in combination with other hormones, can stimulate root formation through mechanisms that regulate the balance of organic and inorganic substances in plant tissues (Zinabu et al., 2021). Auxin is a naturally occurring hormone in plants that is responsible for cell elongation and enlargement, which accelerates root development (Bakti et al., 2018; Hu et al., 2021). This leads to an increase in the dry weight of rice seedling roots. Auxin is a key regulator of many important plant traits, including root architecture, tillering, flowering, seed quality, and stress responses (Wang et al., 2018).

Table 6. Independent effect of sowing method and biostimulant concentration on the dry weight (g) of root rice seedlings at 28 DAP.

Sowing method (S)					
Dry (S1)			Wet (S2)		
0.25b			0.39a		
Seaweed extract concentration (K)					
0% (K0)	0.8% (K1)	1.6% (K2)	2.4% (K3)	3.2% (K4)	4.0% (K5)
0.13b	0.32a	0.36a	0.37a	0.38a	0.35a

Note: Numbers followed by different letters in the same row (a, b) are significantly different according to DMRT at α = 0.05

Total biomass dry weight

Table 7 shows that the independent effect of the sowing method on the average dry weight of rice seedlings at 28 DAP was highest with the wet sowing method (S2), which was significantly different from the dry sowing method (S1). Meanwhile, the independent effect of biostimulant concentration on the average dry weight of seedlings at 28 DAP was highest at a 1.6% concentration (K2), which was not significantly different from the 0.8% (K1), 2.4% (K3), 3.2% (K4), and 4.0% (K5) concentrations, but was significantly different from the no biostimulant treatment (K0).

Table 7. Independent and interaction effect of sowing method and biostimulant concentration on the total biomass dry weight of rice seedlings at 28 DAP.

Sowing method (S)	Total biomass dry weight (g)						Average
	0 % (K0)	0.8% (K1)	1.6% (K2)	2.4% (K3)	3.2% (K4)	4.0% (K5)	
Dry (S1)	1.10b p	1.88a q	1.99a q	2.02a q	2.06a q	2.04a q	1.85q
Wet (S2)	1.19b p	2.95a p	2.94a p	2.67a p	2.70a p	2.77a p	2.54p
Average	1.14b	2.42a	2.47a	2.35a	2.38a	2.41a	

Note: Numbers followed by different letters in the same column (p, q) or row (a, b, c) are significantly different according to DMRT at α = 0.05.

Table 7 presents the wet sowing method with a 0.8% biostimulant concentration (S2K1) produced the highest average dry weight of rice seedlings compared to the dry sowing method with a 3.2% biostimulant concentration (S1K4). Meanwhile, the lowest average dry weight of rice seedlings was found in the dry sowing method with no biostimulant treatment (S1K0). Optimal growth and development of rice seedlings occur in a suitable growing environment, leading to enhanced seedling growth. Leaf formation, leaf area growth, and tiller development will occur optimally in a suitable growing environment with adequate biostimulant and nutritional support (Tables 3, 4, 5, 6, and 7).

Biostimulants, including the hormones found in seaweed liquid extract, stimulate the growth of plant organs such as roots, stems, and leaves, as well as the formation of seedlings. Gibberellin promotes shoot growth, increases cell number and size, and maximizes photosynthesis, leading to improved vegetative growth (Swandari & Faisal, 2023; Ramadhani & Karyawati, 2024). Number of leaves and leaf area contribute to an increase in plant weight (Sianipar et al., 2020). Growth regulation occurs through the production of specific hormones, which influence hormone synthesis, disrupt translocation, or change the location of hormone formation (Mahendra et al., 2017). Auxin is directly involved in the biosynthesis of materials in plants, enabling the production of more plant cells, which results in increased dry matter and reserves in the seeds (Wang et al., 2018; Hu et al., 2021; Sosnowski et al., 2023). However, excessive concentrations of hormones or growth regulators can disrupt cell functions, leading to stunted plant growth. At higher levels, the hormone may inhibit growth, damage plants, or even cause death (Zinabu et al., 2021; Irdika & Nugraha, 2022). Optimal biostimulant application is achieved by using the correct concentration and frequency, which stimulates hormone activity and promotes the development of root, stem, and leaf organs (Saban et al., 2018; Dalero et al., 2019; Sari et al., 2019; Noli et al., 2021).

The *K. alvarezii* seaweed extract contains bioactive compounds, as well as macro and micronutrients essential for plants, including potassium (K), sodium (Na), magnesium (Mg), phosphate (P), iodine (I), and iron (Fe) (Cardoso et al., 2015; Layek et al., 2015; Kilowasid et al., 2023). It also contains plant growth regulators such as IAA, gibberellin, cytokinin, choline chloride, and glycine betaine, which play a role in triggering various physiological responses in plants (Layek et al., 2015). Choline and glycine in the seaweed sap can prevent negative interactions between GA_3 and other hormones (Mondal et al., 2015). The seaweed extract enhances antioxidant properties and improves nutrient absorption from the soil. Additionally, applying the seaweed sap promotes root system development, as it contains endogenous auxins and other compounds, ultimately strengthening nutrient uptake by the roots (Pramanick et al., 2017).

To assess the effectiveness of *K. alvarezii* seaweed as a biostimulant for enhancing seedling vigor, growth, and production, further studies should be conducted. These studies should focus on application volume and frequency during different growth stages of rice plants, in both dry land, irrigated, and rainfed rice field conditions.

CONCLUSIONS

The interaction between seedling method and seaweed extract concentration increased the number of tillers, and dry weight of rice seedlings. The wet seedling method, starting 14 days after planting, was more effective than the dry seedling method in promoting seedling vigor in the nursery. Biostimulant concentrations ranging from 0.8% to 2.4% produced the highest seedling vigor. The wet sowing method combined with a 0.8% liquid seaweed extract biostimulant (S2K1) resulted in the highest seedling vigor, as indicated by seedling dry weight.

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