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Effectiveness of Seed Priming on Vigor and Viability of Moringa Seeds (*Moringa oleifera* L.)

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Abstract

Moringa is often referred to as the "miracle tree" due to its rich nutrient content and medicinal properties. The increasing demand for moringa seeds has highlighted the need for effective propagation methods; however, germination remains limited by the hard seed coats and prolonged dormancy periods. This gap underscores the importance of studying moringa, particularly in efforts to enhance germination success and initial growth. This study aims to evaluate the effects of various seed priming materials, soaking durations, and the interactions between different types of seed priming materials and soaking times on the vigor and viability of moringa seeds. The research was conducted in the Biotechnology Laboratory and greenhouse at the Faculty of Agriculture, National Development University "Veteran" East Java. This study employed a Completely Randomized Design (CRD) with a factorial arrangement of two factors, comprising fifteen combinations and three replications. The first factor is the type of seed priming material (S), which includes Aqua Destillata, seaweed extract, coconut water, PEG 6000, and KNO3. The second factor is the soaking time for the priming solution (L), which consists of 12 hours, 24 hours, and 36 hours. Observations include germinability, maximum growth potential, growth rate, vigor index, and sprout weight. The combination treatment of PEG 6000 as the seed priming material with a soaking time of 12 hours yielded the highest average values for germination and maximum growth potential. A single treatment using PEG 6000 resulted in the highest average vigor index. Additionally, a single treatment with a soaking duration of 12 hours also produced the highest average vigor index. The results of this study can be recommended as optimal treatments to enhance the germination capacity and vigor of Moringa seeds.

Keywords: Germination Power, Growth Index, PEG 6000, Soaking Treatment

1. Introduction

Indonesia boasts a rich biodiversity, making it a valuable asset for global sustainability. Indonesia boasts a diverse range of plants with the potential to offer substantial benefits to human life. One notable example is the moringa plant. The growing demand for moringa leaves has also led to a significant increase in the demand for moringa plant seeds. From January to September 2024, Indonesia experienced a substantial increase in moringa leaf exports, with a total volume reaching 4,350 tons and an export value of $ 13.75 million (BPS, 2024). Therefore, it is essential to employ the most effective propagation method to meet the growing demand for moringa exports.

The propagation of moringa plants on a large scale typically occurs through generative methods. A notable benefit of generative propagation is its capacity to yield plants that exhibit enhanced fruit production and robust root development, resulting in an extended productive lifespan. However, the process of generating new moringa plants through vegetative propagation is challenging due to the low rate of seed germination and survival. Moringa plants possess hard and thick seed coats that impede the ingress of water and oxygen into the seeds, thereby prolonging the dormancy period (Paramita et al., 2018). The hard and thick skin of moringa seeds can also impede the nursery process, as it necessitates a significant amount of energy and time if the seed coat is manually peeled. As Muniarty and Zuhri (2002) explained in Rosalyne et al. (2021), this phenomenon is also observed in robusta coffee beans. In this study, the authors found that peeling 100% of the robusta coffee beans (i.e., completely removing the skin from the beans) can accelerate germination time from 41

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days after sowing (without peeling) to 27 days after sowing.

The problem of seed dormancy can be overcome by breaking seed dormancy treatment, allowing for efficient germination of moringa plant seeds. Invigoration is a physical or chemical treatment carried out to increase and repair seed vigor that has declined, specifically by conditioning the seeds in a manner that enhances their physiological and biochemical characteristics. Priming is one of the invigoration techniques carried out using imbibition media that has a low water potential pressure, allowing the imbibition process to occur in a controlled manner and enabling the metabolic process in the seeds to take place properly.

Priming methods can be in the form of chemical or organic treatments. Priming with chemicals can be achieved by providing a salt solution with a low osmotic potential, such as Polyethylene glycol (PEG) 6000, KNO_3, K_3PO_4, $MgSO_4$, glycerol, and mannitol to accelerate the seed germination process (Saputra et al. 2017) Research on the use of chemical priming, one of which is the use of KNO_3 conducted by (Nengsih and Hartawan 2017), showed the best treatment in testing the dormancy breaking of Liberica coffee seeds, namely soaking in a KNO_3 solution with a concentration of 0.5% and a soaking time of 24 hours. This treatment gave a germination percentage of 58.33%. At the same time, organic *priming* can be in the form of young coconut water, seaweed extract, tomato extract, and banana extract. Organic extracts contain carbohydrates and amino acids that can increase seed germination. Priming with the use of organic materials, one of which is research conducted by Lewu et al. (2023), The concentration of seaweed extract solution (*Sargassum polycystum*) 60% gave the best seed viability in sorghum seeds with viability values consecutively consisting of germination power of (98.45%), growth simultaneity of (98.33%), and vigor index reaching (98.50%). The description above indicates that the provision of seed priming material can enhance seed germination; therefore, it is necessary to conduct experiments to determine the type of seed priming material and the optimal soaking time that can increase the viability and vigor of Moringa seeds (*Moringa oleifera* L.).

2. Material and Methods

2.1. Time and Place

This research was conducted from January to March 2025 at the Laboratory of Biotechnology and Greenhouse, Faculty of Agriculture, National Development University "Veteran" East Java. The research location is situated at coordinates 7º20'1.97" S and 112º47'28.21" E, with an altitude of 6 m above sea level. The average temperature ranges from 25ºC to 33ºC, and the average humidity is 90%.

2.2. Tools and materials

The tools used in this study included a 1000 mL beaker glass, a 1000 mL Erlenmeyer flask, a stirrer, a hand sprayer, a ruler, an analytical balance, a 50 cm x 50 cm germination tank, an oven, label paper, and a *polybag. While the materials used in the study were moringa plant seeds* originating from PT. Moringa Organik Indonesia, water, seaweed extract priming solution, coconut water, PEG 6000, KNO3, fine sand, garden soil, rice husk charcoal, and compost.

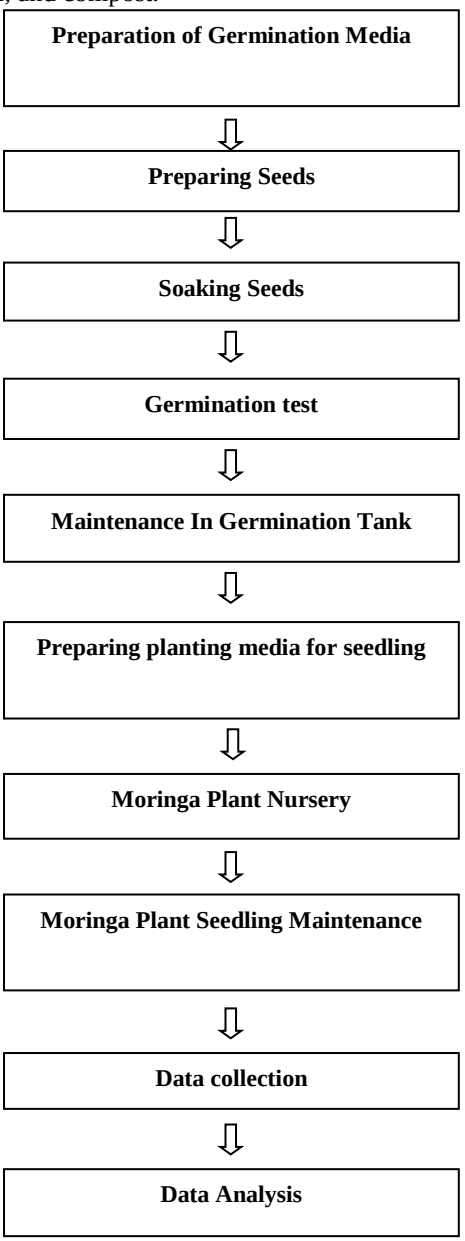

Figure 1. Research flow diagram

2.3. Research methods

This study is a factorial experiment arranged according to a Completely Randomized Design (CRD) with two factors. The first factor is the type of seed priming material (S), consisting of distilled water, seaweed extract, coconut

water, PEG 6000, and KNO3. The second factor is the duration of soaking the priming solution (L), consisting of 12 hours, 24 hours, and 36 hours. Each treatment consists of 100 seeds. Each treatment consists of three replications. The parameters observed were:

a) Increased power (%)

Germination power refers to the number of normal sprouts produced by pure seeds under specific environmental conditions within a specified period. Germination power observations were carried out on the last observation day (final count), namely on the 14th day. Calculation of germination power using the following formula

$$\text{DB} = \frac{total\ ogf\ normal\ germination}{Total\ of\ planted\ seeds} \times 100\%$$

b) Growth Potential (%)

Observations of maximum growth potential were conducted on the last observation day, specifically on seedlings aged 14 days after sowing (HSS). They were calculated based on the percentage of seeds that had grown. The maximum growth potential is calculated using the following formula (Yuliani et al., 2023):

$$\text{PTM} = \frac{Total\ of\ frowing\ seeds}{Total\ of\ germinated\ seeds} \times 100\%$$

c) Additional Growth Rate (hss)

The germination rate can be measured by calculating the number of days required for the emergence of the radicle or seed plumule from the beginning of germination to the final day of observation, namely 14 Days After Sowing (DSS). According to (Lesilolo et al. 2018), the method for calculating the germination rate is as follows:

$$\text{Germination Rate} = \frac{N1T1+N2T2+N3T3+\cdots.+NXTX}{total\ number\ of\ germinated\ seeds}$$

Information:

N = Number of Seeds that Germinate Each Day

T = The amount of time from the start of testing to the end of a specific interval of observation

d) Vigor Index (hss)

Testing is carried out by observing the number of normal shoots that appear each day, starting from the first observation day to the last day of observation. Vigor Index (IV) can be calculated using the following formula (Yuliani et al. 2023) :

$$\text{IV} = \frac{N1}{D1} + \frac{N2}{D2} + \frac{N3}{D3} + \ldots\ldots + \frac{Gn}{Dn}$$

Information :

N = The number of seeds that germinate is within normal range.

D = The time corresponding to that amount

n = Number of days on last calculation

e) Root Length K ec add (grams)

The measurement of the sprout roots' length was carried out by dismantling the sampled plants. The plant roots were washed clean by spraying water until the remaining sand was gone, then dried. The measurement of the length of the sprout roots was carried out on the last day of germination observation, namely the 14th day of HSS. The calculation of the root length was carried out manually using a ruler as a tool.

3. Results and Discussion

3.1. Germination Power (%)

Germination power observation was carried out on the last observation day (final count), namely on the 14th day. The results of the analysis of variance (ANOVA) showed that there was a very significant interaction effect on the combination of seed priming material and soaking time treatments on germination rate (Table 1). This observation of germination power showed that the treatment of seed priming material P EG 6000 and a safe soaking time of 12 hours produced the highest average germination capacity, namely 72%, but there was no significant difference with other treatments, namely treatment with seed priming material P EG 6000 and a safe soaking time of 24 hours and 3 6 hours, treatment with coconut water seed priming material and a safe soaking time of 12 hours, treatment of aqua d es seed priming material and a safe soaking time of 24 hours and 3 6 hours. Treatment of seaweed extract seed priming material with soaking periods of 12 hours, 24 hours, and 36 hours resulted in the lowest average germination capacity, namely 0% and 0.67%.

PEG 6000 treatment reduces the osmotic potential of the solution, thereby inhibiting water absorption in phase I and delaying seed swelling and initial metabolic activation (Wang et al., 2023). In Phase II, the duration can be extended, allowing seeds that have decreased in quality to improve their metabolism before Phase III (Ruliansyah, 2011, as cited. According to Oktafianus and Kannapadang (2025), the physiological germination process involves several key stages, including water absorption, metabolism of food reserves, breakdown of the endosperm, transport of breakdown products to actively growing embryos, reformation of new materials, respiration, and growth. PEG at low to moderate concentrations can increase radicle vigor and growth, especially under environmental stress, but high concentrations reduce germination due to excessive osmotic stress that interferes with the transition to phase III (Farooq et al., 2019)

The seed germination process is also influenced by the duration of seed soaking, where the longer the seed soaking time, the greater the potential for water to penetrate the seed's endosperm. Soaking seeds in water can soften the seed coat, allowing it to break and tear, which enables embryo and endosperm development to occur more quickly. This process also provides access for oxygen to enter the seed. Research conducted by Winarni et al. (2018) showed that seed priming could increase the results, where water soaking for 24 hours on jernang rattan seeds

(Daemonorops draco) produced 80% germination power. The results of this study indicate that treating seed priming materials with the correct soaking time can activate metabolism and accelerate germination.

Table 1. The Effect of Combination of Seed Priming Materials and Soaking Time on the Germination Power of Moringa Seeds

Soaking Time	Germination Power (%)				
	Seed priming materials				
	Aqua dest	Seaweed Extract	Coconut water	PE G 6000	KNO3
12 hours	55.33 ± 4.37 d ef	0 ± 0 a	57.33 ± 1.76 defg	72 ± 4.16 g	54.67 ± 1.76 cdf
24 hours	58 ± 2 defg	0 ± 0 a	39.33 ± 1.761 c	64.67 ± 4.67 fg	48.67 ± 3.71 cd e
36 hours	60 ± 4.16 efg	0.67 ± 0.67 ab	16 ± 2.3 b	60 ± 4 efg	42.67 ± 2.9 cd
BNJ 5%			15.5		

Description: Numbers followed by the same letter indicate no significant difference in the 5% BNJ test.

Figure 2. Visual of Moringa Sprouts from Seed Priming. S0: Aquades; S1: Seaweed Extract; S2: Coconut Water, S3: PEG 6000, S4: KNO3, L1: 12 Hours, L2: 24 Hours, L3: 36 Hours, a: Normal Sprouts, b: Abnormal Sprouts.

3.2. Maximum Growth Potential (%)

Observation of maximum growth potential was conducted by counting the number of sprouts that grew normally or abnormally on the last observation day, specifically those aged 14 HSS. The results of the analysis of variance (ANOVA) showed a very significant interaction effect between the combination of materials (seed priming and soaking time) and the Maximum Growth Potential of Moringa seeds (Table 2).

Observing the maximum growth potential shows that treating P EG 6000 seed priming material with a soaking time of 12 hours produces the highest average maximum growth potential, namely 84%, but the treatment is not significantly different. with other treatments, namely the treatment of seed priming material P EG 6000 and a safe soaking time of 24 hours and 3 6 hours, treatment of seed priming material KNO 3 and safe duration of 12 hours and 24 hours, treatment of seed priming ingredients with coconut water and safe duration of 12 hours, treatment of seed priming ingredients aqua d es and soaking time of 12 hours and 24 hours. The treatment of seaweed extract seed priming material with soaking times of 12 hours, 24 hours, and 36 hours produced the lowest average maximum growth potential values of 6%, 1.33%, and 0.67%, respectively. The treatment of PEG 6000 seed priming material was able to provide the best value, following the opinion of Ahmadvand et al. (2012) in Trisnawaty et al. (2024), that priming Using PEG can affect several processes in seeds, such as improving metabolism, improving seed deterioration, accelerating germination time, increasing germination rate and potential. Seeds can passively absorb water to trigger active metabolic processes.

Research conducted by Hernández-Herrera et al. (2023), stated that horridum seaweed extract plays a role as one of the factors that inhibits the germination process of tomato plant seeds, where the Sargassum extract horridum inhibits tomato seed germination (34%). The presence of sterol or saponin content in seaweed extract may cause the inhibition of the germination process. Research conducted by Sun et al. (2021) found that compounds such as fucosterol, 24-hydroperoxy-24-vinylcholesterol, and saringosterol, identified in Sargassum fusiforme, showed allelopathic activity. According to Tanjung et al. (2023), as cited, allelopathy is a phenomenon where a chemical agent is released into the environment with the aim of affecting other organisms in its vicinity. Allelopathic substances are harmful and can act as inhibitors in the growth process of a plant. The allelopathic effect is selective, meaning it only affects certain types of organisms. This is evidenced by research conducted by Nugroho et al. (2022), which shows that the allelopathy of the gamal plant type, with a concentration of 46%, resulted in the lowest percentage of green bean germination, specifically 33.33%.

Table 2. Effect of Combination of Seed Priming Materials and Soaking Time on the Maximum Growth Potential of Moringa Seeds

Maximum Growth Potential (%)					
Soaking Time (L)	***Seed priming* materials (S)**				
	Aqua dest	**Seaweed Extract**	**Coconut water**	**PE G 6000**	**KNO3**
12 hours	75.33± 6.35 de	6 ± 2.3 a	73.33 ±2.9 de	84 ±1.15 e	77.33± 2.9 de
24 hours	76 ± 4.61 de	1.33± 0.67 a	61.33 ±2.9 cd	70± 4.61 de	77.33± 0.67 de
36 hours	62 ± 4 cd	0.67± 0.67 a	34.67± 2.9 b	66.67± 5.81 cde	50 ± 3.05 bc
BNJ 5%	18.24				

Description: Numbers followed by the same letter indicate no significant difference in the 5% BNJ test.

3.3. Germination Rate (hss)

The germination rate can be measured by calculating the number of days required for the emergence of the radicle or seed plumule from the start of germination to the final day of observation, namely 14 days after sowing. The results of the analysis of variance (ANOVA) showed a significant interaction effect between the combination of seed priming materials and soaking time on the rate of seed germination for Moringa (Table 3).

The results of observations on germination rates showed that treating seed material with coconut water and soaking it for 24 hours resulted in the highest average germination rate, namely 9.37 days. However, this treatment was not significantly different from any other treatment combination and was significantly different from the other treatments. Seed priming ingredients include seaweed extract and a safe drying time of 36 hours. Organic treatments respond more quickly because they directly support the initial metabolic activation of the seed. PEG 6000 and KNO_3 can produce the best vigor and germination power because both repair cell membranes, gradually activate seed metabolism, and increase energy reserves, thereby supporting more optimal seed growth in the long term.

According to Davidz (2023), as cited in the source, the hormones contained in coconut water, serving as growth regulators, include cytokinins (5.8 mg/L), auxins (0.07 mg/L), and gibberellins. Auxin plays a role in increasing the distribution and absorption of water in plants, as well as enhancing photosynthesis activity, thereby facilitating the process of plant tissue development. Additionally, the auxin hormone influences the synthesis of structural proteins, allowing the cell wall to regain its normal structure after being stretched (Saputri et al., 2022). Natural growth regulators can enhance seed embryo development and function as triggers for the germination process. There

are various factors that influence the potential for seeds to grow optimally, one of which is the use of coconut water as a natural growth regulator, which can accelerate the germination process. Based on research conducted by Devitriano and Syarifuddin (2021), it was shown that the treatment of 15% coconut water concentration with a soaking time of 24 hours showed the best results on the germination power, vigor, and dry weight of Moringa plants (Moringa oleifera) with consecutive values, namely germination power 52%; vigor 53.33%; and dry weight 0.26 grams.

Table 3. Effect of Seed Priming Material Combination and Soaking Time on Moringa Seed Germination Rate

Soaking Time	Germination Rate (days)				
	Seed Priming Material				
	Aqua dest	Seaweed Extract	Coconut water	PE G 6000	KNO3
12 hours	6.82 ± 0.150 ab	9.30± 1.153 b	7.44± 0.061 ab	7.03 ±0.384 ab	6.88 ±0.283 ab
24 hours	6.91± 0.356 ab	6.33± 3.28 ab	9.37± 0.037 b	6.47 ±0.110 ab	6.99± 0.298 ab
36 hours	6.44± 0.155 ab	2.33± 2.333 a	8.25 ±0.41 b	6.74± 0.280 ab	6.29± 0.086 ab
BNJ 5%			5.75		

Description: Numbers followed by the same letter indicate no significant difference in the 5% BNJ test.

3.4. Seed Vigor Index (hss)

The vigor index is related to normal sprouts with good shoot growth and roots that are able to absorb nutrients effectively. This Vigor Index test is carried out by observing the number of normal sprouts that appear each day from the first observation day to the last day of sprout observation. The results of the analysis of variance (ANOVA) showed that there was no interaction between the combination of seed priming materials and soaking time on the vigor index. Moringa seeds. Single treatment of seed priming material and soaking time had a very significant effect on the seed vigor index (Table 4).

The results of observing the vigor index showed that a single treatment of the seed priming material PEG 6000 and a single long-term treatment of the safe treatment had a significant influence on the seed vigor index of the lor. A single treatment of the seed priming material P E G 6000 m produced the highest average vigor index, namely 5.41. Still, there was no significant difference between a single treatment of the seed priming material, aqua dest, and KNO3. There was no significant difference between a single treatment of the seed priming material, seaweed extract, and coconut water. A single treatment of seaweed extract yielded the lowest average seed vigor index value, namely 0.04. This finding aligns with research conducted by Afdharani et al. (2020), which showed that the use of PEG 6000 seed priming material yielded higher results compared to other seed priming materials. others. This shows that PEG can inhibit leakage in expired rice seeds by binding water and forming a thin layer that functions to reduce leakage during the imbibition process.

A single treatment with a duration of 12 hours of soaking resulted in the highest average vigor index, namely 3.87, which was not significantly different from a treatment with a duration of 24 hours and was significantly different from a single treatment with a duration of 36 hours. Safe treatment treatment of 3 6 hours resulted in the lowest average vigor index, namely 2.94. Similar results were also found in research by Zahra and Isda (2023), where soaking durations of 36 hours and 48 hours resulted in the lowest germination rates, specifically 56.6%. This result aligns with the statement by Sativa et al. (2022), who noted that soaking for too long can cause anoxia, or a loss of oxygen, in the seeds, thereby limiting the respiration process and affecting germination. Longer soaking can also increase the absorption of water, dissolved oxygen, and additional substances together during the imbibition process to a certain limit.

Table 4. Single Effect of Seed Priming Material and Soaking Time on Moringa Seed Vigor Index

Treatment	In Vigor dex
***Seed Priming* Material**	
Aqua dest	5.28 ± 0.326 c
Seaweed Extract	0.043 ± 0.059 a
Coconut water	1.77 ± 1.479 b
PE G 6000	5.41 ± 0.488 c
KNO3	5.03 ± 1.381 c
BNJ 5%	0.86
Soaking Time	
12 hours	3.87 ± 2.992 b
24 hours	3.71 ± 3.023 b
36 hours	2.94 ± 2.949 a
BNJ 5%	0.50

Note: Numbers followed by the same letter in the same treatment indicate no significant difference in the 5% BNJ test

3.5. Length of Sprout Root (cm)

Measurements were taken from the base of the stem to the tip of the root at the end of observations during the germination period, namely on the 14th day HSS. The

results of the analysis of variance (Anova) showed that there was no interaction between the treatment of seed priming materials and the soaking time on the length of the roots of the Moringa plant seedlings. The single treatment of seed priming materials produced a very significant effect, and the soaking time did not have a significant effect on the length of the roots of the Moringa seedlings (Table 5).

The length of the sprout roots showed that the single treatment of PEG 6000 seed priming material gave a very significant effect on the length of the moringa plant's roots to sprouts. The single treatment of PEG 6000 seed priming material produced the highest average length of roots to sprouts, which was 5.34 cm. However, this difference was not significant compared to the single treatments of other seed priming materials, and it was significantly different from the single treatment of seaweed extract seed priming. The single treatment of seaweed extract seed priming produced the lowest average length of roots to sprouts, which was 0.41 cm. The single treatment of soaking time had no significant effect on the length of roots to sprouts.

The single treatment of PEG 6000 seed priming material produced the highest average root length to the sprout in line with the statement (Arthawijaya et al., 2022) which states that soaking treatment using Polyethylene Glycol (PEG) with the right combination of concentration and soaking duration can optimize the water imbibition process into the seeds. The results of Sari's research (2020) showed that the soaking treatment of rice seeds that had experienced decline using PEG 6000 with a concentration of 5% and a soaking duration of 24 hours showed the highest average viability value at 86% germination power, 80% vigor index, seed water content 23.8%, seed growth simultaneity 84.67% and seed growth rate 18.27%/etmal. Good imbibition will trigger the acceleration of enzyme activity and metabolism that support seed growth, thereby accelerating the development of the radicle due to the fulfillment of water needs and softening of the seed coat. On the other hand, if the concentration and soaking time are not appropriate, water absorption during imbibition becomes limited. This condition will inhibit the activity of enzymes and metabolism that play a role in root growth. In addition, according to , seed metabolic activity will run optimally when the imbibition process is complete. This active metabolism facilitates the seeds' continued growth during the root formation phase. The formation of these roots is significantly influenced by the seeds' ability to adapt morphologically and physiologically. However, excess water can inhibit the continuation of the root formation process, as the seeds are unable to regulate their water needs optimally.

Table 5. The Single Effect of Seed Priming Material and Soaking Time on the Root Length of Moringa Seed Sprouts

Treatment	**Length of sprout roots (cm)**
***Seed Priming* Material**	
Aqua dest	4.53 ± 0.399 b
Seaweed Extract	0.41 ± 0.072 a
Coconut water	4.99 ± 1.811 b
PE G 6000	5.43 ± 0.598 b
KNO3	4.35 ± 1.6917 b
BNJ 5%	1.34
Soaking Time	
12 hours	3.97 ± 3.346
24 hours	3.52 ± 3.379
36 hours	4.33 ± 3.29
BNJ 5%	tn

Description: Numbers followed by the same letter in the same treatment show no significant difference in the 5% BNJ follow-up test; tn = no significant difference.

4. Conclusion

The results indicated that the combination treatment of PEG 6000 seed priming material and a 12-hour soaking time yielded the most favorable outcomes in terms of germination power and maximum growth potential. In contrast, the single treatment of PEG 6000 seed priming material produced the best results for the vigor index and root length at sprouting. Additionally, the single treatment of a 12-hour soaking time resulted in the highest seed vigor index. Key findings from this study suggest that seed priming with PEG 6000 for 12 hours is the optimal method for enhancing the physiological quality of moringa seeds.

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