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Research Article

Examination of Shallot Seedling Quality Through Seed Priming

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Abstract: The study aimed to examine the effect of the seed priming method with zinc micronutrients on the growth of seedlings of two shallot varieties from aging botanical seeds. The research was conducted in the Green House, Agriculture Faculty, Hasanuddin University, Makassar, Indonesia. The study was arranged in a split-plot design (SPD) with a randomized complete block design (RCBD). The main plot was a type of variety, which consisted of 2 combinations, namely Lokananta and Maserati. The subplot was a type of priming comprising six treatments: unpriming, hydropriming, IAA priming, ZnO priming, $ZnSO_4·7H_2O$ priming, and Zn-EDTA priming. Zn-EDTA priming increased germination percentage (71.43%), root length (13.07 cm), and number of root tips (12.04). $ZnSO_4.7H_2O$ priming produced the highest seedling height (28.72 cm). Then, ZnO priming increased the number of leaves (4.18 leaves), pseudo-stem diameter (3.36 mm), fresh weight (2.54 g), dry weight (0.22 g), and seedling quality index (0.0081). Seed priming using Zn improved the growth quality of aging shallot seedlings.

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1. Introduction

Shallot (*Allium ascalonicum* L.) is an important food crop in many parts of the world. Shallot production can be done by using bulbs and seeds as planting material. Seed quality, especially germination and seed vigor (the potential for rapid, uniform emergence and development of normal seedlings under a wide range of field conditions), plays a vital role in producing physically uniform shallot seedlings and growing time in various environmental conditions. Good seed quality will affect growth and yield after planting. Mohammed et al. (2018) reported that using seeds can produce consumption bulbs up to 45.14 t ha^{-1} and true seeds with 98% germination ability with an average growing time of 4 days after sowing.

Shallot planting material could be of better quality. The main reason that causes seeds to have poor quality is faster damage when the storage process is not by the criteria required by the seeds. Seed moisture content and storage temperature are the main factors in decreasing the viability of shallot seeds. A moisture content of 6% and a storage temperature of 4-15 °C can maintain the germination power of shallot seeds for more than one year, while a storage temperature of 5 °C can inhibit biochemical and physiological processes that can reduce seed viability (Thirusendura et al., 2018). Seed deterioration processes, which include DNA damage such as autolysis and hydrolytic, membrane damage due to free radical peroxidation, changes in respiration activity due to mitochondrial damage, enzyme and protein changes, hormonal changes, and production of toxic metabolites due to detoxification system failure, resulting in seedling deformation and reduced seed viability (Demirkaya et al., 2010; Finch et al., 2016; Pehlivan, 2017; Farooq et al., 2021; Małecka et al., 2021).

Quality seeds are indispensable in supporting crop production. Increasing the quality of shallot seeds can be done by pruning during the nursery process (Faried et al., 2024b). Shallot seed has a high market demand for seed viability and vigor. Shallot seeds that have good quality have at least 70% germination. Shallot seeds are tiny and low-quality due to their sensitivity to extended shelf life. Seeds that are of low quality will also produce low and non-uniform germination and abnormal seedlings. The conditions of the growth environment, pre-seeding techniques, and treatments strongly influence the quality of shallot seeds. Seed viability and vigor vary widely according to the superiority of a cultivar (Brar et al., 2019; Hourston et al., 2020). The research conducted by Syam'un et al. (2024), showed that the Lokananta variety gave the best results compared to the Maserati variety. In this case, seed priming has promising potential in improving the survival of seeds that have low quality (Pagano et al., 2023). Sivritepe and Demirkaya (2012) investigated the effects of priming and osmotic conditioning applications on the germination of onion seeds and found that humidification applications gave more positive results on the germination of onion seeds.

One valuable and feasible hydration strategy for accelerating and uniformizing seed growth is seed priming. Seed priming as a plant growth regulator can increase the ability to germinate and grow plant seeds (Pangestuti, 2021). Not only in the germination process but the quality of plant seeds can be improved due to the priming process (Faried et al., 2024a). One of the seed priming agents that can improve the quality of deteriorated seeds is the micronutrient compound zinc (Zn). Protein synthesis, membrane integrity, and enzyme activation depend on zinc. During seed germination, low zinc concentrations disrupt essential metabolic processes, resulting in aberrant cell formation (Cakmak et al., 2023). Abiotic factors can cause zinc deficiencies, leading to poor photosynthetic performance, limited root growth in zinc uptake, decreased carbon assimilation and biomass production, elevated reactive oxygen species (ROS) levels, and accelerated oxidative stress. Administering zinc compounds can counteract plant development's negative impacts by preserving biological membranes' structural and functional integrity, controlling antioxidant activity, and improving photosynthetic performance (Rehman et al., 2018c and 2019; Ullah et al., 2019).

Using small amounts of nutrients to germinate seeds, seed priming makes it easier for seeds to obtain nutrients. Furthermore, compared to unprimed seeds, primed seeds germinate more quickly and uniformly (Farooq et al., 2020). Due to the high Zn concentration in the growing embryo, Zn treatment by seed priming improves plant performance by enabling earlier and more coordinated stand establishment. According to Rehman et al. (2018a and 2018b), this implies a potential function in the germination metabolism of Zn. In one study, chickpeas seed primed with 0.05% Zn solution had increased Zn content and yielded 19% and 29%. Similarly, Zn seed priming with arginine, histidine, and glycine raised the grain's protein level (Seddigh et al., 2016). $ZnSO_4$ seed priming increased grain yields in wheat compared to unprimed seeds (Rehman et al., 2018b and 2018c). Onion seeds (cultivar CO(On)5) were nutriprimed with 0.5% $ZnSO_4$ to boost germination percentage, vigor index, and dry matter production (Saranya et al., 2017).

Many studies have reported the positive impact of Zn application on seeds and plant growth and production. However, only some studies still provide information on using Zn compounds as seed priming agents to improve shallot seed quality. Therefore, this study examined the quality of shallot seedlings through seed priming technology with Zn compounds and explored the mechanism of shallot seedling emergence and growth.

2. Material and Methods

The research was conducted in the Green House, Faculty of Agriculture, Hasanuddin University, Makassar, Indonesia. The research site is located at coordinates 5°7'40.07 "S 119°28'48.94 "E. This research was conducted in June-July 2023 for 40 days. The research was arranged in a separate plot design (SPD) with a randomized group design (RGD). The main plot was the type variety consisting of

2 varieties, namely Lokananta and Maserati. The subplots are the type of Zn priming agent composed of 6, namely unpriming, hydropriming, IAA priming (z2), ZnO priming (z3), $ZnSO_4.7H_2O$ priming (z4), Zn-EDTA priming (z5). Twelve treatment combinations were obtained and repeated thrice, resulting in 36 experimental units—preparation of each solution Zn and IAA with a concentration of 100 ppm. The shallot seeds used were Lokananta and Maserati varieties, which suffered from seed deterioration with an initial germination amount of 15%. Shallot seeds were then primed according to the treatment. 4.5 g of seeds were added to each treatment solution in a 1:20 (W/V) ratio in a glass jar connected to an aerator. Then, the seeds were soaked for a duration according to the results of the lag phase determination for 20 hours (Faried et al., 2023). The seeds were then sown in pots containing a mixture of soil and compost 1:1 (v/v). Measurements were taken after the seedlings were 40 days after sowing. The data were then tabulated and analyzed using variance (ANOVA), and if there was significant data, it was further tested by DMRT test with $\alpha = 0.05$. Correlation analysis, principal component analysis, and path analysis were also conducted. According to Chiomento et al. (2020), the seedling quality index can be calculated through the Dixon Quality Index (DQI) mathematical model as follows:

$$SQI = \frac{SDW}{\frac{SH}{PSD} + \frac{CDW}{RDW}} \quad (1)$$

Description:
SQI = Seedling quality index
SDW = Seedling dry weight
SH = Seedling height
PSD = Pseudo-stem diameter
CDW = Shoot dry weight
RDW = Root dry weight

3. Results

The analysis of variance showed no interaction between the use of shallot seed varieties and the type of seed priming agent that significantly affected the germination percentage, seedling height, number of leaves, pseudo-stem diameter, root length, number of root tips, fresh weight, dry weight, and seedling quality index (Table 1, 2, 3).

Table 1. Effect of two varieties and types of seed priming agents on germination percentage (%), seedling height (cm), and number of leaves

Variety	Germination Percentage (%)	Seedling Height (cm)	Number of Leaves
Lokananta	32.54 ± 10.53 b	27.06 ± 1.33	3.94 ± 0.21
Maserati	56.35 ± 7.91 a	25.47 ± 1.07	3.63 ± 0.16
Priming Agent	**Germination Percentage (%)**	**Seedling Height (cm)**	**Number of Leaves**
Unprimed	16.67 ± 2.38 c	21.31 ± 0.48 b	3.08 ± 0.08 b
Hydropriming	33.33 ± 19.05 bc	26.18 ± 2.42 a	4.00 ± 0.33 ab
IAA	42.86 ± 23.81 abc	26.27 ± 1.86 a	3.83 ± 0.33 ab
ZnO	40.48 ± 21.43 abc	28.61 ± 1.24 a	4.18 ± 0.15 a
$ZnSO_4.7H_2O$	61.90 ± 9.52 ab	28.72 ± 0.13 a	3.66 ± 0.38 ab
Zn-EDTA	71.43 ± 4.76 a	26.51 ± 0.13 a	3.95 ± 0.18 ab

[1]Mean ± SE values with different letters in the column indicate significant ($p < 0.05$) differences by Duncan's new multiple-range test.

Table 2. Effect of two varieties and types of seed priming agents on pseudo-stem diameter (mm), root length (cm), and number of root tips

Variety	Pseudo-Stem Diameter (mm)	Root length (cm)	Number of Root Tips
Lokananta	2.92 ± 0.26	10.04 ± 0.62	11.43 ± 1.05
Maserati	2.87 ± 0.23	10.64 ± 1.33	11.07 ± 1.17
Priming Agent	**Pseudo-Stem Diameter (mm)**	**Root length (cm)**	**Number of Root Tips**
Unprimed	1.89 ± 0.13 b	6.73 ± 0.33 b	6.33 ± 0.67 b
Hydropriming	3.02 ± 0.25 a	10.55 ± 0.72 ab	12.67 ± 1.00 a
IAA	2.77 ± 0.11 a	9.80 ± 1.20 ab	11.62 ± 0.72 a
ZnO	3.36 ± 0.16 a	11.39 ± 0.93 a	11.18 ± 1.18 a
$ZnSO_4.7H_2O$	3.07 ± 0.25 a	10.49 ± 0.33 ab	12.77 ± 0.99 a
Zn-EDTA	3.26 ± 0.47 a	13.07 ± 2.80 a	12.94 ± 1.12 a

[2]Mean ± SE values with different letters in the column indicate significant ($p < 0.05$) differences by Duncan's new multiple-range test.

Table 3. Effect of two varieties and types of seed priming agents on fresh weight (g), dry weight (g), and seedling quality index

Variety	Fresh Weight (g)	Dry Weight (g)	Seedling Quality Index
Lokananta	1.92 ± 0.26	0.15 ± 0.02	0.0053 ± 0.00
Maserati	1.89 ± 0.29	0.16 ± 0.02	0.0061 ± 0.00
Priming Agent	**Fresh Weight (g)**	**Dry Weight (g)**	**Seedling Quality Index**
Unprimed	0.83 ± 0.12 b	0.07 ± 0.00 b	0.0023 ± 0.00 b
Hydropriming	1.88 ± 0.48 a	0.16 ± 0.01 a	0.0059 ± 0.00 a
IAA	1.90 ± 0.24 a	0.15 ± 0.00 a	0.0054 ± 0.00 a
ZnO	2.54 ± 0.06 a	0.22 ± 0.01 a	0.0081 ± 0.00 a
$ZnSO_4.7H_2O$	1.94 ± 0.01 a	0.16 ± 0.00 a	0.0059 ± 0.00 a
Zn-EDTA	2.34 ± 0.45 a	0.18 ± 0.05 a	0.0068 ± 0.00 a

[3] Mean ± SE values with different letters in the column indicate significant ($p < 0.05$) differences by Duncan's new multiple-range test.

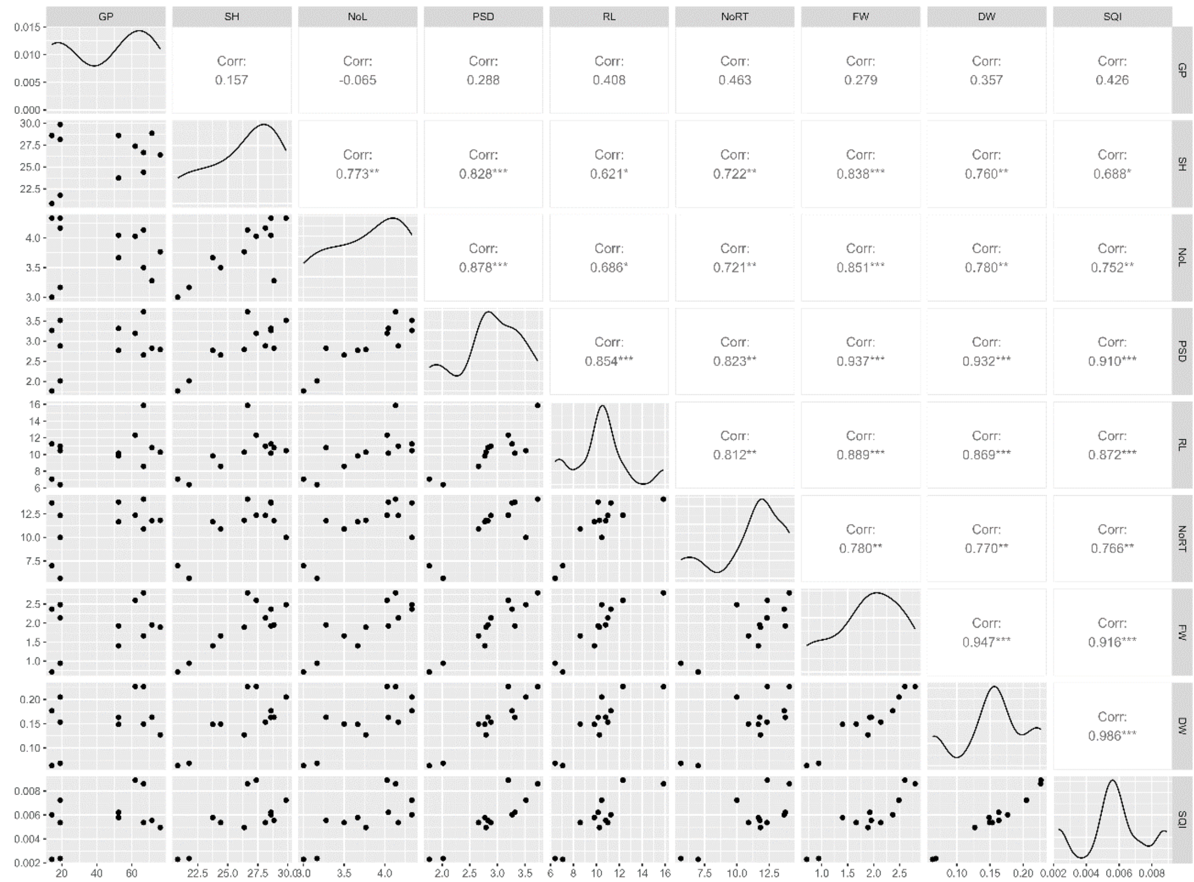

Figure 1. Correlation maps among parameters. (GP) germination percentage, (SH) seedling height, (NOL) number of leaves, (PSD) pseudo-stem diameter, (RL) root length, (NORT) number of root tips, (FW) fresh weight, (DW) dry weight, (SQI) seedling quality index.

The correlation map based on the observation parameters can be seen in (Figure 1). The correlation value is a parameter used in evaluating the relationship between characters. The correlation value is between -1 and 1, where if it is positive, then if the value of a character increases, it will increase the value of other characters. If it is negative, then a character's value increase will result in other characters' values decreasing. The correlation coefficient value is in the range of weak (<0.40), moderate (>0.4), and strong (>0.70) (Schober et al., 2018).

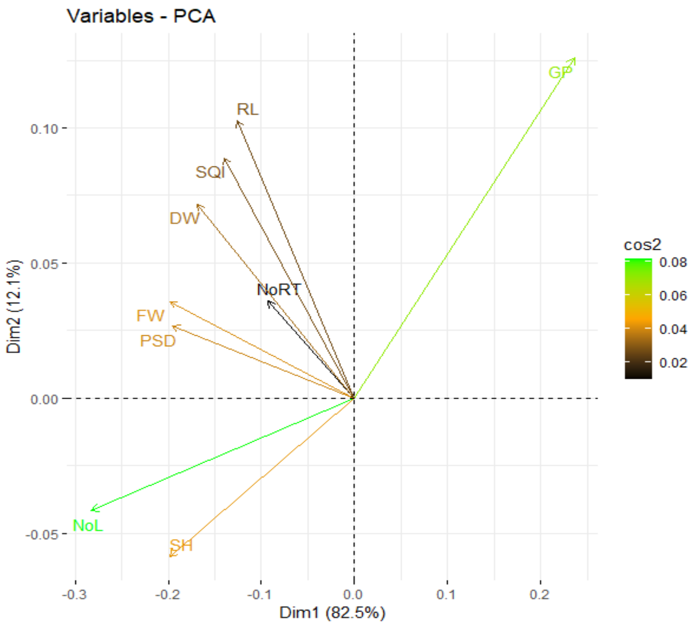

Figure 2. Principal component analysis. (GP) germination percentage, (SH) seedling height, (NOL) number of leaves, (PSD) pseudo-stem diameter, (RL) root length, (NORT) number of root tips, (FW) fresh weight, (DW) dry weight, (SQI) seedling quality index.

The PCA biplot produced a variation pattern visually showing the parameters' contribution in each treatment (Figure 2). This study showed that RL, SQI, DW, NORT, FW, and PSD contributed positively to the PC1 group (82.5%), as the angle between them was less than 90° (figure 2) in the case of the PC2 group, the parameters DW, NORT, FW, PSD, NOL, SH, which have a positive relationship to the PC2 group (12.1%). The GP parameters of NOL and SH showed a negative relationship between the parameters (judging by the angle greater than 90°). The trait relationships formed based on the PCA matrix align with the Pearson correlation trait (Figure 2).

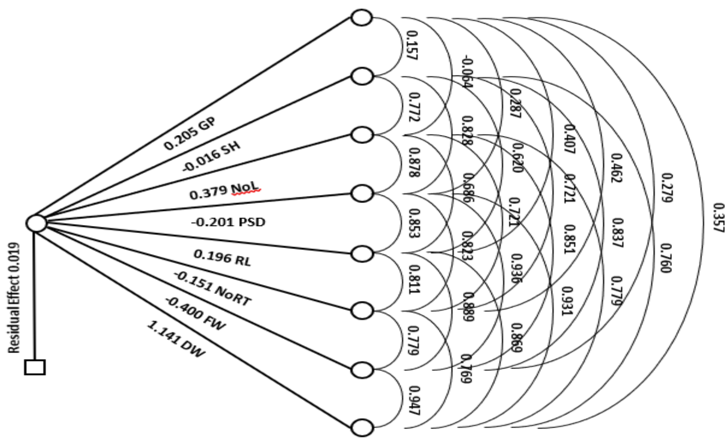

Figure 3. Path coefficient analysis diagram for seedling quality index (SQI). (GP) germination percentage, (SH) seedling height, (NOL) number of leaves, (PSD) pseudo-stem diameter, (RL) root length, (NORT) number of root tips, (FW) fresh weight, (DW) dry weight.

The results of path analysis between observation parameters can be seen (Figure 3). The parameter that provides a positive relationship and a very high direct effect on the seedling quality index is the dry weight (130.25%). The parameter that directly affects the seedling quality index is the number of leaves (14.42%). The parameter that has a medium direct effect on the seedling quality index is germination percentage (4.22%). The parameter with a low direct effect on the seedling quality index is root length (3.87%). The parameters that have a negative direct effect are seedling height (0.03%), pseudo-stem diameter (4.05%), number of root tips (2.29%), and fresh weight (16.00%). So, the seedling quality index can be selected by directly selecting the germination percentage, number of leaves, root length, and dry weight. In contrast, increasing these growth parameters can improve the shallot seedling quality index.

4. Discussion

This study examined the effects of seed priming techniques on two varieties of shallot that had regressed in terms of seedling growth and quality. This study evaluated various priming methods, including nutrient priming, hormone priming, and hydropriming. The outcomes demonstrated that, in comparison to no priming, the seed priming approach had a significant impact on every metric. Imran et al. (2021) state that seed priming treats seeds before sowing. It involves soaking the seeds in water or nutrient solutions, allowing them to air dry, and restoring their moisture content to storage levels. This can boost tolerance to various stress situations, quicken germination, and enhance crop quality.

The experimental results showed that Zn-EDTA priming increased the parameters of germination percentage, root length, and number of root tips. Zn-EDTA is a chelated Zn synthesis compound and a potential source of Zn nutrition in terms of increasing the efficiency of absorption by plants and inhibiting insoluble complex compounds (Poudel et al., 2023). Zhao et al. (2018) reported that applying Zn-EDTA increased the uptake and concentration of Zn content in wheat plants. This study showed a correlation between the number of root tips and the increase in root length, significantly influencing Zn uptake in the rhizosphere region. The increase in root length is due to the activation process of cellular respiration, which allows the seed coat to rupture so that the roots can more easily emerge.

$ZnSO_4.7H_2O$, a widely used Zn compound due to its high solubility, affordability, and availability, was found to play a crucial role in plant growth. The experimental results showed that priming with $ZnSO_4.7H_2O$ increased seedling height parameters. This is because Zn has a vital role in cell division by activating various enzymes involved in the development of plant cells (Hassan et al., 2020). Increasing seed Zn concentration increases plant seedling growth because nutrient reserves are significant when plant growth conditions are nutrient-limited. This agrees with the results of research conducted by Choukri et al. (2022) on corn, where priming $ZnSO_4.7H_2O$ increases the Zn content in seeds. Nutrient reserves, especially Zn, are significant in seeds to maintain seedlings at the beginning of growth until the roots formed can absorb other mineral nutrients from the growth medium.

The experiment results showed that ZnO priming had a significantly positive impact on seedling growth. It increased the number of leaves, pseudo-stem diameter, fresh weight, dry weight, and seedling quality index. The number of leaves illustrates the ability of plants to be able to photosynthesize; the more the number of leaves, the more assimilate produced can affect the growth of plant cells, which can have an impact on enlarging the pseudo-stem diameter and accumulating in plant tissues to increase plant weight. This is the opinion of Kumar et al. (2021), who claimed that the Zn priming treatment produced photosynthetic area, increased leaf area and number, and caused biomass buildup. The increase in plant biomass accumulation is also in line with the increase in the accumulation of Zn levels in plants. This is comparable to a study by Freitas et al. (2020), which discovered that corn seed embryos primed with ZnO had the most Zn accumulation. Compared to the control, Poudel et al. (2023) found that the Zn content of the plants increased by 84.7% in pea plants and 118.4% in sunflower plants. The increased availability of Zn can have a direct translocation effect on young seedlings and encourage the absorption of nitrogen nutrients from the soil during the plant growth phase so that it will produce high-quality seedlings. The provision of Zn through seed priming application is proven to effectively have a significant effect on the growth of shallot seedlings.

Conclusion

Based on the research, seed priming treatment with zinc micronutrients affects the growth of shallot seedlings. Neither variety has different responses to seed priming treatment. Zn-EDTA priming increases the parameters of germination percentage, root length, and number of root tips. $ZnSO_4.7H_2O$ priming produced the highest seedling height. ZnO priming recorded an increased number of leaves, pseudo-stem diameter, fresh weight, dry weight, and seedling quality index.

Ethical Statement

Ethical approval is not required for this study because this research does not cause any negative impacts that are harmful to human life and the environment.

Conflict of Interest

The authors declare that there are no conflicts of interest.

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Author Contributions

Elkawakib Syam'un (conceptualization, methodology, writing original draft, supervision), Syatriabty Andi Syaiful (conceptualization, methodology, supervision, data curation), Abdul Jalil (conceptualization, methodology, writing original draft, resources, formal analysis).

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