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Interactive effects of plant growth-promoting rhizobacteria and drought stress on the growth and yield of shallot (*Allium ascalonicum* L.) cultivated in saline soil

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ABSTRACT

This study evaluated the effects of plant growth-promoting rhizobacteria (PGPR) and drought-stress timing on the growth, yield, and physiological responses of shallot (*Allium ascalonicum* L.) cultivated in saline soil. The experiment was conducted using a factorial randomized block design with PGPR concentrations of 0, 20, and 40 mL L^{-1} and drought-stress timing consisting of no stress, early growth stress (1–14 DAP), vegetative stress (15–28 DAP), and bulb formation stress (29–42 DAP). The final soil EC reached 6 dS m^{-1}, while soil moisture during drought periods declined to 30–50%. PGPR improved plant length, leaf area, tiller number, plant dry weight, and net assimilation rate. PGPR at 40 mL L^{-1} was more effective for vegetative growth, whereas 20 mL L^{-1} tended to enhance proline and flavonoid accumulation. Drought during early and vegetative phases caused the greatest reductions in growth and yield. These findings indicate that PGPR supports shallot performance in saline soil, but its effectiveness depends strongly on water availability during critical growth phases.

Keywords: abiotic stress, drought phase, PGPR, shallot, saline soil.

INTRODUCTION

Shallot (*Allium ascalonicum* L.) is an important horticultural commodity with high economic value and broad utilization as a food ingredient. Its productivity, however, is highly dependent on suitable soil conditions and adequate water availability. In saline soils, high salt concentration increases osmotic pressure and disrupts water and nutrient uptake, thereby inhibiting vegetative growth, reducing photosynthetic efficiency, disturbing ionic balance, and limiting bulb formation. Previous studies reported that salinity can reduce shallot yield components, including bulb number, bulb diameter, and dry bulb weight (Aini et al., 2019; Munns and Gilliham, 2015).

In addition to salinity, drought stress is another major abiotic factor limiting shallot growth and yield. Shallot plants are sensitive to water deficit because they have a relatively shallow root system, making them vulnerable when soil water availability decreases. Drought stress can reduce leaf area, chlorophyll content, photosynthetic rate, biomass accumulation, and bulb development, especially when stress occurs during early growth and vegetative phases. The combination of salinity and drought may impose more complex physiological constraints than a single stress condition because both stresses reduce water availability and intensify osmotic imbalance in plant tissues (Pandey et al., 2017).

Plant growth-promoting rhizobacteria (PGPR) offer a biological approach to improve plant tolerance under unfavorable environments. PGPR can support plant growth through nutrient solubilization, phytohormone production, root development

stimulation, and stress ethylene reduction through ACC deaminase activity (Ahemad and Kibret, 2014). In saline soils, salt-tolerant PGPR may improve crop performance by enhancing nutrient acquisition, regulating stress-related hormones, and reducing ionic toxicity (Egamberdieva et al., 2019). Under drought conditions, PGPR can also improve root growth and plant water status, thereby helping plants maintain water and nutrient uptake during periods of limited water availability (El-Saadony et al., 2024). However, the effectiveness of PGPR in mitigating drought stress at different growth phases of shallot cultivated in saline soil remains unclear, although previous work has shown that PGPR responses in shallot may vary depending on the timing of drought stress (Pratiwi et al., 2024).

Recent studies have emphasized that crop performance under environmental stress is closely linked to the interaction between water availability, root development, microbial activity, and nutrient management. Soil environmental conditions can influence root growth and microbial diversity, which are essential for nutrient cycling and plant adaptation under stress (Patah and Othman, 2024). Water availability is also a central component of agroecosystem resilience because changes in precipitation effectiveness and irrigation efficiency directly affect crop yield stability (Sikder and Mumit, 2024; Wuyep et al., 2026). In addition, nutrient management remains important for supporting crop morphological development under field conditions (Khokhar et al., 2025). These findings strengthen the relevance of evaluating PGPR application under different drought-stress timings in saline soil as part of biological and water-management strategies in shallot production.

MATERIALS AND METHODS

The experiment was conducted in an experimental field located in Wonorejo Village, Poncokusumo District, Malang Regency, East Java, Indonesia, from December 20, 2025, to April 6, 2026. The site is located at 8°02′54″ S and 112°47′03″ E, at an altitude of approximately 500–600 m above sea level, with an average daily temperature of 20–22 °C. Shallot plants were grown in wooden planting boxes measuring 80 × 80 × 30 cm. The planting medium consisted of soil collected from the experimental site, which was loosened and filled into the boxes, leaving approximately 5 cm from the top edge. Soil salinization was conducted before planting by applying an industrial salt solution at a concentration of 5.8 g L^{-1} evenly to the planting medium until field capacity. The soil was then mixed to obtain uniform salinity and incubated for three days. At the end of the experiment, the electrical conductivity (EC) of the planting medium was measured using an EC meter and reached 6 dS m^{-1}, confirming that the growing medium remained within the saline soil category. Each planting box was covered with a transparent polyethylene (PE) plastic shelter supported by bamboo frames to prevent rainfall from entering the planting medium and to create controlled drought-stress conditions. The plastic shelter was arch-shaped, 160 cm wide, 8.6 m long, and approximately 1.5 m high from the surface of the planting box. Soil analysis was conducted three days after salt application to determine the initial characteristics of the saline planting medium. text Soil analysis was conducted three days after salt application to determine the initial chemical characteristics of the planting medium. text Soil analysis was conducted three days after salt application to determine the initial chemical characteristics of the planting medium. The soil had 8% water content, 0.16% total N, 364 ppm available P_2O_5 using the Bray I method, 0.22 cmol(+) kg^{-1} exchangeable K, 0.79% organic C, and 23.28 cmol(+) kg^{-1} cation exchange capacity.

Uniform shallot bulbs of the Tajuk variety were used as planting material. Before planting, the bulbs were selected based on healthy, undamaged, pest-free, disease-free, and relatively uniform size criteria. The PGPR inoculum used in this study was a commercial product, Bactegrowth, produced by PT. Eureeka Great Nusantara, Indonesia. Based on the product information, the formulation contained several plant growth-promoting bacteria, including *Bacillus subtilis* at 9.6×10^{12} CFU g^{-1}, *Pseudomonas fluorescens* at 1.0×10^{12} CFU g^{-1}, *Azotobacter* sp. at 1.0×10^{12} CFU g^{-1}, and *Azospirillum* sp. at 1.0×10^{12} CFU g^{-1}. The PGPR was diluted in water according to the assigned treatment concentrations, namely 0, 20, and 40 mL L^{-1}. PGPR application was carried out four times during the experiment, namely 14 days before planting, at planting, 14 DAP, and 28 DAP, by watering the solution around the shallot root zone at a volume of 100 mL $plant^{-1}$. The viable bacterial population was expressed as colony-forming units (CFU) based on the commercial

formulation information. Planting was carried out two weeks after the first PGPR application by placing one shallot bulb in each planting hole at a spacing of 20 × 20 cm, resulting in 16 plants per planting box. Inorganic fertilization consisted of 200 kg ha^{-1} N, 120 kg ha^{-1} P_2O_5, and 120 kg ha^{-1} K_2O, supplied through urea, ZA, and NPK Phonska Plus 15-15-15. Fertilizer was applied in three stages at 7, 21, and 35 days after planting (DAP) using the dibbling method.

A factorial randomized block design (RBD) was used, consisting of two factors: PGPR concentration and drought-stress timing. The first factor was PGPR concentration with three levels: P0 = 0 mL L^{-1}, P1 = 20 mL L^{-1}, and P2 = 40 mL L^{-1}. The second factor was drought-stress timing with four levels: K0 = without drought stress, K1 = drought stress during the early growth phase (1–14 DAP), K2 = drought stress during the vegetative phase (15–28 DAP), and K3 = drought stress during the bulb formation phase (29–42 DAP). These treatments resulted in 12 treatment combinations, each repeated three times, giving a total of 36 experimental units. Each experimental unit consisted of one planting box containing 16 shallot plants.

Drought stress was imposed by withholding irrigation during specific shallot growth phases. In the non-stressed treatment (K0), plants were irrigated once daily to maintain soil moisture above 80%. For each irrigation event, 10 L of water was applied to each planting box, corresponding to field capacity. In the drought-stress treatments, irrigation was withheld according to the assigned stress periods, and soil moisture during these periods declined to approximately 30–50%, as monitored using a soil moisture meter. In K1, irrigation was withheld during the early growth phase from 1 to 14 DAP and then resumed until harvest. In K2, plants were irrigated normally until 14 DAP, irrigation was withheld from 15 to 28 DAP, and watering was resumed afterward. In K3, plants were irrigated normally until 28 DAP, irrigation was withheld from 29 to 42 DAP, and then resumed until harvest. During non-stress periods, K1, K2, and K3 received the same irrigation regime as K0. Transparent polyethylene shelters were installed above the planting boxes to prevent rainfall from entering the growing medium, thereby maintaining the assigned drought-stress conditions during the experiment.

The observed variables consisted of growth, yield, and physiological parameters. Growth parameters included plant length, number of leaves, leaf area, number of tillers, plant dry weight, net assimilation rate (NAR), and relative growth rate (RGR). For plant dry weight measurement, plant samples were separated into roots, stems, leaves, and bulbs, then oven-dried at 80 °C for 2 × 24 h before weighing. Yield parameters were observed at harvest and included harvest fresh weight, harvest dry weight, and bulb weight. Harvest dry weight was determined after the harvested plants were air-dried. Physiological parameters included proline and flavonoid contents.

$$\mathrm{NAR} = \frac{W_2 - W_1}{t_2 - t_1} \times \frac{ln\, A_2 - ln\, A_1}{A_2 - A_1} \quad (1)$$

$$\mathrm{RGR} = \frac{ln\, W_2 - ln\, W_1}{t_2 - t_1} \quad (2)$$

where: W_1 and W_2 are plant dry weights at times t_1 and t_2 (g), A_1 and A_2 are leaf areas at times t_1 and t_2 (cm^2), and t_1 and t_2 are the observation times (days).

Proline content was analyzed from composite leaf samples collected at harvest for each treatment combination. Proline was determined using the Bates method (Bates et al., 1973). Fresh leaf tissue of 0.5 g was homogenized and extracted with 10 mL of 3% sulfosalicylic acid, then centrifuged at 6000 rpm for 30 min to obtain the supernatant. A 2 mL aliquot of the filtrate was mixed with 2 mL glacial acetic acid and 2 mL acid-ninhydrin reagent. The mixture was heated in a boiling water bath for 1 h and immediately cooled in an ice bath to stop the reaction. Subsequently, 6 mL toluene was added, and the mixture was stirred for 20–30 s to extract the proline-ninhydrin complex into the toluene layer. The toluene phase was separated and measured at 520 nm using a spectrophotometer. Proline concentration was calculated based on a standard curve and expressed as µM g^{-1} fresh weight.

Total flavonoid content was determined using the $NaNO_2$-$AlCl_3$-NaOH colorimetric method described by Jia et al. (1999), with slight modifications as applied in recent plant flavonoid studies. Shallot leaf samples were dried and ground into powder. A 0.05 g sample was extracted using 50% acetone at a ratio of 1:10 (w/v) through maceration for 18 h at room temperature. The extract was filtered, and the filtrate was diluted with acetone at a ratio of 1:5 (v/v). The diluted extract was reacted with 150 µL $NaNO_2$ and allowed to stand for 6 min, followed by the addition of 300 µL $AlCl_3$ and incubation for 5 min. Then, 1 mL of

1 M NaOH was added, and the final volume was adjusted to 5 mL with distilled water before homogenization. Absorbance was measured at 510 nm using a spectrophotometer. Total flavonoid content was determined using a quercetin standard curve and expressed as mg quercetin equivalent per liter (mg QE L^{-1}).

The collected data were analyzed using analysis of variance (ANOVA) based on a factorial randomized block design with DSAASTAT software. When significant treatment effects were detected, mean comparisons were performed using the honestly significant difference (HSD) test at the 5% significance level. Data tabulation, diagrams, and graphical presentations were prepared using Microsoft Excel. The results are presented as mean ± standard deviation (SD) when replicate data were available. Error bars in figures indicate SD. Different letters in tables or figures indicate significant differences among treatments according to the HSD test at $p \leq 0.05$. Because proline and flavonoid analyses were conducted using composite samples, SD and error bars were not calculated for these biochemical variables.

RESULTS AND DISCUSSION

The early vegetative response of shallot was reflected in plant length, number of leaves, and leaf area (Table 1). PGPR application mainly improved plant length and leaf area, indicating a positive contribution of rhizobacteria to canopy development under saline soil conditions. In contrast, drought-stress timing produced a more distinct response, particularly when water deficit occurred during the early growth phase. This treatment consistently resulted in lower plant length, leaf number, and leaf area than the non-stressed treatment, suggesting that water limitation at the initial stage restricted early canopy establishment and reduced the capacity of plants to develop photosynthetic organs.

The improvement in plant length and leaf area under PGPR application suggests that rhizobacteria supported early vegetative growth of shallot in saline soil. The highest plant length and leaf area were observed at 40 mL L^{-1} PGPR, indicating that this concentration was more effective in promoting canopy expansion than the untreated control. This response may be associated with the ability of PGPR to stimulate root development, increase nutrient availability, and produce growth-promoting substances such as auxins, cytokinins, and gibberellins (Ahemad and Kibret, 2014; Gouda et al., 2018). A better-developed root system allows plants to absorb water and nutrients more efficiently, which is particularly important under saline conditions where osmotic pressure can restrict water uptake. Similar responses have been reported in shallot, where PGPR improved root development, canopy growth, leaf area, and photosynthetic activity (Hau et al., 2025; Hamid et al., 2024).

However, PGPR did not significantly affect the number of leaves, indicating that its effect was more pronounced on plant size and leaf expansion than on leaf initiation. Leaf number is often influenced by genetic characteristics and the physiological stage of the plant, whereas leaf area is more sensitive to environmental conditions and plant water status. Therefore, the increase in leaf

Table 1. Effects of PGPR concentration and drought-stress timing on plant length, number of leaves, and leaf area of shallot. Values are presented as mean ± SD. Different letters within the same column indicate significant differences according to the HSD test at $p \leq 0.05$. ns = not significant; DAP = days after planting

Treatment	Result		
	Plant length (cm)	Number of leaves (leaves clump^{-1})	Leaf area (cm^2 clump^{-1})
Without PGPR	15.96 ± 2.71 a	12.35 ± 3.98	25.04 ± 13.90 a
PGPR 20 ml L^{-1}	18.48 ± 3.64 a	13.41 ± 4.48	43.02 ± 22.88 ab
PGPR 40 ml L^{-1}	18.65 ± 2.81 b	12.87 ± 4.39	49.19 ± 34.02 b
HSD 5%	2.43	ns	18,47
Without drought stress	19.80 ± 2.29 b	14.86 ± 1.76 b	57.56 ± 30.82 b
Early phase (1-14 DAP)	14.37 ± 1.61 a	6.55 ± 1.70 a	13.17 ± 9.14 a
Vegetative phase (15-28 DAP)	17.11 ± 3.52 ab	16.13 ± 2.26 b	41.96 ± 25.80 b
Bulb formation phase (29-42 DAP)	19.51 ± 2.07 b	13.97 ± 1.73 b	43.65 ± 13.28 b
HSD 5%	3.11	2.54	23.58

area under PGPR application reflects an improvement in photosynthetic surface development rather than an increase in the number of leaves.

Drought stress imposed during the early growth phase caused the strongest reduction in plant length, number of leaves, and leaf area. This indicates that the early growth phase is a critical period for shallot establishment. During this stage, plants are still developing their root system and initial canopy; therefore, water deficit can directly limit cell division, cell elongation, and leaf expansion. Shallot is particularly sensitive to drought because of its relatively shallow root system, which limits its ability to access deeper soil water (Pejic et al., 2011; Hafizh et al., 2021). Reduced leaf area under drought stress also decreases light interception and photosynthetic capacity, ultimately restricting biomass accumulation and yield formation (Manurung et al., 2022; Unzilaturrohmah et al., 2025).

Compared with early drought, stress during the vegetative and bulb formation phases showed a relatively lower suppressive effect on several growth parameters. This suggests that shallot plants may have developed a stronger root system and canopy structure before drought occurred, allowing partial recovery after stress was relieved. Nevertheless, the overall pattern shows that adequate water availability during early growth is essential for supporting vegetative development. Thus, PGPR application can improve several growth traits, but its effectiveness remains dependent on the timing of drought stress and the availability of water during critical growth stages.

The interaction between PGPR concentration and drought-stress timing was further observed in the number of tillers and plant dry weight (Figure 1). These parameters reflect the ability of shallot plants to maintain vegetative development and biomass accumulation under different stress conditions. The response pattern shows that the effect of PGPR varied depending on the timing of drought stress, indicating that the contribution of PGPR to plant growth was closely related to water availability during each growth phase.

The interaction between PGPR concentration and drought-stress timing in Figure 1 shows that tiller formation and dry matter accumulation were controlled by the combined effect of biological input and water availability. The higher tiller number under PGPR 40 mL L^{-1} indicates that rhizobacteria application may improve the vegetative development of shallot grown in saline soil. This response is consistent with Rahmandhias et al. (2024), who reported that PGPR application improved shallot growth and yield performance under saline soil conditions. In this study, however, the positive response of PGPR was more evident when drought pressure was relatively lower, suggesting that PGPR supported growth but did not completely overcome water limitation.

The highest plant dry weight under PGPR 40 mL L^{-1} without drought stress indicates that biomass accumulation was optimized when PGPR application was accompanied by sufficient water availability. Under favorable moisture conditions, PGPR may enhance root function and nutrient acquisition, allowing plants to maintain better photosynthetic activity and assimilate production.

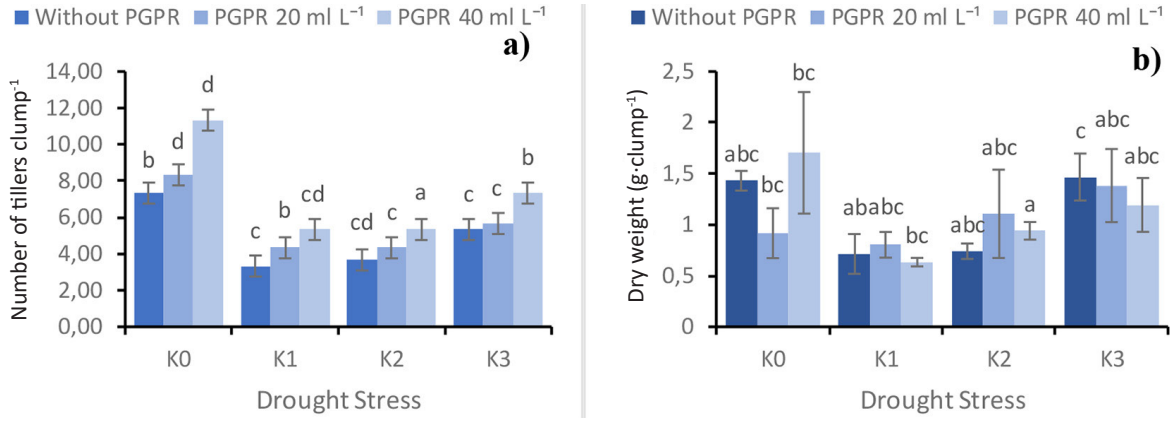

Figure 1. Effects of drought stress (K0 = control; K1 = early growth phase, 1–14 DAP; K2 = vegetative phase, 15–28 DAP; K3 = bulb formation phase, 29–42 DAP) and PGPR concentration (0, 20, and 40 mL L^{-1}) on (a) number of tillers and (b) dry weight of shallot. Bars represent mean values, and error bars indicate standard deviation (SD). Different letters indicate significant differences according to the HSD test at $p \leq 0.05$

Pratiwi et al. (2024) also reported that the response of shallot to PGPR under drought stress depends on the timing of stress exposure, particularly because drought can alter photosynthetic performance. Therefore, the lower dry weight observed under early and vegetative drought stress suggests that water deficit during active growth stages restricted dry matter accumulation despite PGPR application.

The reduction in tiller number and plant dry weight under early and vegetative drought stress may also be related to the combined osmotic pressure caused by saline soil and limited water availability. Salinity can disturb water uptake and ion balance, while drought further reduces plant water status and restricts physiological activity (Shrivastava and Kumar, 2015). Under combined abiotic stress, plants often show more complex responses than under a single stress condition because several physiological processes are affected simultaneously (Pandey et al., 2017). Thus, PGPR application can be considered a supportive strategy for improving shallot growth in saline soil, but adequate water management during early and vegetative phases remains essential to maintain biomass production.

The decline in growth under early and vegetative drought stress confirms that water availability is a major determinant of crop performance, as reported by Wuyep et al. (2026), who emphasized the relationship between effective precipitation and yield stability. In addition, the positive tendency of PGPR-treated plants may be associated with improved root function and rhizosphere activity, since soil conditions strongly influence root development and microbial diversity (Patah and Othman, 2024).

The effects of PGPR concentration and drought-stress timing on shallot yield components are presented in Table 2. Yield performance was evaluated based on harvest fresh weight, harvest dry weight, and bulb weight. Unlike several growth parameters, the yield response was more strongly influenced by drought-stress timing than by PGPR concentration. This indicates that water availability during critical growth phases played an important role in determining final bulb production.

Table 2 shows that PGPR concentration did not significantly affect harvest fresh weight, harvest dry weight, or bulb weight, although the mean values tended to be higher under PGPR application than without PGPR. PGPR at 20 mL L^{-1} increased the mean values of harvest fresh weight, harvest dry weight, and bulb weight compared with the untreated control, but the increase was not statistically significant. This indicates that PGPR had the potential to support yield formation, but its effect was not strong enough to produce a significant difference under saline soil and drought-stress conditions. PGPR may improve plant performance in saline environments by enhancing nutrient availability and reducing the negative effects of salt stress, but its effectiveness depends on root colonization, environmental conditions, and water availability (Paul and Lade, 2014; Rubin et al., 2017).

In contrast, drought-stress timing had a clearer effect on yield components. The non-stressed treatment produced the highest harvest fresh weight, harvest dry weight, and bulb weight, indicating that sufficient water availability was essential for bulb development. Drought

Table 2. Effects of PGPR concentration and drought-stress timing on harvest fresh weight, harvest dry weight, and bulb weight of shallot. Values are presented as mean ± SD. Different letters within the same column indicate significant differences according to the HSD test at $p \leq 0.05$; ns = not significant; DAP = days after planting

Treatment	Result		
	Fresh harvest weight (g clump^{-1})	Dry harvest weight (g clump^{-1})	Bulb weight (g clump^{-1})
Without PGPR	14.16 ± 6.89	11.30 ± 5.20	10.35 ± 4.97
PGPR 20 ml L^{-1}	18.60 ± 9.01	14.51 ± 6.30	13.08 ± 6.34
PGPR 40 ml L^{-1}	16.71 ± 5.31	13.26 ± 3.94	12.17 ± 3.83
HSD 5%	ns	ns	ns
Without drought stress	23.34 ± 8.27 a	17.94 ± 5.70 a	16.62 ± 5.79 a
Early phase (1-14 DAP)	12.79 ± 4.00 b	10.22 ± 2.60 b	9.34 ± 2.42 b
Vegetative phase (15-28 DAP)	13.65 ± 6.55 b	10.81 ± 5.18 b	9.80 ± 5.02 b
Bulb formation phase (29-42 DAP)	16.18 ± 5.07 ab	13.12 ± 3.66 ab	11.70 ± 3.63 ab
HSD 5%	8.64	6.24	6.20

stress during the early and vegetative phases caused marked reductions in yield because these stages are closely related to root establishment, canopy development, and assimilate production. Limited water availability during these phases may reduce photosynthetic capacity and restrict assimilate translocation to bulbs, resulting in lower bulb biomass. Similar findings were reported by Ariska and Rachmawati (2017), who stated that bulb formation is strongly related to photosynthate accumulation during vegetative growth and bulb filling.

The reduction in bulb weight under drought stress may also be associated with the combined effect of saline soil and water deficit. Salinity can increase osmotic pressure around the root zone, making water absorption more difficult, while drought further limits water availability for physiological processes. Under such conditions, plants require more energy to maintain osmotic adjustment, leaving less assimilate available for bulb enlargement. Aini et al. (2019) reported that salinity can reduce shallot yield components, including bulb number, bulb diameter, and dry bulb weight. Therefore, although PGPR application showed a positive tendency, water availability during early and vegetative growth remained the dominant factor determining shallot yield in saline soil.

The responses of net assimilation rate (NAR) and relative growth rate (RGR) to PGPR concentration and drought-stress timing are shown in Figure 2. These parameters describe the physiological efficiency of plants in producing and accumulating dry matter during growth. Changes in NAR and RGR indicate how shallot plants adjusted their assimilate production and growth rate under different PGPR concentrations and drought-stress phases.)

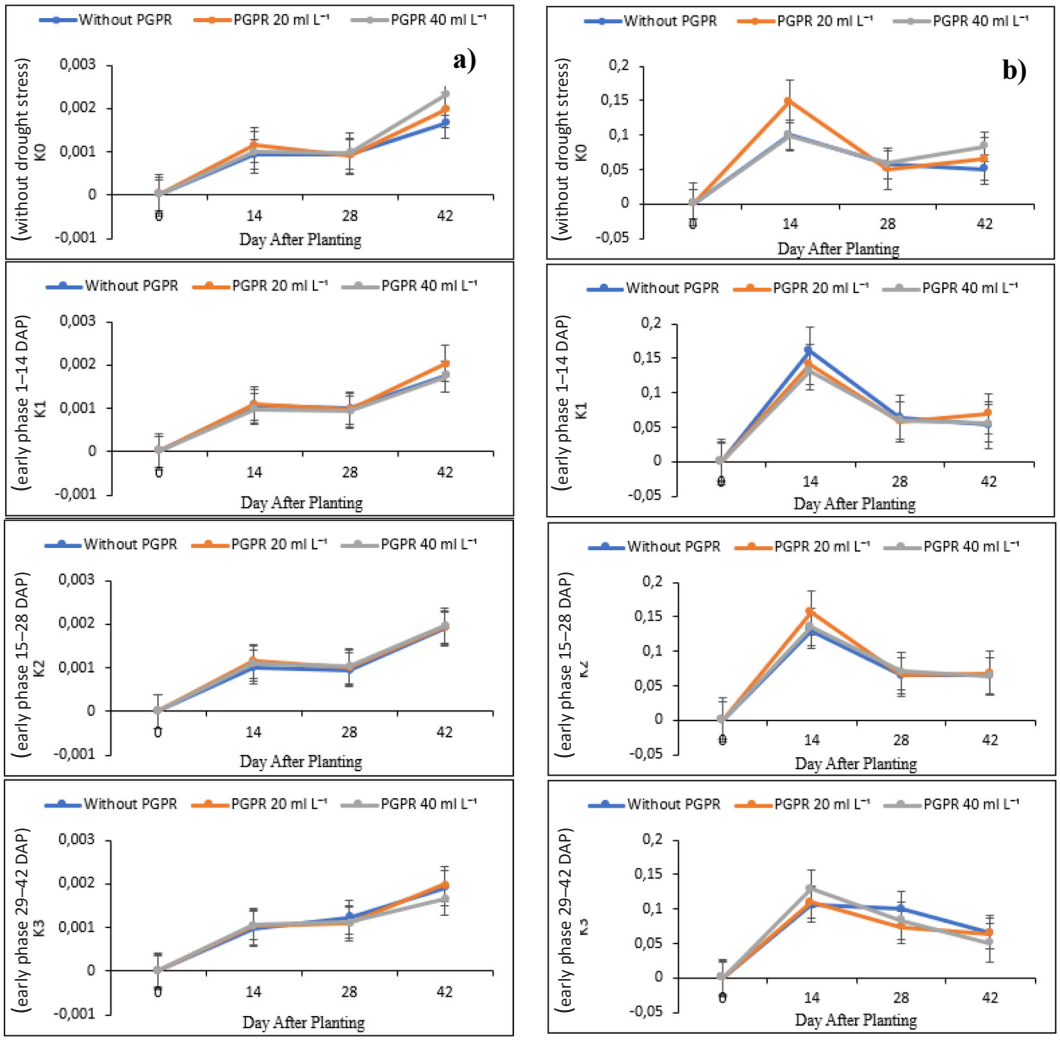

Figure 2. Effects of drought stress (K0 = control; K1 = early 1–14 DAP; K2 = vegetative 15–28 DAP; K3 = bulb formation phase 29–42 DAP) and concentration of PGPR (0, 20, 40 ml L^{-1}) on (a) net assimilation rate, (b) relative growth rate. Values are presented as mean, and error bars indicate standard deviation (SD

Figure 2 shows that PGPR application influenced the physiological growth efficiency of shallot, particularly through changes in NAR and RGR. The higher NAR under PGPR application indicates that rhizobacteria may improve the efficiency of dry matter formation per unit leaf area. In this study, PGPR at 20 mL L^{-1} tended to provide a stronger physiological response than the untreated control, suggesting that a moderate PGPR concentration was sufficient to support assimilate production. This response may be associated with improved root activity and better physiological performance of shallot under stress conditions, as reported by Saryanah et al. (2024), who found that growth-promoting bacteria enhanced biochemical and growth responses of shallot under drought stress.

The response of NAR and RGR was also affected by the timing of drought stress. Drought imposed during the early growth phase resulted in lower growth efficiency because plants were still establishing roots and leaves. Limited water availability at this stage can reduce leaf expansion, nutrient uptake, and photosynthetic capacity, leading to lower dry matter accumulation. Swasono (2012) reported that drought stress can suppress root growth and reduce water and nutrient absorption in shallot. Therefore, the decrease in NAR and RGR under early drought stress reflects the limitation of plants in producing and converting assimilates into new biomass.

Interestingly, drought stress during the bulb formation phase did not always result in the lowest NAR and RGR. This may indicate that shallot plants had already developed sufficient vegetative structure before stress occurred, allowing them to maintain assimilate production and direct photosynthates toward bulb development. During bulb formation, assimilates are increasingly allocated to storage organs, so changes in NAR and RGR may reflect a shift from vegetative expansion to dry matter partitioning into bulbs. Similar patterns have been explained by Bararyenya et al. (2020), who stated that assimilate allocation in bulb crops shifts from vegetative growth toward bulb enlargement. Thus, NAR and RGR responses in this study suggest that PGPR can support physiological growth efficiency, but the final response depends strongly on the growth phase during which drought stress occurs.

The physiological responses of shallot to PGPR concentration and drought-stress timing were further evaluated through proline and flavonoid contents (Figure 3). These compounds are closely associated with plant defense mechanisms under abiotic stress. Proline functions as an osmoprotectant that helps maintain cellular water balance, while flavonoids act as antioxidant compounds that protect plant tissues from oxidative damage. Therefore, changes in proline and flavonoid contents indicate the biochemical adjustment of shallot plants under saline soil and drought-stress conditions.

Figure 3 shows that PGPR application and drought-stress timing altered the biochemical response of shallot, as indicated by changes in proline and flavonoid contents. PGPR at 20 mL L^{-1} produced higher proline content than the untreated control, suggesting that this concentration may stimulate osmotic adjustment under saline and drought-stress conditions. Proline accumulation is commonly associated with plant adaptation to

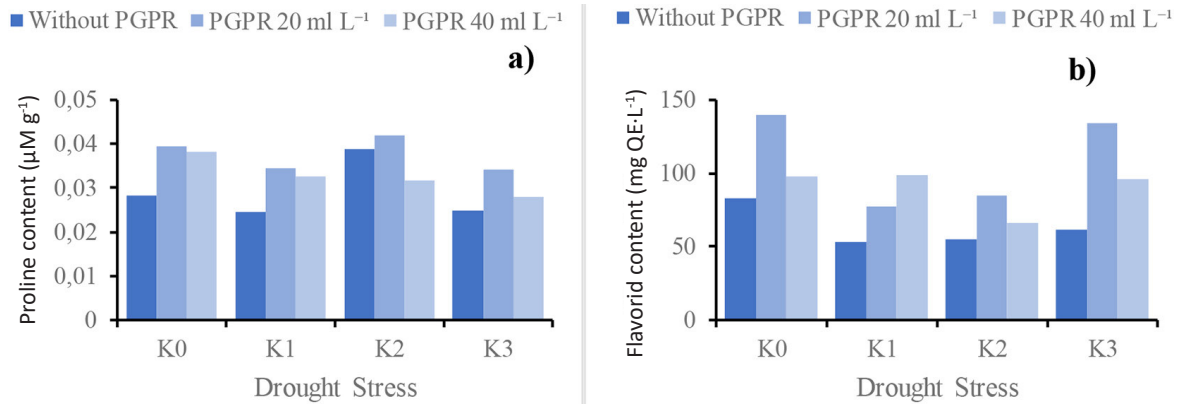

Figure 3. Effects of drought stress (K0 = control; K1 = early 1–14 DAP; K2 = vegetative 15–28 DAP; K3 = bulb formation phase 29–42 DAP) and concentration of PGPR (0, 20, 40 ml L^{-1}) on (a) proline content, (b) flavonoid content. Values were obtained from composite samples for each treatment combination; therefore, error bars are not shown

water deficit because it helps maintain cell turgor, protects cellular structures, and supports osmotic balance during stress exposure (Larasani and Violita, 2021). Therefore, the increase in proline under PGPR application indicates that rhizobacteria may contribute not only to growth promotion but also to stress-response regulation.

Drought-stress timing also affected proline accumulation, with the vegetative phase showing the highest proline content. This response indicates that drought during active vegetative growth imposed stronger physiological pressure on shallot plants. During this phase, leaf expansion, photosynthesis, and biomass accumulation occur intensively; therefore, water limitation may trigger stronger osmotic adjustment. Proline acts as an osmoprotectant that helps plants maintain turgor and cellular stability under drought conditions (Meena et al., 2019). The increase in proline during vegetative drought therefore reflects an adaptive response to maintain physiological function when water availability becomes limited.

Flavonoid content showed a different response pattern. PGPR at 20 mL L^{-1} produced the highest flavonoid content, indicating that moderate PGPR application may stimulate secondary metabolite accumulation. Flavonoids are antioxidant compounds involved in reducing oxidative damage caused by abiotic stress. However, flavonoid content was higher under non-stressed conditions than under drought stress, suggesting that severe or prolonged water limitation may reduce the plant's capacity to synthesize secondary metabolites. This condition may occur because drought stress can suppress photosynthesis and reduce assimilate availability for secondary metabolism (Ahmed et al., 2021). Thus, the biochemical response of shallot in this study indicates that PGPR may enhance proline and flavonoid accumulation, but the magnitude of this response depends on drought timing and plant physiological status.

CONCLUSIONS

This study demonstrated that PGPR application and drought-stress timing affected the growth, yield, and physiological responses of shallot cultivated in saline soil. The interaction between both factors was observed in tiller number and plant dry weight, indicating that the effect of PGPR depended on the timing of drought stress. PGPR at 40 mL L^{-1} was more effective in improving vegetative growth, while PGPR at 20 mL L^{-1} supported physiological responses through higher proline and flavonoid contents. However, drought stress during the early growth and vegetative phases remained the main limiting factor for shallot growth and yield. Therefore, PGPR application can be used as a supportive strategy to improve shallot performance in saline soil, but adequate water management during early and vegetative growth is essential to maintain optimal yield.

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REFERENCES

1. Ahemad, M., Kibret, M. (2014). Mechanisms and applications of plant growth promoting rhizobacteria: Current perspective. *Journal of King Saud University - Science*, *26*(1), 1–20. https://doi.org/10.1016/j.jksus.2013.05.001
2. Ahmed, U., Rao, M. J., Qi, C., Xie, Q., Noushahi, H. A., Yaseen, M., Shi, X., Zheng, B. (2021). Expression profiling of flavonoid biosynthesis genes and secondary metabolites accumulation in Populus under drought stress. *Molecules*, *26*(18), 5546. https://doi.org/10.3390/molecules26185546
3. Aini, N., Yamika, W. S. D., Aini, L. Q., Azizah, N., Sukmarani, E. (2019). Pengaruh rhizobacteria pada pertumbuhan dan hasil tanaman bawang merah (*Allium ascalonicum* L.) pada kondisi salin. *Jurnal Hortikultura Indonesia*, *10*(3), 182–189. https://doi.org/10.29244/jhi.10.3.182-189
4. Ariska, N., Rachmawati, D. (2017). Pengaruh ketersediaan air berbeda terhadap pertumbuhan dan hasil tiga kultivar bawang merah (*Allium cepa* L.). *Jurnal Agrotek Lestari*, *4*(2), 42–50. https://doi.org/10.35308/jal.v3i2.609
5. Bararyenya, A., Olukolu, B. A., Tukamuhabwa, P., Gruneberg, W. J., Ekaya, W., Low, J.,

Ochwo-Ssemakula, M., Odong, T. L., Talwana, H., Badji, A., Kyalo, M., Nasser, Y., Gemenet, D., Kitavi, M., Mwanga, R. O. M. (2020). Genome-wide association study identified candidate genes controlling continuous storage root formation and bulking in hexaploid sweetpotato. *BMC Plant Biology*, *20*, 3. https://doi.org/10.1186/s12870-019-2217-9

6. Bates, L. S., Waldren, R. P., Teare, I. D. (1973). Rapid determination of free proline for water-stress studies. *Plant and Soil*, *39*, 205–207. https://doi.org/10.1007/BF00018060
7. Egamberdieva, D., Wirth, S., Bellingrath-Kimura, S. D., Mishra, J., Arora, N. K. (2019). Salt-tolerant plant growth promoting rhizobacteria for enhancing crop productivity of saline soils. *Frontiers in Microbiology*, *10*, 2791. https://doi.org/10.3389/fmicb.2019.02791
8. El-Saadony, M. T., Saad, A. M., Mohammed, D. M., Fahmy, M. A., Elesawi, I. E., Ahmed, A. E., Algopishi, U. B., Elrys, A. S., Desoky, E.-S. M., Mosa, W. F. A., Abd El-Mageed, T. A., Alhashmi, F. I., Mathew, B. T., AbuQamar, S. F., El-Tarabily, K. A. (2024). Drought-tolerant plant growth-promoting rhizobacteria alleviate drought stress and enhance soil health for sustainable agriculture: A comprehensive review. *Plant Stress*, *14*, 100632. https://doi.org/10.1016/j.stress.2024.100632
9. Gouda, S., Kerry, R. G., Das, G., Paramithiotis, S., Shin, H.-S., Patra, J. K. (2018). Revitalization of plant growth promoting rhizobacteria for sustainable development in agriculture. *Microbiological Research*, *206*, 131–140. https://doi.org/10.1016/j.micres.2017.08.016
10. Hafizh, M., Rambe, R. D. H., Asbur, Y. (2021). Pertumbuhan dan produksi tanaman bawang merah (*Allium cepa* L.) terhadap cekaman kekeringan dan dosis pupuk kendang sapi. *AGRILAND Jurnal Ilmu Pertanian*, *9*(1), 7–11.
11. Hamid, H., Yanti, Y., Haryanda, F. (2024). Application effect of rhizobacteria Bacillus strain on plant growth and pest insects in shallot plantation (*Allium ascalonicum* L.). *IOP Conference Series: Earth and Environmental Science*, *1346*(1), 12023. https://doi.org/10.1088/1755-1315/1346/1/012023
12. Hau, W. D., Kerihi, E. C. G., Manu, C. M. A., & Bunyani, N. A. (2025). Pengaruh pemberian plant growth promoting rhizobacteria (PGPR) dengan dosis berbeda terhadap pertumbuhan dan hasil tanaman bawang merah (*Allium cepa* L.). *HUMANITIS: Jurnal Humaniora, Sosial Dan Bisnis*, *3*(1), 305–313.
13. Jia, Z., Tang, M., Wu, J. (1999). The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chemistry*, *64*(4), 555–559. https://doi.org/10.1016/S0308-8146(98)00102-2
14. Khokhar, K. H., Demiraj, E., Lahori, A. H., Memon, M., Babar, H., Sethar, S., Panhwar, M. A., Kumar, M., Khokhar, Z. A., Afzal, A. (2025). Field experiment to evaluate the phosphate fertilizers effect on wheat morphological traits. *Trends in Ecological and Indoor Environmental Engineering*, *3*(4), 1–11. https://doi.org/10.62622/TEIEE.025.3.4.01-11
15. Larasani, I., Violita. (2021). Prolin sebagai indikator ketahanan tanaman terhadap cekaman kekeringan. *Prosiding SEMNAS BIO 2021*, 1728–1738.
16. Manurung, G. P., Kusumiyati, Hamdani, J. S. (2022). Pengaruh interval penyiraman terhadap pertumbuhan dan adaptasi tiga bawang merah komersial. *Jurnal Kultivasi*, *21*(1), 24–32. https://doi.org/10.24198/kultivasi.v21i1.34836
17. Meena, M., Divyanshu, K., Kumar, S., Swapnil, P., Zehra, A., Shukla, V., Yadav, M., Upadhyay, R. S. (2019). Regulation of L-proline biosynthesis, signal transduction, transport, accumulation and its vital role in plants during variable environmental conditions. *Heliyon*, *5*(12), e02952. https://doi.org/10.1016/j.heliyon.2019.e02952
18. Munns, R., Gilliham, M. (2015). Salinity tolerance of crops: What is the cost? *New Phytologist*, *208*(3), 668–673. https://doi.org/10.1111/nph.13519
19. Pandey, P., Irulappan, V., Bagavathiannan, M. V, Senthil-Kumar, M. (2017). Impact of combined abiotic and biotic stresses on plant growth and avenues for crop improvement by exploiting physio-morphological traits. *Frontiers in Plant Science*, *8*, 537. https://doi.org/10.3389/fpls.2017.00537
20. Patah, S. F. A., Othman, N. M. I. (2024). Influence of soil temperature on root development and microbial diversity in paddy fields: A comprehensive review. *Trends in Ecological and Indoor Environmental Engineering*, *2*(4), 1–9. https://doi.org/10.62622/TEIEE.024.2.4.01-09
21. Paul, D., Lade, H. (2014). Plant-growth-promoting rhizobacteria to improve crop growth in saline soils: A review. *Agronomy for Sustainable Development*, *34*(4), 737–752. https://doi.org/10.1007/s13593-014-0233-6
22. Pejic, B., Gvozdanovic-Varga, J., Milic, S., Ignjatovic-Cupina, A., Krstic, D., Cupina, B. (2011). Effect of irrigation schedules on yield and water use of onion (*Allium cepa* L.). *African Journal of Biotechnology*, *10*(14), 2644–2652. https://doi.org/10.5897/AJB10.1059
23. Pratiwi, A., Maghfoer, M. D., Widaryanto, E., Aini, N. (2024). Effects of different timings of drought stress and plant growth-promoting rhizobacteria inoculation on the photosynthetic characteristics of shallot (*Allium ascalonicum* L.). *Journal of Ecological Engineering*, *25*(5), 230–243. https://doi.org/10.12911/22998993/186357
24. Rahmandhias, D. T., Karyawati, A. S., Hariyono, D.,

Maghfoer, M. D. (2024). Effect of plant growth-promoting rhizobacteria (PGPR) on growth and yield of shallots on saline soils. *Journal of Degraded and Mining Lands Management*, *11*(4), 6461–6469. https://doi.org/10.15243/jdmlm.2024.114.6461

25. Rubin, R. L., van Groenigen, K. J., Hungate, B. A. (2017). Plant growth promoting rhizobacteria are more effective under drought: A meta-analysis. *Plant and Soil*, *416*(1–2), 309–323. https://doi.org/10.1007/s11104-017-3199-8
26. Saryanah, N. A., Bidara, I. S., H., R., S., H., Irawati, A. F. C., Sulastri, S., Sukmadi, R. B., Roswanjaya, Y. P., Asiani, N. (2024). Alleviating drought stress on Allium cepa mediated by drought-tolerant plant growth-promoting bacteria. *IOP Conference Series: Earth and Environmental Science*, *1359*(1), 12092. https://doi.org/10.1088/1755-1315/1359/1/012092
27. Shrivastava, P., Kumar, R. (2015). Soil salinity: A serious environmental issue and plant growth promoting bacteria as one of the tools for its alleviation. *Saudi Journal of Biological Sciences*, *22*(2), 123–131. https://doi.org/10.1016/j.sjbs.2014.12.001
28. Sikder, A., Mumit, S. H. (2024). Investigation of the effectiveness of using luffa cylindrica to reduce water consumption in agriculture. *Trends in Ecological and Indoor Environmental Engineering*, *2*(4), 18–27. https://doi.org/10.62622/TEIEE.024.2.4.18-27
29. Swasono, F. D. H. (2012). Karakteristik fisiologi toleransi tanaman bawang merah terhadap cekaman kekeringan di tanah pasir pantai. *Jurnal AgriSains*, *3*(4), 88–102.
30. Unzilaturrohmah, L., Triani, N., Hidayat, R. (2025). Peran pupuk KCl terhadap pertumbuhan dan hasil tanaman bawang merah (*Allium ascalonicum* L.) varietas tajuk pada cekaman kekeringan. *BIOFARM: Jurnal Ilmiah Pertanian*, *21*(1), 162–169. https://doi.org/10.31941/biofarm.v21i1.6406
31. Wuyep, S. Z., Ogbole, A. S., James, W. (2026). Precipitation effectiveness and rice yield: Implications for agroecosystem resilience in Langtang South, Nigeria. *Trends in Ecological and Indoor Environmental Engineering*, *4*(2), 35–43. https://doi.org/10.62622/TEIEE.026.4.2.35-43