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PLANT GROWTH-PROMOTING RHIZOBACTERIA INOCULATION MITIGATES DROUGHT STRESS AND ENHANCES GROWTH AND NUTRIENT UPTAKE IN WHEAT

Abstract: Drought is one of the most critical environmental constraints limiting wheat productivity worldwide, especially under organic and low-input agricultural systems where synthetic fertilizers are restricted. Microbial inoculants, particularly plant growth-promoting rhizobacteria (PGPR), offer a sustainable strategy for enhancing plant tolerance to water deficit by improving physiological performance and nutrient acquisition. In this study, germinated seeds of the wheat cultivar ‘Rakhshan’ were inoculated with eight PGPR strains – *Azotobacter chroococcum* (Azto478), *A. salinestrus* (Azto474), *Azospirillum lipoferum* (A443), *Pseudomonas* sp. (P196 and P241), *Pseudomonas fluorescens* (P187), *Pseudomonas putida* (P186), and *Bacillus subtilis* (B271). The experiment was conducted under greenhouse conditions using a completely randomized factorial design. Drought stress was applied at three moisture levels corresponding to 100%, 60%, and 40% of field capacity (FC), and soil moisture was maintained throughout the 100-day growth period. Measured traits included grain, shoot, and root dry weights; plant height; spike length; grain nitrogen, phosphorus, and potassium concentrations. As expected, increasing drought severity markedly reduced all growth and nutrient parameters. However, PGPR inoculation substantially mitigated these declines, demonstrating the capacity of specific bacterial strains to counteract drought-induced physiological limitations. Among the tested inoculants, *Pseudomonas fluorescens* P187 consistently exhibited the strongest positive effects. Under moderate drought, P187 increased grain dry weight by approximately 95%, and under severe drought it enhanced shoot and root dry weights by approximately 75% compared with the uninoculated control. P187 also improved macronutrient accumulation, outperforming all other strains across nitrogen, phosphorus, and potassium contents. *Pseudomonas* sp. P196 ranked second, showing notable but comparatively lower improvements. Overall, the findings identify P187 and to a lesser extent P196 as highly promising PGPR candidates for developing microbial biofertilizers aimed at enhancing drought resilience and nutrient use efficiency in wheat. These strains warrant further evaluation under field conditions and in multi-strain or consortium-based formulations to maximize their agronomic potential.

Keywords: biofertilizers, drought, inoculation, stress mitigation, water deficit, wheat.

Introduction

Drought stress is one of the most critical limiting factors affecting plant growth and crop productivity worldwide (Kuwayama et al., 2019). Iran, located between latitudes 25° and 38°N, lies within arid and semi-arid climatic zones and has recently experienced a significant decline in precipitation along with an increased frequency of drought events. As a substantial portion of the country’s water resources is allocated to agriculture, this sector is the most severely impacted by drought (Yazdandoost, 2016; Yousefi et al., 2024).

A promising and environmentally sustainable strategy for mitigating drought stress in agriculture

is the application of microbial inoculants containing plant growth-promoting rhizobacteria (PGPR) (Ali & Glick, 2025). Representative genera of PGPR include *Azotobacter*, *Pseudomonas*, *Bacillus*, and *Azospirillum* (Shah et al., 2021). These bacteria enhance plant drought tolerance primarily by promoting root development through phytohormone production, facilitating biological nitrogen (N₂) fixation, mediating iron uptake via siderophores, and solubilizing insoluble phosphate compounds in the soil. Additional mechanisms include the production of exopolysaccharides (EPS) and the enzyme ACC deaminase, both of which contribute to drought stress alleviation (Ali & Glick, 2025; Khosravi, 2019; Khosravi & Mahmoudi, 2013; Khosravi et al., 2024; Shirinbayan et al., 2019).

Wheat bread is a major source of caloric intake in Iranian households and plays a fundamental role in the national diet (Abdi et al., 2016). Currently, approximately 58% of Iran’s cultivated land is dedicated to wheat production (Center for Statistics, Information Technology, and Communications, 2024). Given wheat’s strategic importance, extensive cultivation area, and the increasing occurrence of drought and water scarcity, the adoption of biotechnology-based approaches such as microbial inoculants is essential for promoting sustainable agricultural development.

This study aimed to evaluate the effects of inoculating wheat with selected native PGPR strains under different soil moisture regimes and drought stress levels. The ultimate objective is to develop effective microbial formulations that enhance wheat resilience to drought stress and improve crop performance under water-limited conditions.

Material and methods

Bacterial selection and inoculum preparation

In this study, the bacterial strains *Azotobacter chroococcum* Azto478, *Azotobacter salinestris* Azto474, *Azospirillum lipoferum* A443, *Bacillus subtilis* B271, *Pseudomonas* sp. P241, *Pseudomonas* sp. P196, *Pseudomonas putida* P186, and *Pseudomonas fluorescens* P187 were used to prepare the inoculants for the greenhouse experiments. These strains were selected from a collection of 63 isolates maintained in the microbial collection of the Soil and Water Research Institute (SWRI) (<https://gcm.wdcm.org/cc?wdcmnumber=891>). They were originally isolated from cultivated soils across Iran and had previously been screened for plant growth-promoting traits. The plant growth-promoting characteristics of the selected bacterial strains used in this study are summarized in Table 1 (Khosravi et al., 2025).

Table 1
Characteristics of the selected plant growth-promoting bacteria (Khosravi et al., 2025)

Bacterial strain	Indole-3-acetic acid (IAA)	Inorganic phosphorus solubilization	Organic phosphorus solubilization	Potassium release from muscovite	Potassium release from biotite	Exopolysaccharide production	Insoluble iron compounds solubilization	Siderophore production
			$\mu\text{g}\cdot\text{mL}^{-1}$			$\text{g}\cdot\text{L}^{-1}$	halo/colony diameter ratio	
<i>Azospirillum lipoferum</i> A443	24.90	34.20	40.35	16.67	29.59	0.00	1.90	1.42
<i>Azotobacter chroococcum</i> Azto478	27.89	27.06	17.27	5.33	7.33	4.79	0.00	0.00
<i>Azotobacter salinestris</i> Azto474	40.67	11.16	17.35	5.37	5.92	0.70	0.00	0.00
<i>Pseudomonas putida</i> P186	8.00	274.18	46.51	22.33	15.86	5.21	3.55	2.33
<i>Pseudomonas fluorescens</i> P187	43.90	334.31	74.78	14.31	13.79	5.38	3.77	1.92
<i>Pseudomonas</i> sp. P241	44.93	214.15	48.55	14.38	16.45	5.92	3.88	1.66

To prepare the inoculate, a loopful of each purified single colony grown on agar plates was transferred into a 100 mL Erlenmeyer flask containing the appropriate selective medium. The flasks were incubated at 30°C for 72 hours on a Rotary Shaker Incubator (AG CH, Infors, Switzerland). The following media were used for bacterial cultivation: Winogradsky medium for *Azotobacter*, RC medium for *Azospirillum*, *Bacillus* agar medium for *Bacillus*, and King’s B medium for *Pseudomonas* (Himedia, India).

Soil and water testing used for greenhouse cultivation

The soil samples used in this study were collected from the top 20 cm of a field at the Central Research Station of the SWRI (Karaj, Iran). Soil and water analyses were subsequently performed at the institute’s Chemistry, Physics, and Fertilizer Technology Laboratory to determine the physicochemical properties of the samples. Total nitrogen (N) content was measured using the Kjeldahl method, available potassium (K) was extracted using 1N am-

monium acetate, and available phosphorus (P_2O_5) was determined using the Olsen method (Sparks et al., 2020). Micronutrients (Zn, Fe, Cu, and Mn) were extracted using the DTPA method and quantified by atomic absorption spectrophotometry (Novaspec II, Pharmacia, Sweden). Electrical conductivity (EC) was determined using a conductivity meter (4510, Jenway, UK), while pH was measured with a pH meter (691, Metrohm, Switzerland). Organic carbon content in the soil saturation extract was determined using the Walkley–Black method (Standard volumetric titration method) (Sparks et al., 2020). Soil calcium carbonate equivalent was determined using the Total Neutralizing Value (TNV) method based on acid neutralization, following standard soil chemical procedures (Sparks et al., 2020). Soil texture components – sand, silt, and clay – were determined using the Bouyoucos hydrometer method, and the soil texture class was identified using the soil texture triangle. Field capacity (FC) was measured using the pressure plate method at 0.3 bar, and the permanent wilting point (PWP) was determined at 15 bars using the same technique (Soil moisture Equipment Corp) (Dane & Topp, 2020).

Irrigation water samples were collected following standard hydrochemical sampling procedures in pre-cleaned, acid-washed polyethylene bottles to avoid contamination. Samples were immediately transported to the laboratory and stored at 4°C prior to analysis to preserve their chemical integrity.

Soluble cations (Na^+ , Ca^{2+} , Mg^{2+} , and K^+) were determined after filtration of samples through 0.45 μm membrane filters. Sodium and potassium concentrations were measured using flame photometry (PFP7, Jenway, Germany), while calcium and magnesium were determined by atomic absorption spectrophotometry (Novaspec II, Pharmacia, Sweden), following standard methods used in soil and irrigation water studies (Sparks et al., 2020).

Soluble anions (Cl^- , SO_4^{2-} , and NO_3^-) were analyzed using standard colorimetric and titrimetric methods, in accordance with established soil science laboratory procedures (Sparks et al., 2020).

The sodium adsorption ratio (SAR) was calculated based on soluble cation concentrations expressed in $meq L^{-1}$ using the following equation: $SAR = Na^+ / \sqrt{((Ca^{2+} + Mg^{2+}) / 2)}$ (Sparks et al., 2020).

Plastic pots measuring 23 cm in height and 29 cm in top diameter, each equipped with drainage holes, were used in the experiment. Each pot was filled with 10 kg of soil sieved through a 5 mm mesh. Based on the soil analysis results, nitrogen deficiency was cor-

rected through split applications at three key growth stages: one week after planting, during stem elongation, and at heading.

Experimental design

The experiment was conducted in the greenhouse of the Central Research Station of the SWRI, Karaj, Iran, using a factorial arrangement based on a completely randomized design (CRD) with three replications. Treatments included two factors: drought stress and bacterial inoculation.

Drought stress was applied at three levels:

- D0 (non-stress): soil moisture maintained at 100–80% of field capacity (FC), corresponding to irrigation at 20% moisture depletion;
- D1 (moderate stress): soil moisture maintained at 60% of FC, corresponding to 40% depletion;
- D2 (severe stress): soil moisture maintained at 40% of FC, corresponding to 60% depletion.

The inoculation factor consisted of nine levels: a non-inoculated control (B0) and inoculation with the bacterial strains A443, B271, Azto474, Azto478, P186, P187, P196, and P241. Inoculation was performed at sowing by applying 1 mL of bacterial suspension to each seed, with a concentration of approximately 1.5×10^7 cells mL^{-1} .

Planting, growth conditions, and harvesting

Four pre-germinated seeds of the wheat cultivar ‘Rakhshan’, provided by the Seed and Plant Improvement Institute (Karaj, Iran) were sown in each pot. One week after sowing, seedlings were thinned to maintain two plants per pot. The greenhouse was kept under a 12-hour photo period, with temperatures ranging from 20 to 30°C, for 100 consecutive days.

At the end of the experiment, the plants were harvested, and the following traits were measured: grain dry weight, shoot dry weight, root dry weight, plant height, spike length, and the percentage concentrations of nitrogen (N), phosphorus (P), and potassium (K) in dry grains. To determine grain N, P, and K contents, oven-dried grain samples were ground and digested using an H_2SO_4 – H_2O_2 acid mixture. Total nitrogen was quantified using the Kjeldahl method, phosphorus was measured colorimetrically with a spectrophotometer, and potassium was determined by flame photometry (Sparks et al., 2020). All nutrient concentrations were expressed as percentages (%).

Data was analyzed using DSAASAT.XLS software (Version 2010.5.04). A one-way ANOVA was used to determine treatment effects, and mean comparisons were performed using the least significant difference (LSD) test at the 5% significance level.

Results and discussion

Soil and water test results. The physical and chemical properties of the soil used for greenhouse cultivation are presented in Table 2.

The characteristics of the irrigation water are shown in Table 3.

Effect of inoculation treatments and drought stress on growth plant indices. The results of the analysis of variance (ANOVA) for the effects of different treatments and drought stress on wheat growth indices are presented in Table 4.

Comparison of mean values showed that under non-stress conditions (D0), all bacterial treatments significantly increased grain dry weight compared to the uninoculated control (B0), with the highest value observed for inoculation with strain P187. Under moderate drought stress (D1), strain P187 again exhibited the highest grain dry weight, showing a 95% increase relative to the control. However, under severe drought stress (D2), inoculation did not result in a significant enhancement of grain dry weight compared to the control (Figure 1).

Table 2

Physical and chemical properties of the soil

Texture	Clay	Sand	Silt	Organic C (OC)	Total Neutraliz- ing Value (T.N.V.)	FC	PWP	pH	EC	N	K	P ₂ O ₅	Zn ²⁺	Cu ²⁺	Fe ²⁺	Mn ²⁺
				%					dS·m ⁻¹	%			mg·kg ⁻¹			
Sandy Clay Loam	28	46	26	0.65	14.5	18.25	11.93	7.78	1.44	0.065	285	12.6	1.21	1.21	1.8	5.46

Table 3

Chemical properties of irrigation water

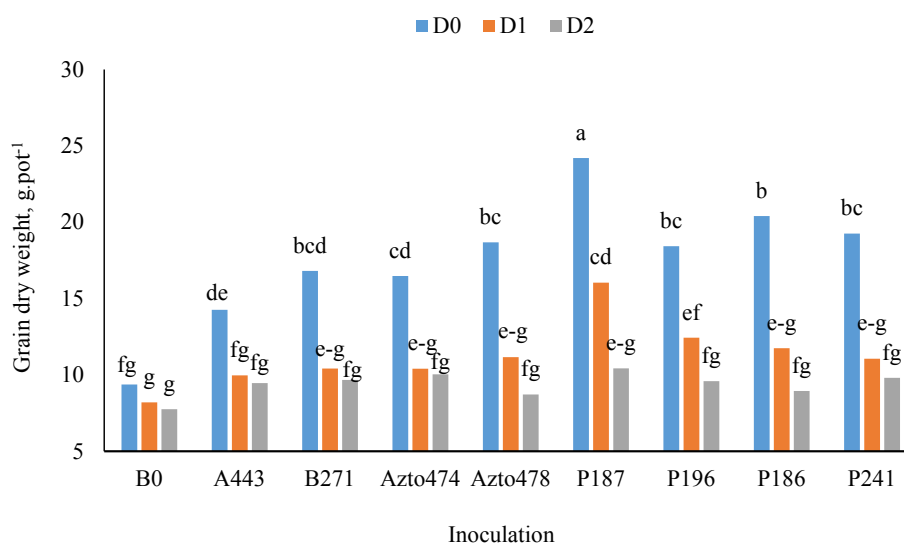
EC, dS·m ⁻¹	pH	SAR, meq·L ⁻¹	K ⁺ , mmol·L ⁻¹	Na ⁺ , mmol·L ⁻¹	Ca ²⁺ , mmol·L ⁻¹	Mg ²⁺ , mmol·L ⁻¹	Ca ²⁺ / Mg ²⁺ , mmol·L ⁻¹	Cl ⁻ , mmol·L ⁻¹	SO ₄ ²⁺ , mmol·L ⁻¹	NO ₃ ⁺ , mmol·L ⁻¹
0.42	7.92	1.8	0.015	1.87	0.84	0.24	3.54	0.17	0.69	0.31

Table 4

Variance analysis of inoculation and drought stress on various growth indices (mean square)

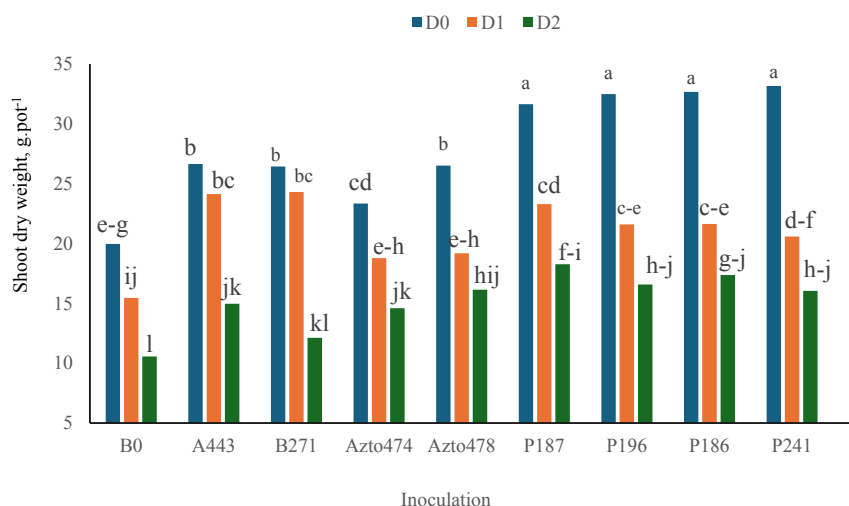
S.O.V.	df	Grain dry weight, g/ pot	Shoot dry weight, g/ pot	Root dry weight, g/ pot	Plant height, cm	Spike length, cm	Nitrogen content, %	Phosphorous content, %	Potassium content, %
Inoculation	8	45.25**	72.20**	10.70**	13.13**	10.43**	3.20**	0.0014**	0.07**
Drought stress	2	492.91**	1130.12**	32.11**	156.94**	37.28**	11.89**	0.0142**	0.09**
Inoculation * Drought stress	16	10.91**	14.27**	1.70**	2.43**	0.53**	0.33*	0.0006**	0.01**
Error	54	4.12	3.10	0.40	1.11	0.29	0.00019	0.00	0.001
CV, %		15.98	8.24	7.80	2.42	7.11	15.78	3.82	3.42

Note: * significant at $p < 0.05$; ** significant at $p < 0.01$

Figure 1*Effect of inoculation and drought stress on grain dry weight**(D0: no stress, D1: 60% of field capacity, D2: 40% of field capacity)*

With increasing drought severity, shoot dry weight decreased significantly in most treatments. Under D2 conditions, shoot dry weight was reduced by 47% compared with D0. Inoculation with all bacterial strains under D0 and D1 significantly increased

shoot dry weight relative to the uninoculated control (B0). Under D2 conditions, all strains except B271 enhanced shoot dry weight, with the highest value recorded for strain P187, which resulted in a 75% increase compared with the control (Figure 2).

Figure 2*Effect of inoculation and drought stress on shoot dry weight**(D0: no stress, D1: 60% of field capacity, D2: 40% of field capacity)*

An increase in drought stress was associated with a reduction in root dry weight. Under severe stress (D2), the lowest root dry weight was ob-

served in the B0 treatment. Under both D1 and D2 conditions, all inoculated treatments significantly increased root dry weight relative to the control

(B0), with strains P187 and P196 showing the most pronounced effects. Under D1, P187 increased root weight by approximately 75% compared to the control (Figure 3)

Plant height decreased across all treatments with increasing drought stress. In the uninoculated con-

trol (B0), plant height under D2 was reduced by 17% compared to D0. Inoculation under D2 had a significant effect, as all treated groups showed considerable increases in plant height relative to the control, with strains Azto474 and P187 producing the tallest plants (Figure 4).

Figure 3

Effect of inoculation and drought stress on root dry weight

(D0: no stress, D1: 60% of field capacity, D2: 40% of field capacity)

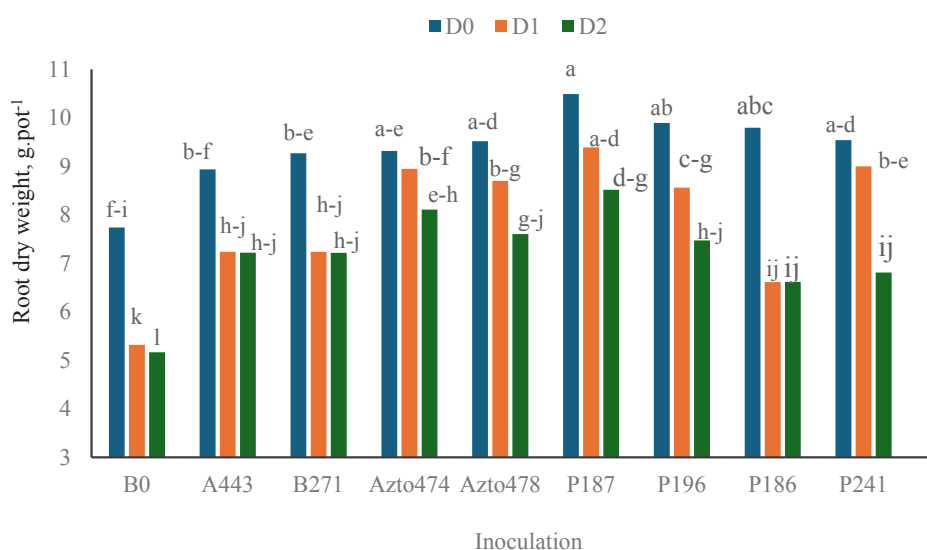
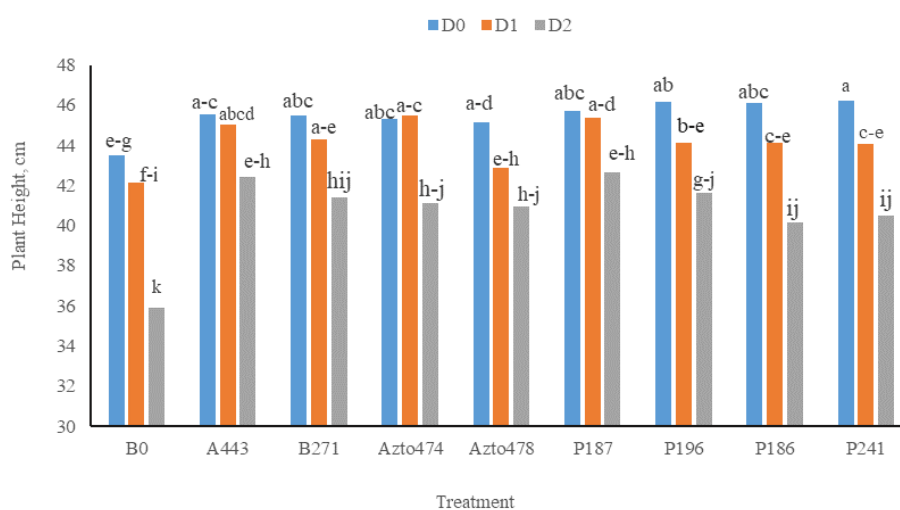


Figure 4

Effect of inoculation and drought stress on plant height

(D0: no stress, D1: 60% of field capacity, D2: 40% of field capacity)



Under D1 conditions, the most significant increases in spike length were observed for strains P196 and P187. However, under D2, inoculation did not significantly affect spike length (Figure 5).

Mean comparisons revealed that drought stress led to a reduction in grain nitrogen content. Under

moderate stress (D1), only inoculation with strain P187 resulted in a significant increase in nitrogen content compared to the control. Under severe stress (D2), strains P187, P196, and B271 significantly increased nitrogen content, with P187 showing the highest value (Figure 6).

Figure 5

Effect of inoculation and drought stress on spike length

(D0: no stress, D1: 60% of field capacity, D2: 40% of field capacity)

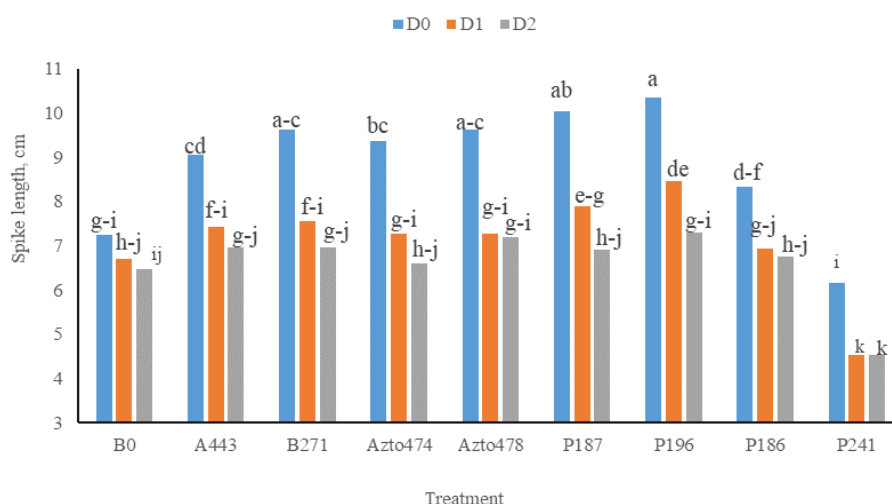
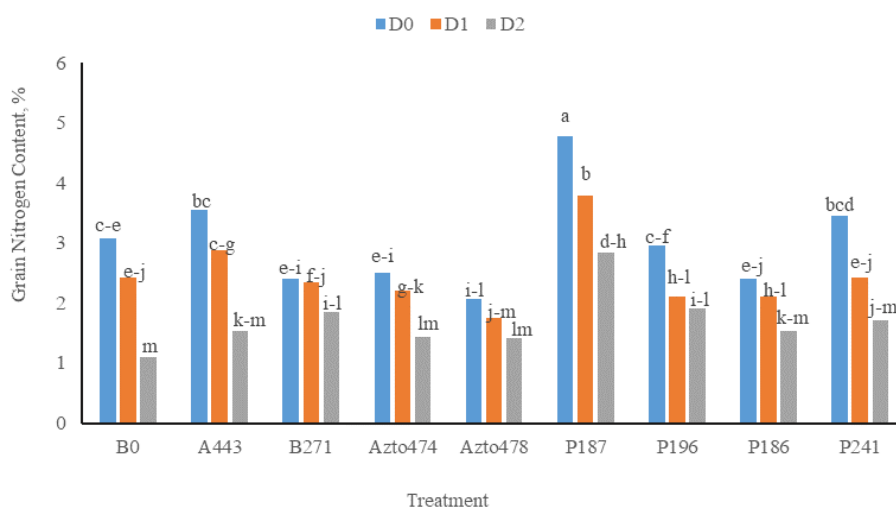


Figure 6

Effect of inoculation and drought stress on nitrogen content in plants

(D0: no stress, D1: 60% of field capacity, D2: 40% of field capacity)



Analysis of mean values indicated that drought stress reduced grain phosphorus content. Under D1, only strain P187 caused a significant increase in phosphorus levels. Under D2, all treatments except P196 significantly increased phosphorus content compared to the control, with strain P187 showing the greatest enhancement (Figure 7).

Drought stress also led to a reduction in grain potassium content. Under D1, inoculation with strains P196, P187, and P186 significantly increased potassium levels compared to the control. Under D2, strains P196 and P187 significantly elevated potassium content relative to the control (Figure 8).

Figure 7

*Effect of inoculation and drought stress on phosphorous content in plants
(D0: no stress, D1: 60% of field capacity, D2: 40% of field capacity)*

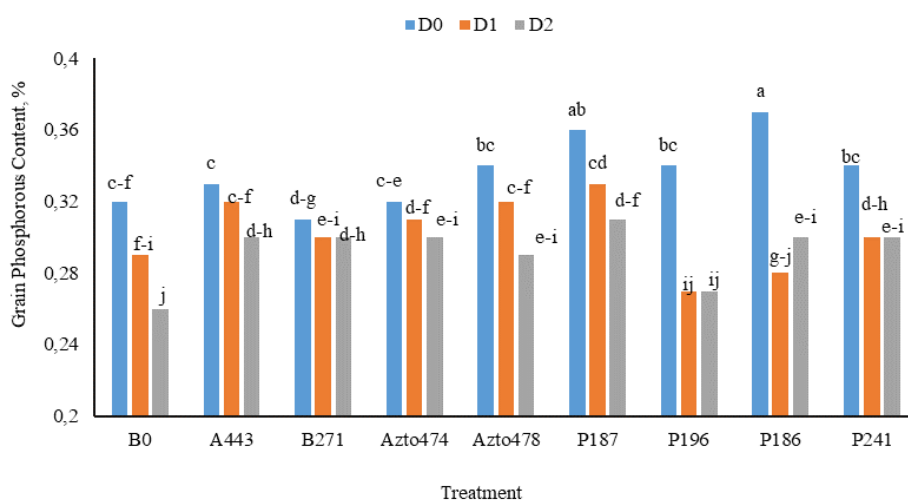
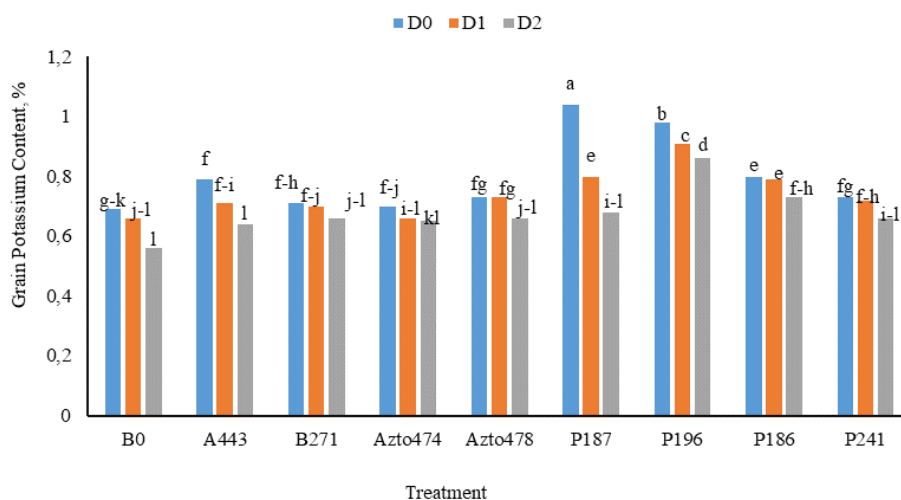


Figure 8

*Effect of inoculation and drought stress on potassium content in plants
(D0: no stress, D1: 60% of field capacity, D2: 40% of field capacity)*



The results demonstrated that drought stress, at both moderate (D1) and severe (D2) levels, significantly reduced wheat growth indices and the concentrations of nitrogen (N), phosphorus (P), and potassium (K) in grains. However, inoculation with selected bacterial strains mitigated these adverse effects by enhancing several growth parameters and nutrient concentrations under drought conditions (Figures 1-8).

Among the tested strains, *Pseudomonas fluorescens* P187 led to a significant increase in grain dry weight under moderate drought stress (D1) compared to the uninoculated control (Figure 1). This finding is consistent with previous studies reporting that inoculation with plant growth-promoting rhizobacteria improves wheat yield under drought conditions (Creus et al., 2004). *Pseudomonas spp.* has been shown to enhance grain yield in both irrigated and water-limited environments (Chandra et al., 2019).

Shoot dry weight also increased significantly at both drought levels following inoculation with all strains, P187 showing the highest mean values (Figure 2). These observations align with earlier reports demonstrating the beneficial effects of *Pseudomonas* on straw biomass (Chandra et al., 2019) and of *Bacillus* and *Azospirillum* on shoot biomass under drought stress (Kasim et al., 2013).

Similarly, root dry weight was significantly improved under both D1 and D2 conditions across all treatments, P187 again producing the greatest increase (Figure 3). The role of *Azospirillum* in promoting root biomass under drought has been well-documented. Enhanced root development under stress is particularly important for improving water and nutrient uptake, thereby supporting overall plant growth (Agami et al., 2014).

Drought stress led to a reduction of plant height, but inoculation significantly mitigated this effect, especially under severe drought (D2) (Figure 4). This result is consistent with prior findings indicating that PGPR can enhance plant height under stress conditions (Kasim et al., 2013).

Although spike length declined with increasing drought severity, significant improvement due to inoculation was observed only under moderate stress (D1), with strains P187 and P196 producing the longest spikes (Figure 5). These findings agree with earlier studies showing that PGPR can enhance spike length under water-deficient conditions (Chandra et al., 2019). Additionally, *Azospirillum lipoferum* was previously reported to increase spike length under 60% evapotranspiration (ETc) relative to 100% ETc (Agami et al., 2014).

Drought stress also decreased grain macronutrient concentrations, but inoculation partially offset this reduction (Figures 6-8). Under moderate drought (D1), only strain P187 significantly improved nitrogen and phosphorus levels, and it showed the strongest effect under severe stress (D2) as well. A similar trend was noted for potassium content (Figure 8). These outcomes are supported by prior research indicating that PGPR can improve N, P, and K concentrations in grains under drought stress (Chandra et al., 2019). Likewise, inoculation with *Azotobacter* has been shown to enhance macronutrient uptake in maize grown under drought conditions in greenhouse trials (Shirinbayan et al., 2019).

PGPR enhance plant growth through several mechanisms, including the production of phytohormones (e.g., auxins), biological nitrogen fixation, solubilization of insoluble phosphates, siderophore release, silicate decomposition, and mobilization of essential nutrients such as potassium (Khosravi et al., 2024). The bacterial strains used in the present study were previously screened and characterized based on these functional traits (Khosravi et al., 2025).

Previous research has also demonstrated that bacterial inoculation can mitigate drought-induced damage and increase wheat stress tolerance via both direct mechanisms (e.g., hormone production) and indirect mechanisms (e.g., improved nutrient availability) (Kasim et al., 2013).

Interestingly, although *Azotobacter* strains (Azto478 and Azto474) and *Azospirillum* (A443) are known nitrogen (N₂) fixers, they did not significantly influence grain nitrogen content. In contrast, *Pseudomonas fluorescens* P187, which lacks nitrogen-fixing ability, significantly enhanced nitrogen content under all moisture conditions (D0, D1, and D2) compared to the control (Figure 6). This suggests that mechanisms other than nitrogen fixation – likely involving plant growth-promoting activities such as enhanced root development – are responsible for the observed improvements in nitrogen uptake. Laboratory analyses have confirmed that P187 exhibits superior traits, including indole-3-acetic acid (IAA) production, phosphate solubilization, and exopolysaccharide secretion (Khosravi et al., 2025). Similar enhancements were observed for phosphorus and potassium concentrations, further underscoring the effectiveness of this strain (Figures 7 and 8). These results are consistent with other reports highlighting the central role of microbial growth-promoting traits in improving root and shoot development and, subsequently, nutrient accumulation in plants (Ahmed & Hasnain, 2014; Alaskar & Al-Shwaiman, 2023; Çakmakçı et

al., 2020; Grover et al., 2021; Pantoja-Guerra et al., 2023).

Conclusion

This study provides clear evidence that inoculating wheat with selected PGPR strains, including *Azotobacter*, *Azospirillum*, *Bacillus*, and *Pseudomonas*, can significantly enhance plant performance under drought stress, both in terms of growth attributes and macronutrient acquisition. Among all evaluated strains, *Pseudomonas fluorescens* P187 consistently produced the strongest positive responses across multiple growth and nutrient parameters, indicating substantial drought-mitigating potential. The performance of *Pseudomonas sp.* P196, although slightly lower, further supports the superior adaptability and functional versatility of *Pseudomonas* species in water-limited environments. These results collectively highlight the value of exploring multi-strain or consortium-based inoculation strategies in future research, as such formulations may broaden the functional spectrum and yield more stable improvements under variable field conditions.

A particularly noteworthy finding is the marked increase in grain nitrogen content in plants inoculated with P187, despite the strain's lack of nitrogen-fixing ability. This suggests that indirect mechanisms such as stimulation of root system architecture, enhancement of rhizosphere nutrient mobilization, or modulation of hormone-mediated nutrient uptake pathways may play a central role in improving nitrogen assimilation under drought. In contrast, classical nitrogen-fixing PGPR such as *Azotobacter* and *Azospirillum* did not produce comparable effects, indicating that nitrogen fixation alone is insufficient to guarantee improved N accumulation when plants are exposed to moisture stress.

Overall, these observations emphasize the need to adopt a mechanism-oriented and trait-based per-

spective when selecting bacterial inoculants for wheat drought tolerance. Considering attributes such as phytohormone production, siderophore release, osmotic-adjustment facilitation, and nutrient-mobilization capacity will be essential for developing reliable PGPR-based bioformulations aimed at strengthening crop resilience and maintaining yield stability under increasingly frequent drought events.

Author contributions

Houshang Khosravi: Conceptualization, Investigation, Data Curation, Writing-Original Draft Preparation, Writing-Review and Editing, Visualization, Supervision, and Project Administration

Akram Otadi: Conceptualization, Investigation, Data Curation, and Writing-Original Draft Preparation

Hassan Etesami: Conceptualization, Visualization, and Supervision

Hossein Ali Alikhani: Supervision and Visualization

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Conflict of interest

The authors declare that they have no conflicts of interest.

References

1. Abdi, F., Atarodi, Z., Mirmiran, P., & Esteki, T. (2016). Review of nutritional status in Iranian population. *Focus on Sciences*, 2(3), 1–4. <https://doi.org/10.20286/focsci-020332>
2. Agami, R. A., Ghramh, H. A., & Hasheem, M. (2017). Seed inoculation with *Azospirillum lipoferum* alleviates the adverse effects of drought stress on wheat plants. *Journal of Applied Botany and Food Quality*, 90(1), 165–173.
3. Ahmed, A., & Hasnain, S. (2014). Auxins as one of the factors of plant growth improvement by plant growth promoting rhizobacteria. *Polish Journal of Microbiology*, 63(3), 261–266. <https://doi.org/10.33073/pjm-2014-035>
4. Alaskar, A. A., & Al-Shwaiman, H. A. (2023). The effect of plant growth-promoting rhizobacteria on soil properties and the physiological and anatomical characteristics of wheat under water-deficit stress conditions. *Agriculture*, 13(11), 2042. <https://doi.org/10.3390/agriculture13112042>

5. Ali, S., & Glick, B. (2025). Bacterial alleviation of drought stress in plants: recent advances and future challenges. In H. Etesami & Y. Chen (Eds.), *Sustainable Agriculture under Drought Stress* (pp. 367-383). Academic Press. <https://doi.org/10.1016/B978-0-443-23956-4.00022-3>
6. Çakmakçı, R., Mosher, G., Milton, A. H., Alatürk, F., & Ali, B. (2020). The effect of auxin and auxin-producing bacteria on the growth, essential oil yield, and composition in medicinal and aromatic plants. *Current Microbiology*, 77(4), 564–577. <https://doi.org/10.1007/s00284-020-01917-4>
7. Center for Statistics, Information Technology, and Communications. (2024). *Agricultural statistics* (Vol. 1). Ministry of Agricultural-Jahad. (In Persian)
8. Chandra, D., Srivastava, R., Gupta, V. V., Franco, C. M., Paasricha, N., Saifi, S. K., et al. (2019). Field performance of bacterial inoculants to alleviate water stress effects in wheat (*Triticum aestivum* L.). *Plant and Soil*, 441, 261–281. <https://doi.org/10.1007/s11104-019-04115-9>
9. Creus, C. M., Sueldo, R. J., & Barassi, C. A. (2004). Water relations and yield in *Azospirillum*-inoculated wheat exposed to drought in the field. *Canadian Journal of Botany*, 82(2), 273–281. <https://doi.org/10.1139/b03-119>
10. Dane, J. H., & Topp, C. G. (Eds.). (2020). *Methods of soil analysis: Part 4. Physical methods*. John Wiley & Sons.
11. Grover, M., Bodhankar, S., Sharma, A., Sharma, P., Singh, J., & Nain, L. (2021). PGPR mediated alterations in root traits: way toward sustainable crop production. *Frontiers in Sustainable Food Systems*, 4, 618230. <https://doi.org/10.3389/fsufs.2020.618230>
12. Kasim, W. A., Osman, M. E., Omar, M. N., Abd El-Daim, I. A., Bejai, S., & Meijer, J. (2013). Control of drought stress in wheat using plant-growth-promoting bacteria. *Journal of Plant Growth Regulation*, 32, 122–130. <https://doi.org/10.1007/s00344-012-9283-7>
13. Khosravi, H. (2019). Response of maize to inoculation with *Azotobacter* under drought stress conditions. *Journal of Water Research in Agriculture*, 33(1), 29–38. (In Persian) <https://doi.org/10.22092/jwra.2019.119112>
14. Khosravi, H., Khoshru, B., Nosratabad, A. F., & Mitra, D. (2024). Exploring the landscape of biofertilizers containing plant growth-promoting rhizobacteria in Iran: progress and research prospects. *Current Research in Microbial Sciences*, 6, 100268. <https://doi.org/10.1016/j.crmicr.2024.100268>
15. Khosravi, H., Mahmoudi, M. (2013). Evaluation of effects of *Azotobacter* inoculation and manure on growth of rainfed wheat. *Journal of Soil Management and Sustainable Production*, 3(2), 205–219. (In Persian).
16. Khosravi, H., Otadi, A., Alikhani, H., & Etesami, H. (2025). Evaluation and comparative analysis of plant growth-promoting traits in diverse groups of rhizosphere bacteria. *Journal of Soil Biology*, 12(2), 235–260. <https://doi.org/10.22092/sbj.2025.367286.271>
17. Kuwayama, Y., Thompson, A., Bernknopf, R., Zaitchik, B., & Vail, P. (2019). Estimating the impact of drought on agriculture using the US Drought Monitor. *American Journal of Agricultural Economics*, 101(1), 193–210. <https://doi.org/10.1093/ajae/aay037>
18. Pantoja-Guerra, M., Valero-Valero, N., & Ramírez, C. A. (2023). Total auxin level in the soil–plant system as a modulating factor for the effectiveness of PGPR inocula: a review. *Chemical and Biological Technologies in Agriculture*, 10(1), 6. <https://doi.org/10.1186/s40538-022-00370-8>
19. Shah, A., Nazari, M., Antar, M., Msimbira, L. A., Naamala, J., Lyu, D., & Smith, D. L. (2021). PGPR in agriculture: a sustainable approach to increasing climate change resilience. *Frontiers in Sustainable Food Systems*, 5, 667546. <https://doi.org/10.3389/fsufs.2021.667546>
20. Shirinbayan, S., Khosravi, H., & Malakouti, M. J. (2019). Alleviation of drought stress in maize (*Zea mays*) by inoculation with *Azotobacter* strains isolated from semi-arid regions. *Applied Soil Ecology*, 133, 138–145. <https://doi.org/10.1016/j.apsoil.2018.09.015>
21. Sparks, D. L., Page, A. L., Helmke, P. A., & Loeppert, R. H. (Eds.). (2020). *Methods of soil analysis: Part 3. Chemical methods*. John Wiley & Sons.
22. Yazdandoost, F. (2016). Dams, drought and water shortage in today's Iran. *Iranian Studies*, 49(6), 1017–1028. <https://doi.org/10.1080/00210862.2016.1241626>
23. Yousefi, H., Ahani, A., Moridi, A., & Razavi, S. (2024). The future of droughts in Iran according to CMIP6 projections. *Hydrological Sciences Journal*, 69(7), 951–970. <https://doi.org/10.1080/02626667.2024.2348720>

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