ACS OMEGA

Open Access

http://pubs.acs.org/journal/acsodf

Article

Salinity Stress Resilience in *Sorghum bicolor* through *Pseudomonas*-Mediated Modulation of Growth, Antioxidant System, and Eco-Physiological Adaptations

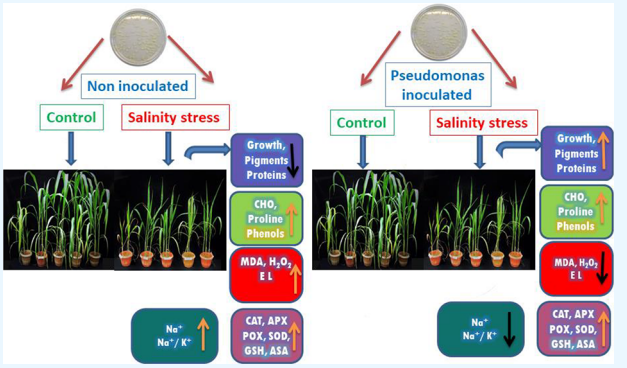
Muhammad Azeem,* Robina Sultana, Naeem Ahmed, Muhammad Waseem Abbasi, Khwaja Ali Hasan, Renjie Dong, Saud Alamri, and Alanoud T. Alfagham

Cite This: *ACS Omega* 2025, 10, 940−954

Read Online

ACCESS | Metrics & More | Article Recommendations

ABSTRACT: Increased soluble salts in soil and irrigation water threaten the sustainability of crops. This causes food insecurity directly by reducing the staple crop yield and indirectly by limiting fodder and forage production. Recently, plant-growth-promoting rhizosphere microorganism utilization improved crop productivity under stress. Therefore, this research was conducted to find the *Sorghum bicolor* growth improvement potential by exogenous application of five different *Pseudomonas* strains under salinity in a pot experiment. The salinity was applied with a 1/2-strength Hoagland's nutrient solution as 0 and 100 mM NaCl for 30 days. Results indicated that salinity reduced the vegetative growth parameters and stress-responsive biochemicals in nonbacterial treated plants. However, *Pseudomonas* strains applied to plants exhibited notable increases in growth, relative water content, antioxidant enzyme activities, osmolytes, and photosynthetic pigments under salinity. The ionic imbalance was also reduced due to *Pseudomonas* strains by improving K^+ and K^+/Na^+ ratios under salinity. *P. aeruginosa* strain SAHK (OQ194056) and *P. putida* strain AHK_SHA007 (OR468335) were found to be promising compared to other strains in increasing growth and stress tolerance. The augmentation of the plant's antioxidant system and maintenance of ion homeostasis by *Pseudomonas* strains served as a strategy to enhance the plant salt tolerance.



1. INTRODUCTION

Stress-resilient crop cultivation in semiarid and arid regions is the prerequisite goal for farmers and researchers to provide food for increasing populations in the prevailing global climate change situation.[1] Climate change is accelerating the intensity of different environmental stresses including drought, salinity, floods, heat, chilling, and heavy metal stresses, which in turn seriously affect crop productivity globally.[2,3] Moreover, soil and irrigation water-induced salinity is proving to be the most destructive environmental stress factor causing serious crop losses.[4] About 1125 million hectares (i.e., 10% of the world's arable land) is salt affected, and about 1.5 million hectares of land annually adds to this salt-affected land.[5] Increased salinity in plants imposes a variety of metabolic and physiological impairments due to osmotic, ionic, and oxidative stresses. Hyperaccumulation of salts causing specific ion toxicity along with nutritional imbalances triggers a secondary stress factor, oxidative damage, due to the surplus generation of active oxygen species.[6,7]

Sorghum bicolor, a multipurpose C4 plant, holds the fifth position in global agricultural importance and is notably the second most crucial crop in Africa.[8] Throughout various regions in Asia and Africa, sorghum cultivation is prioritized for its diverse uses, serving as a vital resource for ensuring food security, animal feed, and bioethanol production.[9] While sorghum exhibits a commendable tolerance to salinity at moderate levels, higher salt concentrations present a significant obstacle, particularly impeding seed germination and obstructing seedling establishment,[10] hence significantly reducing the growth and yield.[11,12] Considering the importance and requirement of sorghum, its production needs to be improved on saline lands via an agriculturally sustainable method.

Various sustainable techniques are in consideration to stimulate salinity tolerance in plants including exogenous

Received: September 3, 2024
Revised: November 30, 2024
Accepted: December 3, 2024
Published: January 5, 2025

Table 1. Detail of the Exogenous *Pseudomonas* Treatments Applied to *Sorghum* with Isolate Name, Species Name with Strain Name, and Accession Number[a]

treatments	isolate name	species name	strain name	accession number	CFU·mL^{-1}
NT					
B1		P. monteilii	SHA_AHK	OR468769	9×10^{9}
B2	MRFP-206	*P. aeruginosa*	PS3	MN850314	15.7×10^{10}
B3	MRFP-207	*P. aeruginosa*	ZSR2	OQ220350	14.8×10^{10}
B4	EPP-161	*P. aeruginosa*	SAHK	OQ194056	15.6×10^{10}
B5	EFP-56	*P. putida*	AHK_SHA007	OR468335	16.9×10^{10}

[a]The amounts of different *Pseudomonas* strains applied are given in colony-forming units (CFU) per milliliter.

application of growth regulators, including secondary metabolites and phytohormones.[13] The current trend involves a growing interest in the development of transgenic lines, although this approach is not considered helpful in solving crop production challenges.[14] Therefore, researchers are focusing toward the solution that is more sustainable, cost-effective, and eco-friendly to increase crop productivity.[15] The growth-promoting potential exhibited by root colonizing rhizobacteria is extensively documented in scientific literature and proves helpful to protect crops and improve agricultural productivity by improving soil fertility under unfavorable environmental conditions.[16] Plant-growth-promoting rhizobacteria (PGPR) help to improve the plant productivity through diverse mechanisms, involving facilitation of nutrient availability and the synthesis of phytohormones required for growth. PGPR also demonstrated the stress tolerance capacity by modulating several plants' physiological processes that confer protection.[17,18] Considering this, the exogenous application of the PGPR to *Sorghum* is helpful to increase the salt tolerance and the productivity, emphasizing the vital importance of research in this domain.

Some of the PGPRs like *Pseudomonas, Rhizobium, Bacillus, Azospirillum,* and *Enterobacter* contribute to mitigate the stresses in plants.[19−21] The utilization of these bacteria with growth-promoting properties represents an environmentally sustainable strategy, as they can establish colonization in the plant's rhizosphere. This colonization, in turn, induces salt tolerance by upholding nutritional equilibrium and fostering plant growth and development.[22] Furthermore, numerous reports on exogenous application of PGPR bacteria including *Bacillus* and *Pseudomonas* species showed increased salt tolerance.[23,24] The *Pseudomonas* species exogenous application has attained widespread attention due to its proficiency in root surface colonization, enzyme activity changes, and augmentation of metabolite production.[25] *Pseudomonas* species have the potential to produce growth-enhancing traits in plants including production of ACC deaminase activity, exopolysaccharides (EPS), siderophores, indole-acetic acid (IAA), and ammonia and efficiently solubilize essential nutrients such as zinc and phosphate.[26,27] These growth-promoting properties could improve plants' physiological and nutrient status and make *Pseudomonas* species a potential bioinoculant to promote growth and productivity under abiotic stress conditions. The studies on applying *Pseudomonas* species to plants under drought conditions improved the tolerance of jujube[28] and wheat.[26] Application of *Pseudomonas* species has also shown to improve the salt tolerance in *Arabidopsis thaliana*,[29] red pepper,[30] tomato,[31] soybean, and corn.[32] Considering this evidence, bioinoculation of *Pseudomonas* could be regarded as an environment-friendly and sustainable alternative method to overcome the problem of salinity and also an alternative to harmful chemical fertilizers.[33,34] Studies conducted on improving *Sorghum* growth by applying *Pseudomonas* strains are limited. Recently, the halotolerant *P. stutzeri* ISE12 has shown effectiveness in reducing salt stress in *Sorghum*, suggesting that other *Pseudomonas* strains should also be evaluated.[35] Given these, the current investigation is undertaken to evaluate the potential of various *Pseudomonas* strains to improve salt tolerance and their physiological mechanism in the *Sorghum* plant.

2. MATERIALS AND METHODS

2.1. Bacterial Strains. Five different *Pseudomonas* isolates were collected from the Department of Botany, University of Karachi culture collection. These isolates were preidentified at morphological and biochemical basis at the genus level, and their species was confirmed through molecular identification.

2.2. Molecular Identification of *Pseudomonas* Species and Phylogenetic Relationship. The genomic DNA extraction was carried out via a miniprep kit (Bio Basic, Canada) as per vendor instructions. The quality of genomic DNA extracted was confirmed by running on 1% agarose gel electrophoresis. The 16S rDNA region was amplified by using a genus-specific primer set as a molecular barcode for identification. The polymerase chain reaction mixture comprising 50 ng of extracted bacterial DNA, 1 μL of forward and reverse primer (10 μM), and 25 μL of 2X DreamTaq Green PCR was accomplished at a 50 μL final volume, using a Bio-Rad S1000 thermal cycler following, as described earlier.[36] The amplicons were subjected to sequencing PCR followed by sequence elucidation through the CEQ 8000 Genetic Analysis System (Beckman Coulter). The 16S rDNA sequences were used to establish a phylogenetic relationship among the *Pseudomonas* species used in this study via the MEGA software. The 16S rDNA sequences of the *Pseudomonas* strains were submitted to the NCBI GenBank by the OR468769, MN850314, OQ220350, OQ194056, and OR468335 accession numbers.

2.3. Experimental Setup in Greenhouse. Selected *Pseudomonas* strains were exogenously applied on *Sorghum bicolor* plants exposed to two different NaCl treatments (0 and 100 mM) under greenhouse conditions. Initially, surface-disinfected seeds were divided into six sets: five sets were exogenously applied microbial suspensions for 10 min and one nontreated disinfected set of seeds (NT; without microbial application). Ten treated seeds of respective bacterial and nonbacterial treatments were sown in respective pots filled with quartz sand (2 kg). These pots containing treated seeds were provided with 1/2-strength Hoagland's solution (prepared in sterilized distilled water) through the subirrigation method. At 15 days of germination, five equal-sized plants were maintained in each pot. Bacterial suspension (50 mL) was

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

applied in each pot (with almost the same CFU/mL) as for the soil drenching method. Three days after bacterial inoculation, salt concentrations (0 and 100 mM NaCl) mixed in half-strength nutrient solution (Hoagland's) were applied to the plants. These plants were allowed to grow in a greenhouse and after 30 days of salinity treatment carefully harvested for growth, biochemical, and physiological analyses. The details of the *Pseudomonas* treatments with isolate number, species name, strain name, accession number, and amount of bacteria given for treatments are given in Table 1.

2.4. Evaluation of Growth Parameters. The growth parameters of *Sorghum* plants treated with different *Pseudomonas* strains under nonsalinity and salinity conditions were evaluated after 30 days of salinity treatment. The plants of various treatments were uprooted under running tap water to avoid damage to the roots. These harvested plants were placed on a blotting paper to remove excess water. The shoot and root lengths, number of leaves, stem thickness, intermodal distance, and shoot and root fresh weights were immediately measured. The stem thickness was measured with the help of a vernier caliper. The shoots and roots of plants from different treatments were separated and dried in an oven at 65 °C for 48 h for dry weights.

2.5. Estimation of Relative Water Contents (RWC) and Electrolyte Leakage (EL). The RWC of leaves were estimated by following the methodology given by Weatherly.[37] Fresh leaf disc (0.6 mm) samples of each *Pseudomonas* strain-treated and nontreated were immediately weighed for fresh weight and then transferred in Petri dishes having distilled water at 4 °C. After 8 h, leaf disc samples were transferred to blotting paper to remove excess water and weighed for turgid weight. These leaf disc samples were dried by transferring to an incubator at 60 °C for 24 h. Later, the dry weights of the leaf disc samples were measured. The RWC from the above measured weights were calculated according to the following formula: RWC (%) = ((fresh weight − dry weight)/(turgid weight − dry weight)) × 100. The percent EL in leaf tissues was estimated by the Dionisio-Sese and Tobita method.[38] Briefly, the 20 discs of 6 mm diameter were made from the fresh leaves. These leaf discs were transferred in a test tube containing 20 mL of distilled water for 2 h, and then the electrical conductivity (EC_1) value was measured. After measuring EC_1 values, the test tubes were transferred in an autoclave at 121 C for 30 min to disrupt all the membranes. This autoclaved solution was used to measure the EC_2 values. The EC_1 and EC_2 values were measured on an electrical conductivity meter (Jenway 4510). The EL (%) from EC_1 and EC_2 values was calculated by the following equation:

$$\mathrm{EL}(\%) = \mathrm{EC}_{1(\text{initial reading})}/\mathrm{EC}_{2(\text{reading after 30 min. of boiling})} \times 100 \quad (1)$$

2.6. Estimation of Photosynthetic Pigments. Leaf samples harvested from plants of different treatments were cleaned, macerated in acetone (80%), and centrifuged, and the absorbance of supernatants was noted at 646 and 663 nm for chlorophylls, and 470 nm for the carotenoids.[39] The absorbance values were used to calculate the chlorophylls and carotenoids in mg·g^{-1} of leaf fresh weight from the following equations:

$$\text{chlorophylls a}(C_a) = \frac{(12.21A_{663} - 2.81A_{646})}{a \times 1000 \times W} \times V \quad (2)$$

$$\text{chlorophylls b}(C_b) = \frac{(20.13A_{646} - 5.03A_{663})}{a \times 1000 \times W} \times V \quad (3)$$

$$\text{total chlorophylls} = \frac{(C_a + C_b)}{a \times 1000 \times W} \times V \quad (4)$$

$$\text{carotenoids}(C_{x+c}) = \frac{(1000A_{470} - 3.27C_a - 104C_b)}{a \times 1000 \times W} \times V \quad (5)$$

where V = volume of leaf extract, W = leaf fresh weight, a = path of a light

2.7. Estimation of Soluble Carbohydrates, Total Phenols, and Proline. Hot water extracts of dry plant material were prepared for soluble carbohydrates and proline. The dry sample powder (100 mg) was dissolved in 10 mL of distilled water. This extract was boiled in a water bath at 100 °C for 1 h. Then, this hot water extract was cooled at room temperature and centrifuged at 4000 × *g* for 10 min. The supernatant collected was used to measure soluble carbohydrates and proline concentration by the method described by Yemm and Willis[40] and Bates et al.,[41] respectively. For total phenols, the leaf dry material (100 mg) was homogenized in 10 mL of 80% methanol. After 12 h of shaking, this mixture was centrifuged. The supernatant was separated and used for measuring phenolic contents according to Singleton and Rossi.[42]

2.8. Estimation of Ascorbic Acid (ASA) and Reduced Glutathione (GSH). The fresh leaf samples were frozen in liquid nitrogen and later homogenized in a 3% trichloroacetic acid (TCA) solution and vortexes and centrifuged at 12,000 rpm for 15 min at 4 °C. The supernatant was separated and used to determine the concentrations of ASA and GSH according to the method described by Guri.[43] Briefly, 1 mL of supernatant was mixed with 1 mL of DCPIP (2,6-dichlorophenolindophenol) and 0.5 mL of distilled water and incubated for 15 min. The absorbance at 600 nm was recorded for the ASA concentration determination. The GSH was analyzed in a reaction mixture composed of 0.5 mL of distilled water, 0.5 mL of supernatant, 0.5 mL of 0.2 M sodium phosphate buffer (pH 7.0), and 0.1 mL of 5,5-dithiobis(2-nitrobenzoic acid). The reaction mixture was incubated for 30 min, and absorbance was recorded at 412 nm.

2.9. Estimation of Protein and Antioxidant Enzyme Activities. For the protein concentration and antioxidant enzyme activities, fresh *Sorghum* leaf samples (0.5 g) each of respective NT and *Pseudomonas* strain treatments were frozen immediately in liquid nitrogen. These frozen samples of different treatments were stored at −20 °C and at the time of analysis were homogenized in an extraction buffer made by 50 mM potassium phosphate (pH 7.5) and supplemented with 1 mM ascorbic acid, 5 mM disodium EDTA, and 2% PVP. The homogenate was allowed to centrifuge at 17,000 rpm for 20 min in a refrigerated centrifuge machine. The supernatant of each extract was collected and transferred in five Eppendorf tubes separately for activities of proteins and four antioxidant enzymes. The protein concentration of these extracts was determined by the Bradford assay reagent method.[44] The absorbance of the protein assay reaction was recorded at 590 nm through a UV/vis spectrophotometer (Jenway 6305).

This protein extract was also used for the antioxidant enzyme assay. For the CAT assay, 3 mL of reaction mixture contained 50 mM potassium phosphate buffer (pH 7.0), 10

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

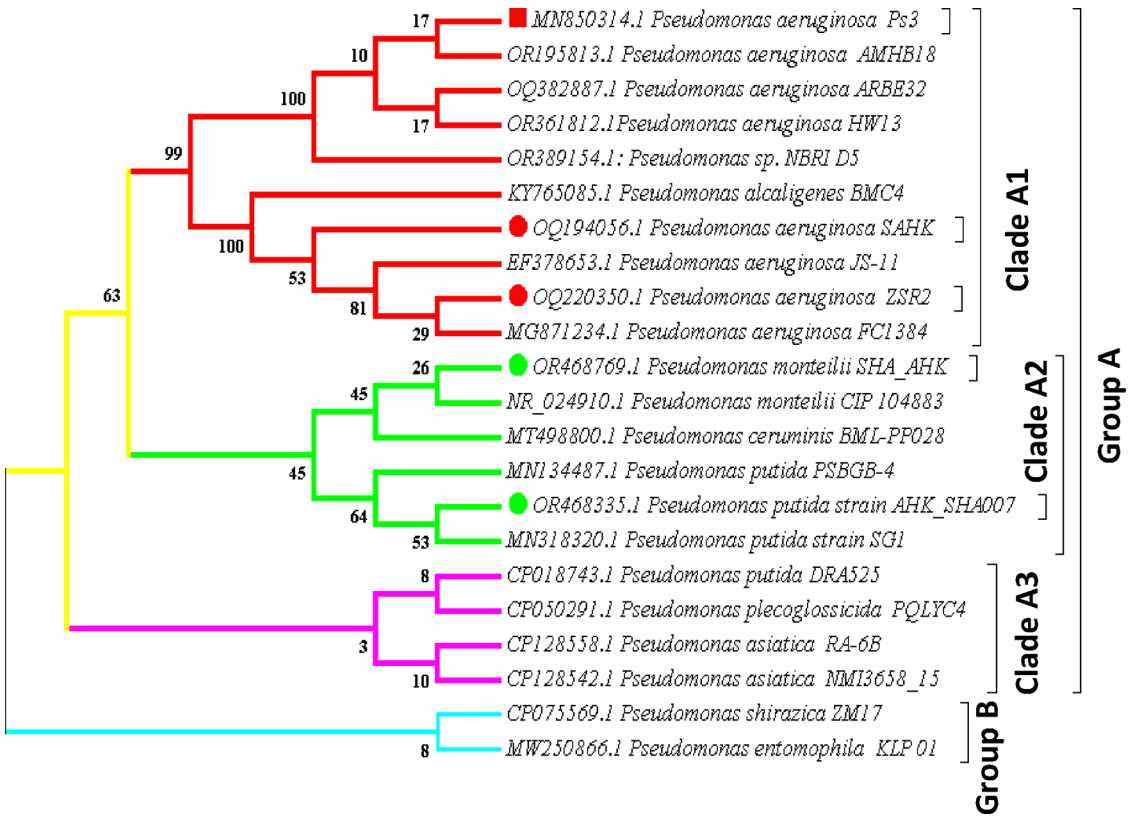


Figure 1. Evolutionary relationships of the different *Pseudomonas* strains. The studied *Pseudomonas* strains were highlighted with red and green markers on the left side and with brackets on the right side. The numbers shown next to the branches represent the percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates).

mM H_2O_2, and 100 μL of enzyme extract. The linear decrease in absorbance of the reaction mixture was recorded immediately after the addition of enzyme extract at 240 nm for 1 min.[45] For the APX assay, 3 mL of reaction mixture contained 50 mM potassium phosphate buffer (pH 7.0), 0.55 mM ascorbate, 0.1 mM H_2O_2, and 50 μL of enzyme extract. The decrease in the absorbance of the reaction mixture was recorded at 290 nm for 1 min at 25 °C.[46] For the POX assay, 3 mL of reaction mixture contained 100 mM potassium phosphate buffer (pH 7.0), 20 mM guaiacol, 10 mM H_2O_2, and 50 μL of enzyme extract. The increase in absorbance of the reaction mixture was measured at 470 nm for 1 min.[47] For the SOD assay, 3 mL of reaction mixture contained 100 mM potassium phosphate buffer (pH 7.8), 33 μM nitro blue tetrazolium chloride, 10 mM methionine, 0.6 mM EDTA, 0.10 μM riboflavin, and 50 μL of enzyme extract. The reaction of SOD activity was started by placing the reaction mixture containing test tubes in a fluorescent light (two 20 W bulbs) inside the box. After 7 min, reaction was stopped and absorbance was recorded at 560 nm.[48]

2.10. Estimation of Hydrogen Peroxide (H_2O_2) and Malondialdehyde (MDA) Concentration. The same 3% TCA extract used for nonenzymatic antioxidants was also used for the H_2O_2 and MDA determination. The H_2O_2 and MDA concentrations were determined by the methods of Velikova et al.[49] and Heath and Packer,[50] respectively. For H_2O_2 concentration determination, the reaction mixture consisted of 0.5 mL of TCA extract, 0.5 mL of potassium phosphate buffer (pH 7.0), and 1 mL of 1 M potassium iodide. The reaction mixture was incubated in the dark for 10 min, and the absorbance was recorded at 390 nm. For MAD concentration determination, the reaction mixture comprised 0.5 mL of 3% TCA extract and 0.5 mL of 0.5% 2-thiobarbituric acid made in 20% TCA solution. The reaction mixture was incubated in a water bath at 95 °C for 30 min. The reaction was terminated by placing test tubes in an ice bath and then centrifuged. The absorbance of the supernatant was recorded at 450, 532, and 600 nm.

2.11. Estimation of Sodium (Na^+) and Potassium (K^+) Ions. The hot water extract of shoot and root dry powder was made by boiling on a water bath, and the clear solution after filtration was obtained. The Na^+ and K^+ ions in the clear solution were determined by the method of Khan et al.[51] A Corning flame photometer (model 410) was utilized for measuring ions.

2.12. Statistical Analysis. All of the data sets of the experimental results presented as bars in the graphs are the mean ± SD of five replicates (n = 5). Statistical analyses of all the data sets were performed on R (Version 4.1.3), and the figures were generated on the ggplot2 program of RStudio. Two-way ANOVA (analysis of variance) was tested on the data sets to find the significance levels among salinity and bacterial treatments. Tukey's HSD test was employed to compare the mean of each data set ($P < 0.05$), and the significant differences were represented by different letters on each bar. Pearson correlation analysis was done for all the tested parameters, and a correlation matrix was generated to show the relationship among different variables.

3. RESULTS

3.1. Molecular Identification and Evolutionary Relationship of *Pseudomonas* Species. The isolates were

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

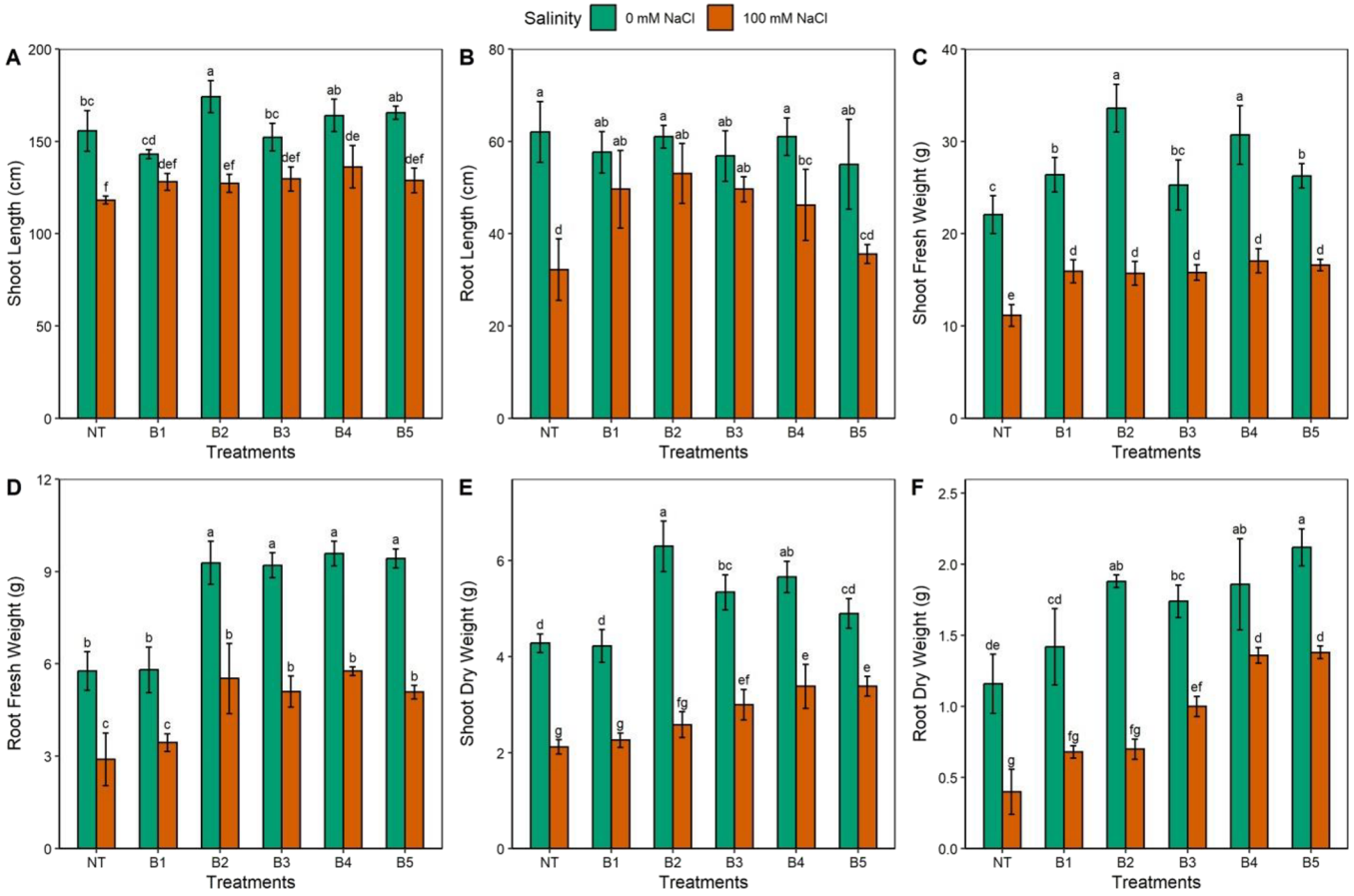


Figure 2. Effect of different *Pseudomonas* strains on the growth parameters (shoot length (A), root length (B), shoot fresh weight (C), root fresh weight (D), shoot dry weight (E), and root dry weight (F) of *Sorghum bicolor* plants grown under salt stress (0 and 100 mM NaCl). NT = nontreated, B1 = *P. monteilii* strain SHA_AHK; B2 = *P. aeruginosa* strain PS3; B3 = *P. aeruginosa* strain ZSR2; B4 = *P. aeruginosa* strain SAHK; B5 = *P. putida* strain AHK_SHA007. The bars are presented as treatment mean ± SD (n = 5). The different alphabetic letters above the bars represent the significant differences at $P < 0.05$ according to Tukey's HSD test following ANOVA to compare the effect of each treatment.

identified as *P. monteilii* strain SHA_AHK (Accession No. OR468769), MRFP-206 as *P. aeruginosa* strain PS3 (Accession No. MN850314), MRFP-207 as *P. aeruginosa* strain ZSR2 (Accession No. OQ220350), EPP-161 as *P. aeruginosa* strain SAHK (Accession No. OQ194056), and EFO-56 as *P. putida* strain AHK_SHA007 (Accession No. OR468335). The evolutionary relationship showed that the strain *P. aeruginosa* (MN850314, OQ220350, and OQ194056) belongs from Clade A1, whereas the other two strains *P. monteilii* (OR468769) and *P. putida* (OR468335) belong to Clade A2 of Group A *Pseudomonas* (Figure 1).

3.2. Growth Parameters. *Sorghum* growth (lengths of shoot and root, fresh and dry weights of shoot and root, internodal distance, stem thickness, and number of leaves) was reduced significantly ($P < 0.001$) under salinity. However, those plants inoculated with different *Pseudomonas* strains including *P. monteilii* strain SHA_AHK (B1); *P. aeruginosa* strains PS3 (B2), ZSR2 (B3), and SAHK (B4); and *P. putida* strain AHK_SHA007 (B5) significantly ($P < 0.001$) increased the growth parameters except number of leaves under nonsalinity and salinity conditions as compared to nonbacterial treatment (NT). *Pseudomonas* strain-inoculated roots showed higher fresh and dry weights except *P. monteilii* (B1) treatment under both nonsalinity and salinity conditions. In shoot, *P. aeruginosa* (B2) inoculations showed higher fresh (52%) and dry (47%) weights and performed better than other *Pseudomonas* strains under nonsalinity condition, whereas *P. aeruginosa* (B4) and *P. putida* (B5)-inoculated higher shoot fresh (53 and 48%, respectively) and dry weights (59% respectively) were noticed under salinity treatment compared to no bacteria (NT) (Figure 2).

The inoculation of *Pseudomonas* strains also significantly ($P < 0.001$) augmented the stem thickness in all inoculated plants under salinity compared to NT, and the highest increase (101%) was found with B4 treatment (Figure 3A). The intermodal distance was nonsignificantly increased in *P. aeruginosa* (B4) and *P. putida* (B5) inoculation under salt stress (Figure 3B). However, a nonsignificant increase for the number of leaves was observed due to inoculation under salinity (Figure 3C). Overall, the growth data showed that *Pseudomonas* strains enhanced the sorghum growth under salinity mainly *P. aeruginosa* (B4) and *P. putida* (B5) increased more dry weights of shoot and root and stem thickness. The leaf RWC was also decreased under salinity; however, *Pseudomonas* strain inoculation increased the RWC. Particularly, B3 increased (49%) more RWC compared to NT under salinity (Figure 3D).

3.3. Photosynthetic Pigments. The levels of chlorophylls (a, b, and total) in NT plant leaves were significantly ($P < 0.001$) decreased under salinity (Figure 4A–C). However, *Pseudomonas* strain inoculation increased the chlorophylls under salinity. Particularly, *P. aeruginosa* (B4) and *P. putida*

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

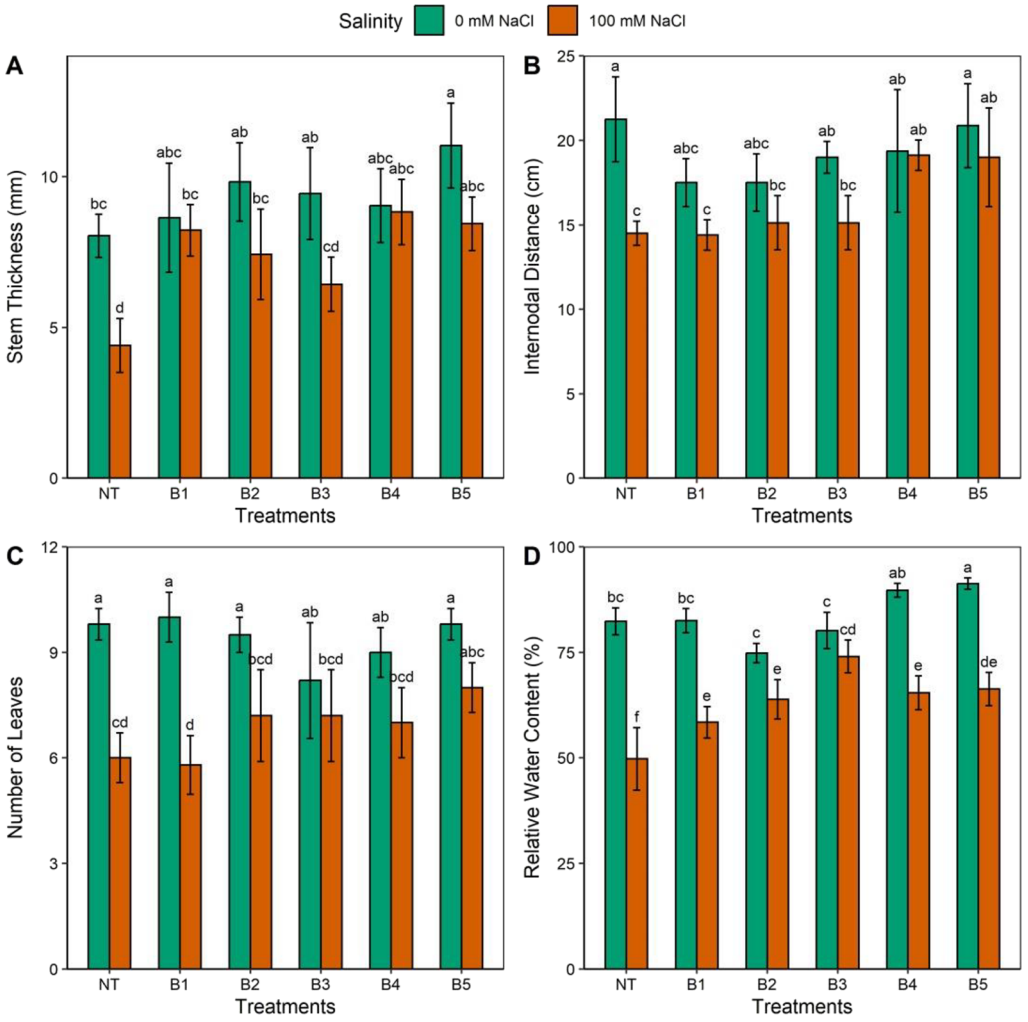


Figure 3. Effect of different *Pseudomonas* strains on the stem thickness (A), internodal distance (B), number of leaves (C), and relative water content (RWC) (D) of *Sorghum bicolor* plants grown under salt stress (0 and 100 mM NaCl). The details on the treatments are given in Figure 2. The bars are presented as treatment mean ± SD (n = 5). The different alphabetic letters above the bars represent the significant differences at P < 0.05 according to Tukey's HSD test following ANOVA to compare the effect of each treatment.

(B5) treatments increased chlorophyll levels compared to other treatments, respectively. This increase was more due to "chlorophyll a" contents, while the *P. aeruginosa* (B2) treatment increased (38%) chlorophyll b contents compared to NT under salinity. Carotenoid content had a direct relation with salt stress; therefore, the carotenoid concentration was improved significantly (P < 0.01) in *Pseudomonas* strain-treated plants under no salinity and salinity. However, the highest increase (49%) in carotenoid contents was observed in *P. aeruginosa* (B4)-treated plants compared to NT under salinity (Figure 4D).

3.4. Proteins, Soluble Sugars, Proline, and Phenols. The results showed a 101% reduction in protein contents of *Sorghum* plants under salinity. The protein contents were decreased under nonstress, while they were slightly increased under salinity due to *Pseudomonas* strain treatment (Figure 5A). The amount of soluble carbohydrate, proline, and phenolic concentration was increased significantly (P < 0.001) under salinity. *Pseudomonas* strain inoculation increased the abovementioned biochemicals more under salinity. Interestingly, all *Pseudomonas* strains increased the soluble carbohydrates levels under no salinity and salinity. The higher levels of soluble carbohydrates were noticed in *P. putida* (B5) and *P. aeruginosa* (B2) treatments (31 and 30%, respectively) compared to NT under salinity (Figure 5B). The proline and phenolic contents were slightly increased with *Pseudomonas* strains under salinity (Figure 5C,D).

3.5. Ascorbic Acid (ASA) and Reduced Glutathione (GSH). Inoculation of *Pseudomonas* strains significantly (P < 0.001) increased the ASA and GSH concentrations in *Sorghum* compared to NT under no-salinity and salinity conditions. The treatments of *P. monteilii* (B1) and *P. aeruginosa* (B4) showed higher ASA contents compared to other *Pseudomonas* strain treatments, while *P. aeruginosa* (B2) and *P. putida* (B5) treatments showed higher GSH contents (54 and 76%) compared to NT under salinity (Figure 5E,F).

3.6. Oxidative Damage Markers (EL, H_2O_2, and MDA). The oxidative stress damage markers (i.e., H_2O_2, MDA, and EL,) were significantly (P < 0.001) increased in *Sorghum* under salinity (Figure 6). However, the inoculation of *Pseudomonas* strains significantly (P < 0.001) reduced this damage under salinity. Different *Pseudomonas* strains reduced H_2O_2 levels from 35 to 48% compared to NT under salinity conditions, and *P. aeruginosa* (B3 and B4) strain inoculation showed maximum H_2O_2 reduction in salinity. In comparison, MDA was reduced from 12 to 38 due to different *Pseudomonas*

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

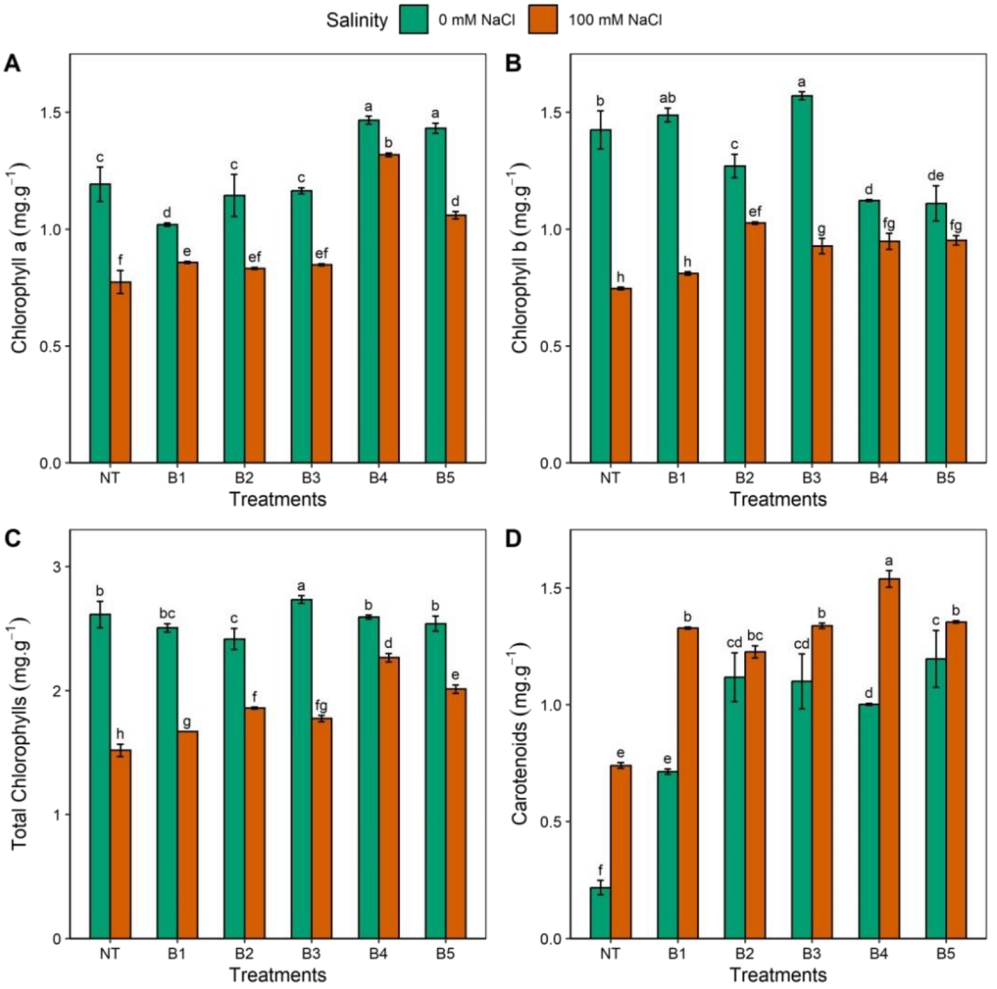


Figure 4. Effect of different *Pseudomonas* strains on photosynthetic pigments including chlorophyll a (A), chlorophyll b (B), total chlorophylls (C), and carotenoids (D) of *Sorghum bicolor* plants grown under salt stress (0 and 100 mM NaCl stress). The details on the treatments are given in Figure 2. The bars are presented as treatment mean ± SD (n = 5). The different alphabetic letters above the bars represent the significant differences at $P < 0.05$ according to Tukey's HSD test following ANOVA to compare the effect of each treatment.

strains and *P. aeruginosa* (B2) showed a maximum decrease of 38% compared to NT under salinity. All of the *Pseudomonas* strains reduced the EL from 37 to 45% under salinity.

3.7. Antioxidant Enzyme Activities. Antioxidant enzymes (CAT, APX, POX, and SOD) in *Sorghum* showed significantly ($P < 0.001$) higher activities under salinity (Figure 7). This increase was more significant with inoculation of *Pseudomonas* strains. Mainly, the *P. aeruginosa* (B2) inoculation showed higher activities of CAT, SOD (84 and 36%, respectively) as compared to NT, whereas the *P. monteilii* (B1) and *P. putida* (B5) inoculation showed higher APX and POX activity compared to other treatments under salinity, respectively.

3.8. Na^+ and K^+ Ion Concentrations. The sodium ion (Na^+) levels in both *Sorghum* shoot and root tissues were found significantly ($P < 0.001$) higher under salinity. However, the *Pseudomonas* strain treatments decreased the Na^+ levels in both tissues under salinity (Figure 8A,B). In the shoot, *Pseudomonas* strains contributed equally to decrease the Na^+ levels under salinity, whereas in root, *P. aeruginosa* (B3) inoculation performed better than other *Pseudomonas* strains and NT. The K^+ levels were decreased significantly ($P < 0.001$) in the *Sorghum* shoot under salinity. The *Pseudomonas* strain treatments increased the K^+ levels in the shoot and root under no-salinity and salinity conditions. This increase in the K^+ levels was more prominent in the root (Figure 8C,D). The Na^+/K^+ ratio in shoots was found nonsignificant under no salinity conditions but declined significantly ($P < 0.001$) with *Pseudomonas* strain treatments, whereas the root Na^+/K^+ ratio declined significantly under no-salinity and salinity conditions (Figure 8E,F). In addition, the shoot K^+/Na^+ ratio was nonsignificant under no-salinity conditions; however, it increased with *Pseudomonas* strain treatments (Figure 8G). In root, the K^+/Na^+ ratio changes were more prominent and increased with *Pseudomonas* strains under no-salinity and salinity conditions (Figure 8H). The maximum increase was found with *P. aeruginosa* (B4) inoculation (67%) under no-salinity conditions and B3 inoculation (88%) under salinity, respectively.

3.9. Correlation between Growth Parameters and Physiological Changes. *Pseudomonas* treatments to *Sorghum* plants showed that the growth parameters (such as shoot and root fresh and dry weights) were positively correlated with chlorophylls (a, b, and total), RWC, and K^+ levels, whereas they were negatively correlated with oxidative stress damage markers (MDA, H_2O_2, and EL), nonenzymatic antioxidants (ASA, GSH, and phenols), antioxidant enzyme activities (CAT, APX, GPX, and SOD), and osmotica (prolines and

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

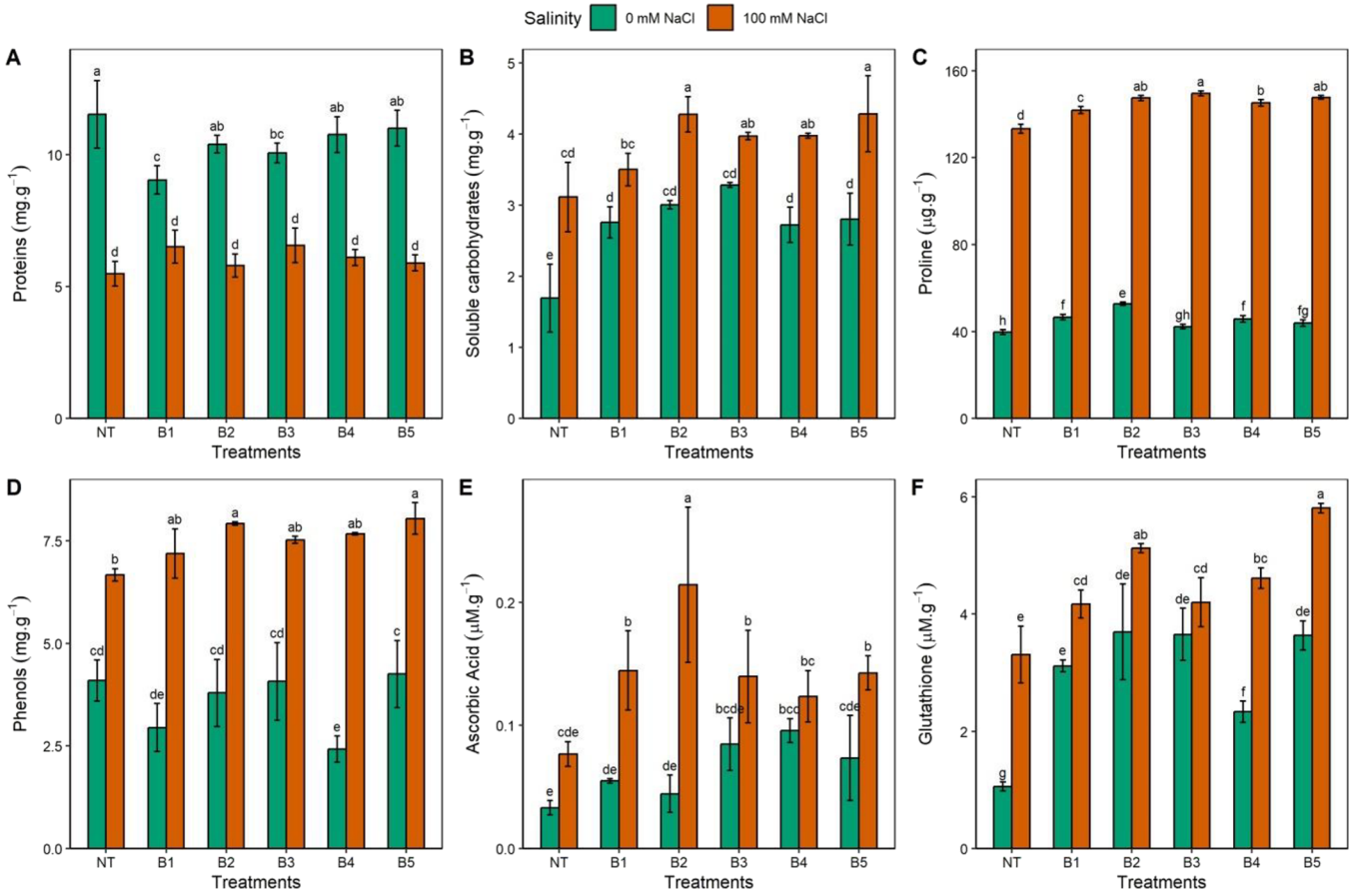


Figure 5. Effect of different *Pseudomonas* strains on osmolytes and nonenzymatic antioxidants. Proteins (A), soluble sugars (B), proline (C), phenols (D), ascorbic acid (E), and glutathione (F) of *Sorghum bicolor* plants grown under salt stress (0 and 100 mM NaCl). The details on the treatments are given in Figure 2. The bars are presented as treatment mean ± SD (n = 5). The different alphabetic letters above the bars represent the significant differences at $P < 0.05$ according to Tukey's HSD test following ANOVA to compare the effect of each treatment.

soluble carbohydrates), and Na^+ levels (Figure 9). These results showed that *Pseudomonas* treatments improved the *Sorghum* growth under salinity by increasing chlorophylls, K^+ levels, and RWC and reducing oxidative stress. The positive correlation between oxidative stress markers and the antioxidant system showed a protective role for *Pseudomonas* strains.

4. DISCUSSION

The application of plant-beneficial microbes has gained greater agronomical attention by increasing crop yield under stress.[52,53] The growth-promoting *Pseudomonas* strains were applied in this research work to promote *S. bicolor* growth under salinity. Our investigations showed that *Sorghum* growth (such as lengths and fresh and dry weights of root and shoot) was greatly reduced under salinity. This growth reduction in *Sorghum* due to salinity was reported due to salt-induced negative consequences on the physiological and anatomical modifications.[54,55] The *Sorghum* growth reduction because of salinity has been documented in earlier reports.[10,12,56] Here, the application of *Pseudomonas* strains proved their positive role by promoting *Sorghum* growth under salinity. The *Pseudomonas* species possess multiple growth-promoting roles including synthesis of plant essential phytohormones like IAA,[57] improvement in the nutrient absorption, enhanced nitrogen fixation, siderophore production,[58] and phosphorus solubilization.[59] These *Pseudomonas* beneficial roles may help to contribute in growth improvement of salt-stressed *Sorghum* plants. Among the *Pseudomonas* strains, the most promising growth enhancement was found with *P. aeruginosa* (B2, B4) and *P. putida* (B5) strains, respectively, under no-salinity and salinity conditions. *Pseudomonas* strain-inoculated *Sorghum* plants showed increased shoot and root fresh and dry weights, stem thickness, and internodal distance compared to NT *Sorghum* plants under no-salinity and salinity conditions, probably because of promotion in bacterial assisted cell division and elongation.[60] Bacterial treated plant growth improvement under salinity might happen by enhanced multiple growth-promoting traits of *Sorghum* plants such as an increase in antioxidant enzymatic activities, enhanced synthesis of osmolytes, reduced oxidative damage, reduced photosynthetic damage, and selective uptake of essential ions (particularly K^+) through roots.[56] The involvement of *Pseudomonas* in *Sorghum* physiological processes has also been linked with the improved transpiration rate and its stimulatory action on ACC-deaminase enzyme (enzyme known for ethylene inhibition), which could reduce salt-activated excess ethylene synthesis in plants.[61] By this growth-promoting action, the biomass of *Pseudomonas* strain-treated *Sorghum* plants was improved compared to NT plants. The reduction in RWC is a well-known phenomenon under saline conditions, also confirmed in our study. However, the RWC was increased due to the *Pseudomonas* strain treatment under salinity. This might happen because of *Pseudomonas* EPS

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

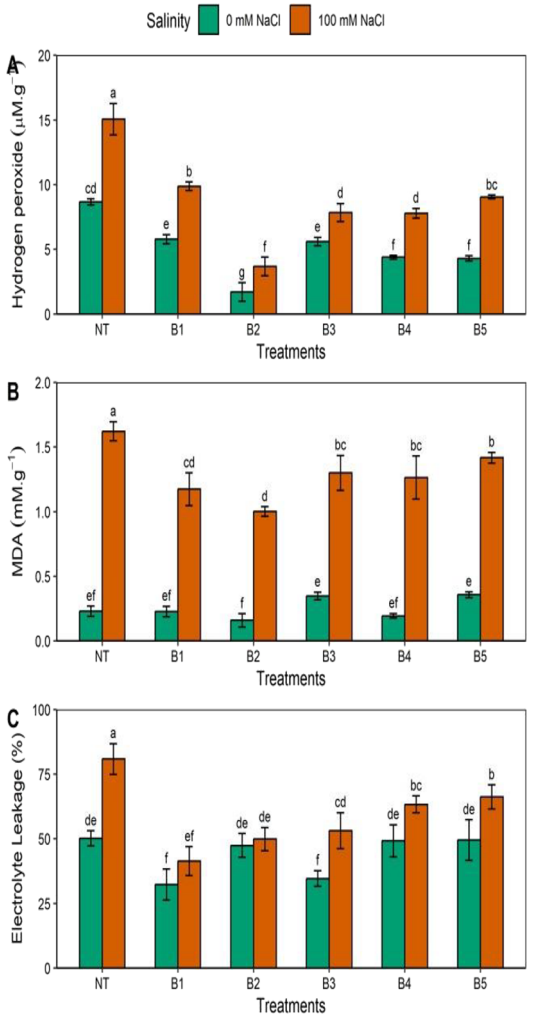


Figure 6. Effect of different *Pseudomonas* strains on oxidative stress-damaged markers. H_2O_2 (A), MDA (B), and electrolyte leakage (C) *Sorghum bicolor* plants grown under salt stress (0 and 100 mM NaCl). The details on the treatments are given in Figure 2. The bars are presented as treatment mean ± SD (n = 5). The different alphabetic letters above the bars represent the significant differences at $P < 0.05$ according to Tukey's HSD test following ANOVA to compare the effect of each treatment.

producing ability, which binds excess Na^+ with bacterial cell surfaces and improves water uptake from the roots.[62] However, the reduced RWC in NT under salt stress might be because excess salt in soil water hindered the water absorption capability of roots under reduced soil water osmotic potential and increased transpiration rate due to absorbed Na^+ in leaf tissues, which has also been recorded in alfalfa plant under 10 $dS{\cdot}m^{-1}$ salinity treatment, too.[63]

Prolonged salinity also harms plants' photosynthetic machinery because of constant reactive oxygen species (ROS) generation that triggers chlorophyll degradation.[64] The present investigation illustrated that *Pseudomonas* treatments, especially *P. aeruginosa* (B4) and *P. putida* (B5), considerably improved photosynthetic pigments (chlorophylls and carotenoids) of plants compared to NT plants under salinity. Greater pigment reduction in NT salt-stressed plants could happen due to irreplaceable alterations in food manufacturing mechanisms such as distortion of lamellar organization resulting in deformed ultrastructure of chloroplast, decreased photosynthetic pigment synthesis together with reduced PSII system performance, and reduced stomatal functions and gaseous exchange mechanism.[65,66] Similarly, our results of reduced photosynthetic pigments under salt stress follow the earlier studies performed on the sesame and tomato.[67,68] The improvement in photosynthetic pigments by *Pseudomonas* strains in our study has also been supported by previous findings on salt-stressed pepper leaves in which inoculation with *Pseudomonas* strain caused modulation in food manufacturing machinery of the plant due to enhanced chlorophyll synthesis by boosted nitrogen absorption in plants.[69]

The protein concentration was reduced in *Sorghum* plants of all treatments under saline conditions, which might be linked with reduced protein biosynthesis and enhanced protease activity as reported in comparative proteomic and gene expression analysis of chickpea plant under salinity.[70] However, in our study, only a slight augmentation in proteins was noticed with different strains of *Pseudomonas* treatment under salt stress. Exogenously applied *Pseudomonas* strain treatment boosted the synthesis of proteins due to amino acid synthesis in salt-stressed plants.[71] Another work has also documented a slight increase in protein concentration by *P. monteilii* treatment in maize seedlings under water−salt−alkali-combined stresses.[72]

The soluble carbohydrates in the *Sorghum* plant were increased under salinity; however, the *Pseudomonas* strain treatments raised the soluble carbohydrate levels. *P. aeruginosa* (B2) and *P. putida* (B5) treatments were found to be most promising under salinity. This increase in soluble carbohydrates of *Pseudomonas* strain-treated plants might be due to better plant growth, increased photosynthesis, and translocation of photosynthetic products,[73] which could be achieved by overcoming osmotic imbalance in the cytosol or by lowered ROS production;[74] these changes ultimately strengthen cell wall extensibility due to increased levels of low methyl esterified homogalacturonans, which increases the cell wall thickness as reported in *Brassica napus* applied with *P. stutzeri* ISE12 strain under saline conditions.[75] This was also in accordance to previous findings where PGPR application enhanced carbohydrate biosynthesis, which maintained the cytosolic osmotic potential and protected plants from overproduction of ROS.[76] The proline concentrations were noted to be higher in all salt-stress-exposed plants of all *Pseudomonas* strains and NT plants. The *P. aeruginosa* (B2) inoculation showed enhanced proline contents; however, the difference between different *Pseudomonas* strains and NT was not very prominent. This increase points out that PGPR application has some involvement in the activation of proline-synthesizing enzymes, which is evident from the improved proteins of microbial-treated plants. This strategy of bacteria could be considered due to improvement in turgor pressure and protection against water loss by the plants due to several environmental stress conditions including salinity.[77,78]

Excess salts in the cytosol proved destructive for plants like the enhancement in lipid peroxidation and damage to membranes because of the salt-induced ROS production.[79] Nontreated (NT) plants showed increased levels of the oxidative damage markers like H_2O_2, MDA, and EL; however, lower levels of these were noticed in *Pseudomonas* strain-treated plants under salinity. The decreased levels of these oxidative damage markers were also reported due to *P. aeruginosa* and fluorescent *Pseudomonas* isolate treatments in

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

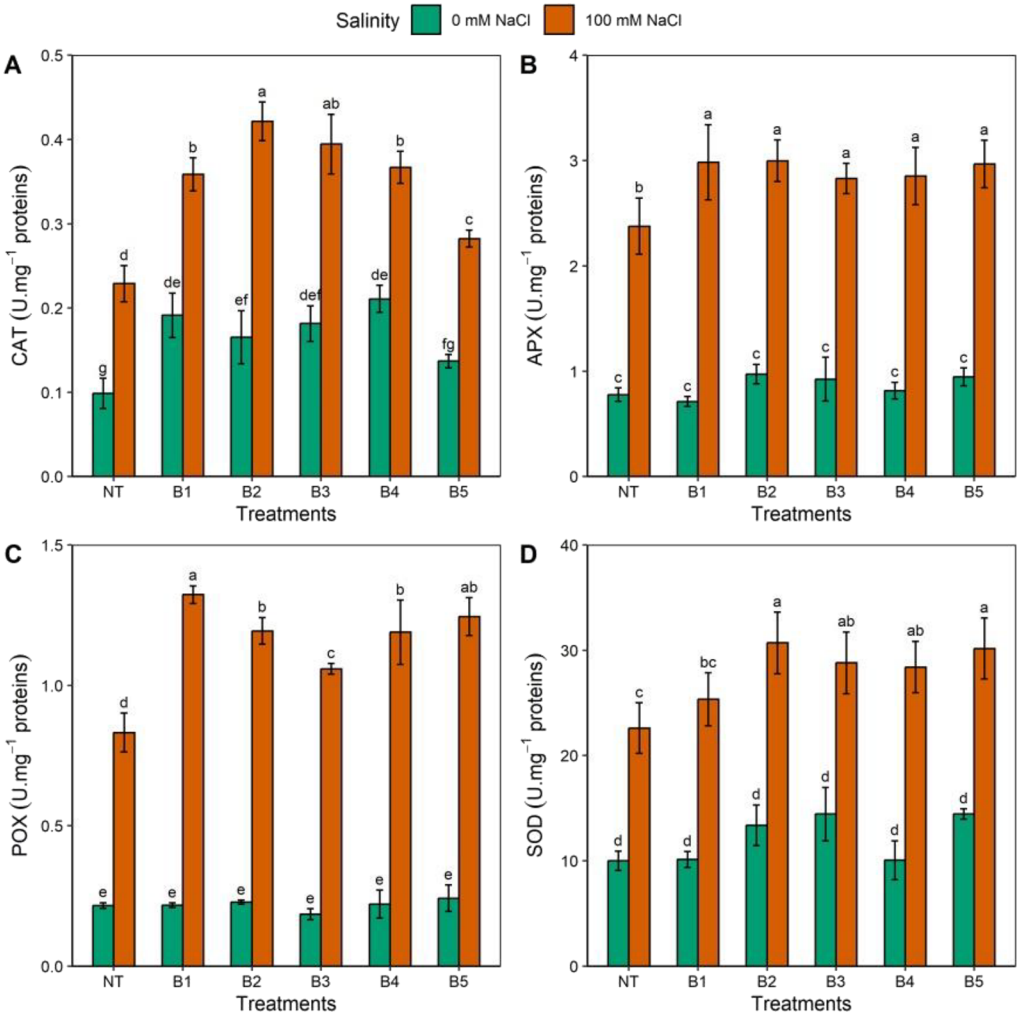


Figure 7. Effect of different *Pseudomonas* strains on the antioxidant enzymatic activities. Catalase activity, CAT (A); ascorbate peroxidase activity, APX (B); guaiacol-peroxidase activity, POX (C); and superoxide dismutase activity, SOD (D) of *Sorghum bicolor* plants grown under salt stress (0 and 100 mM NaCl). The details on the treatments are given in Figure 2. The bars are presented as treatment mean ± SD (n = 5). The different alphabetic letters above the bars represent the significant differences at $P < 0.05$ according to Tukey's HSD test following ANOVA to compare the effect of each treatment.

wheat and finger millet under zinc and salinity stresses, respectively.[80,81] The reduction in MDA and H_2O_2 due to *P. aeruginosa* under zinc stress in wheat has been observed at 34 and 17%, respectively, and the reduction in our study of both the parameters was 12 to 38% and 34 to 48% due to different *Pseudomonas* strains. Similarly, various fluorescent *Pseudomonas* isolates in finger millet reduced MDA and H_2O_2 to 65 and 37% under salinity, respectively. This reduction in oxidative damage in *Sorghum* plants due to *Pseudomonas* strains was supposed to be due to the protective role of the antioxidant system by their up-regulation (including nonenzymatic antioxidant concentrations and enzymatic antioxidant activities). Our results showed that nonenzymatic antioxidant concentrations (e.g., ascorbic acid and reduced glutathione) rapidly increased in *Pseudomonas* strain treatments and most prominently in *P. aeruginosa* (B2)- and *P. putida* (B5)-treated plants. This prominent increase might be the strategy of PGPR to scavenge salt-induced overly produced ROS.[81] Additionally, the phenolic compounds also contribute to scavenge ROS under salinity.[82] In our results, the phenolic contents were increased in all *Pseudomonas* strain-treated plants under salinity treatments compared to NT plants. This might indicate that *Pseudomonas* species enhanced the activity of phenolic compound biosynthetic enzymes, which contributed to defending the plants against ROS.[83] This increase in phenolic compounds was also proved from the study conducted on *P. putida* strain H-2−3-treated soybean plants under salinity and drought.[84] Our finding also revealed that *Pseudomonas* strains improved antioxidant enzyme activities under salinity, which also showed a positive contribution for increasing *Sorghum* salt tolerance. These enzymes have been well documented in the previous literature due to their protective role in plants against the oxidative damage and scavenging of ROS due to variety of stresses.[85]

Increased concentration of salts in irrigation water increases the absorption and retention of toxic ions (e.g., Na^+ and Cl^-) in cytosol and, consequently, decreases essential mineral (K^+, Ca^{2+}, P, and Mg^{2+} ions) uptake necessary for the normal growth and development of plants.[86] Particularly, increased Na^+ levels in the cytosol eventually disrupt the ion homeostasis by decreasing the K^+/Na^+ ratio.[87,88] This same Na^+ increase was found in untreated *Sorghum* (NT) plants. However, the *Pseudomonas* strain treatment showed decreased Na^+ levels and increased K^+ levels; thereby, the K^+/Na^+ ratio was restored

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

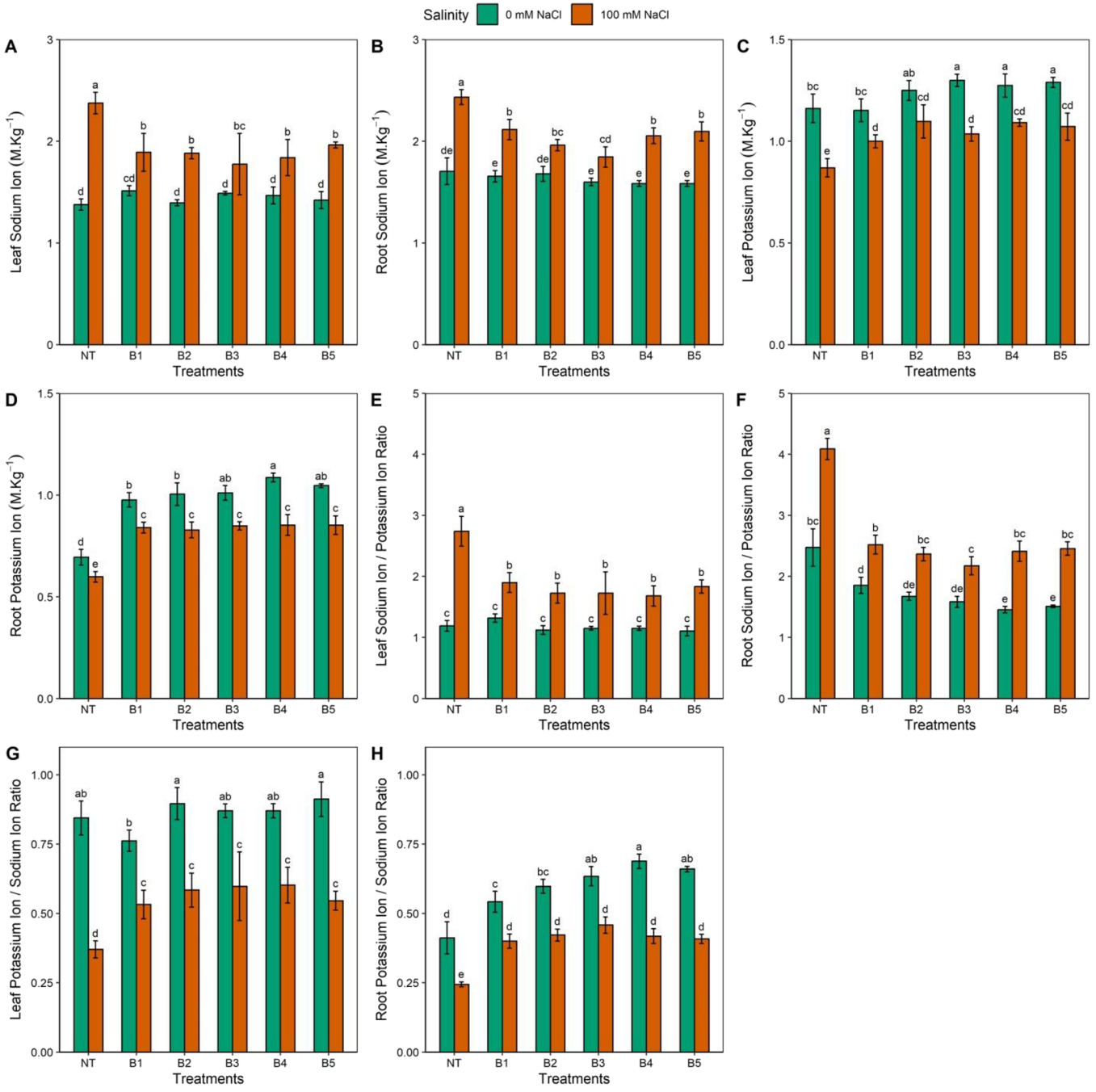


Figure 8. Effect of different *Pseudomonas* strains on ion concentration in leaf and root of *Sorghum bicolor* plants grown under salt stress (0 and 100 mM NaCl). Leaf Na^+ concentration (A), root Na^+ concentration (B), leaf K^+ concentration (C), root K^+ concentration (D), leaf Na^+/K^+ ratio (E), root Na^+/K^+ ratio (F), leaf K^+/Na^+ ratio (G), and root K^+/Na^+ ratio (H). The details on the treatments are given in Figure 2. The bars are presented as treatment mean ± SD ($n = 5$). The different alphabetic letters above the bars represent the significant differences at $P < 0.05$ according to Tukey's HSD test following ANOVA to compare the effect of each treatment.

under salinity. This improvement in ion homeostasis contributed to improving plant growth under stress conditions. This might happen because the exogenous application of *Pseudomonas* strains accumulated EPS on roots. These EPS capture excess Na^+ and promote Na^+ exclusion from root;[89] therefore, the uptake of K^+ was enhanced and transported into leaves of *Pseudomonas*-treated plants under saline conditions.[90] Enhanced uptake of K^+ is linked with increased salt tolerance because of its involvement in defensive compound biosynthesis and strengthening the plant cellular systems under salinity.[86,87,91]

Overall, this research work on the exogenous application of *Pseudomonas* strains improved the growth of the *Sorghum* plant due to modification in physiological changes and antioxidant responses under nonstress and salinity conditions. Although this work was conducted under sterilized conditions, it showed the response of only individual strains. However, in field conditions, diverse microbiota exist and plants recruit beneficial microorganisms that provide plants with different growth-promoting substances and nutrients. It is also believed that the bioinoculation of beneficial bacteria also help plants to recruit other beneficial microbiota;[92,93] however, direct

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

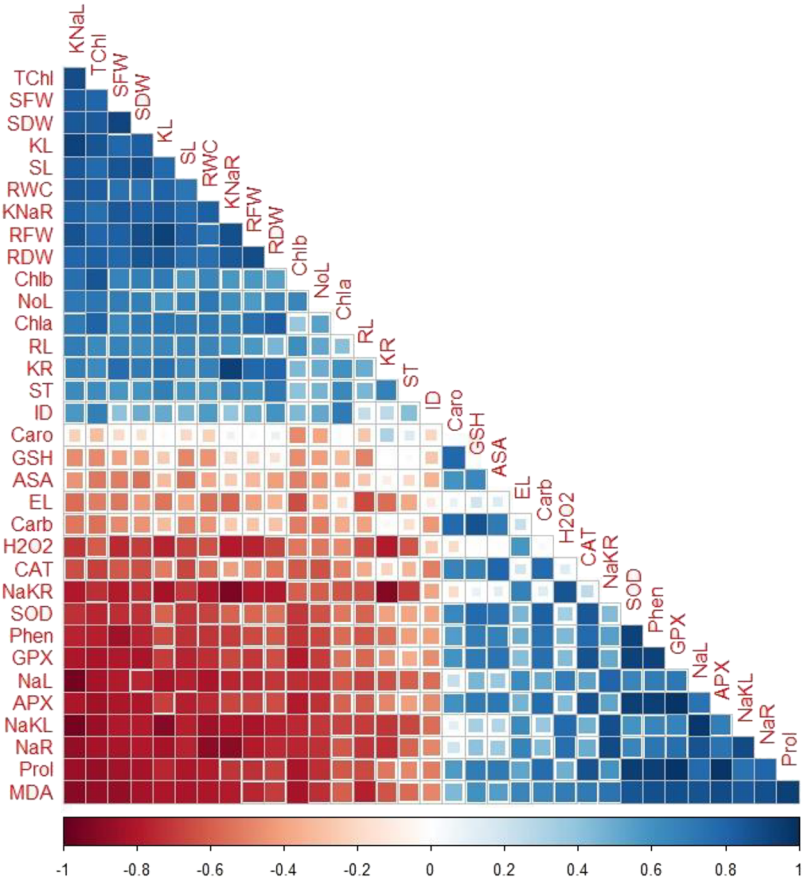


Figure 9. Pearson's correlation matrix between growth, antioxidant system, and eco-physiological parameters modified by the application of *Pseudomonas* strains under salinity. APX = ascorbate peroxidase activity; ASA = ascorbic acid; Carb = soluble carbohydrates; Caro = carotenoids; CAT = catalase activity; Chla = chlorophylls a; Chlb = chlorophylls b; EL = electrolyte leakage; GSH = reduced glutathione; H_2O_2 = hydrogen peroxide; ID = intermodal distance; KL = potassium ion in leaf; KNaL = potassium:sodium ion ratio in leaf; KNaR = potassium:sodium ion ratio in root; KR = potassium in root; MDA = malondialdehyde; NaKL = sodium:potassium ions ratio in leaf; NaKR = sodium:potassium ion ratio in root; NaL = sodium ions in leaf; NaR = sodium ions in root; NoL = number of leaves; POX = guaiacol peroxidase activity; Prol = proline; RDW = root dry weight; RFW = root fresh weight; RL = root length; RWC = relative water content; SDW = shoot dry weight; SFW = shoot fresh weight; SL = shoot length; SOD = superoxide dismutase activity; ST = stem thickness; and TChl = Total chlorophylls.

evidence supporting this presumption is limited. Pepper seedlings preinoculated with *Bacillus velezensis* NJAU-Z9 strain were shown to modify the rhizospheric microbiota including bacteria and fungi to increase the yield.[94] Similarly, the bioinoculation of various bacterial strains also modified the rhizospheric microbiota and increased the beneficial bacterial diversity leading to better plant health.[95] These could suggest that the exogenous application of *Pseudomonas* strains is beneficial for *Sorghum* growth under salinity; however, before being utilized as bioinoculant, their interaction with the existing microbiota would be evaluated under controlled and field conditions because the coexisting microbial communities would form a stable system to provide nutrients and metabolites that help to improve plant resilience to environmental stresses including salinity.

5. CONCLUSIONS

It is evident from the above discussion that bioinoculation with different *Pseudomonas* strains has proved helpful in increasing salt tolerance of *Sorghum*. This was achieved by improving plants' antioxidant defense system by increasing antioxidant enzymes (CAT, APX, POX, and SOD) and nonenzymatic antioxidants (ascorbic acid, glutathione), reducing oxidative damage components (H_2O_2, MDA, and EL), enhancing osmoprotectant accumulation (soluble sugars, proline, phenolic compounds, and protein), and balancing nutritional equilibrium by reducing Na^+ and improving K^+ and K^+/Na^+ ratio, which ultimately increased the photosynthetic pigments (chlorophylls and carotenoids), thus improving growth of *Sorghum* under salinity conditions. *P. aeruginosa* (B2 and B4) and *P. putida* (B5) were found more beneficial in improved morphological, biochemical, and eco-physiological attributes of *Sorghum* under salinity. Therefore, it is to be suggested that the above selected *Pseudomonas* strains could be utilized in enhanced salt tolerance of the high bioenergy crop *S. bicolor*.

AUTHOR INFORMATION

Corresponding Author

Muhammad Azeem − *Department of Botany, University of Karachi, Karachi 75270, Pakistan;* orcid.org/0000-0003-1668-3830; Email: m.azeem@uok.edu.pk

Authors

Robina Sultana − *Department of Botany, University of Karachi, Karachi 75270, Pakistan*

Naeem Ahmed − *Department of Botany, University of Karachi, Karachi 75270, Pakistan*

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

Muhammad Waseem Abbasi − *Department of Botany, University of Karachi, Karachi 75270, Pakistan*
Khwaja Ali Hasan − *Molecular and Structural Biochemistry Research Unit, Department of Biochemistry, University of Karachi, Karachi 75270, Pakistan*
Renjie Dong − *College of Engineering, China Agricultural University, Beijing 100083, China*
Saud Alamri − *Department of Botany and Microbiology, College of Science, King Saud University, Riyadh 11451, Saudi Arabia*
Alanoud T. Alfagham − *Department of Botany and Microbiology, College of Science, King Saud University, Riyadh 11451, Saudi Arabia*

Complete contact information is available at: https://pubs.acs.org/10.1021/acsomega.4c08110

Author Contributions

M.A.: conceptualization, resources, and supervision. R.S.: investigation and methodology. N.A.: software, formal analysis, and visualization. K.A.H.: bacterial molecular identification. N.A. and M.A.: writing—original draft and revision. R.D., S.A., N.A., M.A., and M.W.A., and A.T.A.: writing—review editing. S.A.: open access funding. All authors actively participated in manuscript writing, revision, and approval of the article.

Funding

The authors would like to extend their sincere appreciation to the Higher Education Commission of Pakistan (HEC) for the NRPU research project no. 17396. The authors are thankful to the Researchers Supporting Project number (RSP2025R194), King Saud University, Riyadh, Saudi Arabia, and also extend their sincere appreciation to Prof. Dr. Ehtesham ul Haq (late) for providing bacterial strains and Muhammad Noman Syed for providing technical support regarding bacterial strain identification.

Notes

The authors declare no competing financial interest.

REFERENCES

(1) Singh, B. K. Creating New Business, Economic Growth and Regional Prosperity through Microbiome-based Products in the Agriculture Industry. *Microb. Biotechnol.* **2017**, *10* (2), 224−227.
(2) Awan, S. A.; Ilyas, N.; Khan, I.; Raza, M. A.; Rehman, A. U.; Rizwan, M.; Rastogi, A.; Tariq, R.; Brestic, M. Bacillus Siamensis Reduces Cadmium Accumulation and Improves Growth and Antioxidant Defense System in Two Wheat (Triticum Aestivum L.) Varieties. *Plants* **2020**, *9* (7), 878.
(3) Abdul Rahman, N. S. N.; Abdul Hamid, N. W.; Nadarajah, K. Effects of Abiotic Stress on Soil Microbiome. *Int. J. Mol. Sci.* **2021**, *22* (16), 9036.
(4) Kibria, M. G.; Hoque, Md. A. A Review on Plant Responses to Soil Salinity and Amelioration Strategies. *Open J. Soil Sci.* **2019**, *09* (11), 219−231.
(5) Mishra, A. K.; Das, R.; George Kerry, R.; Biswal, B.; Sinha, T.; Sharma, S.; Arora, P.; Kumar, M. Promising Management Strategies to Improve Crop Sustainability and to Amend Soil Salinity. *Front. Environ. Sci.* **2023**, *10*, No. 962581.
(6) Tomar, R. S.; Kataria, S.; Jajoo, A. Behind the Scene: Critical Role of Reactive Oxygen Species and Reactive Nitrogen Species in Salt Stress Tolerance. *J. Agron. Crop Sci.* **2021**, *207* (4), 577−588.
(7) Klein, A.; Hüsselmann, L.; Keyster, M.; Ludidi, N. Exogenous Nitric Oxide Limits Salt-Induced Oxidative Damage in Maize by Altering Superoxide Dismutase Activity. *South Afr. J. Bot.* **2018**, *115*, 44−49.
(8) Satish, L.; Shilpha, J.; Pandian, S.; Rency, A. S.; Rathinapriya, P.; Ceasar, S. A.; Largia, M. J. V.; Kumar, A. A.; Ramesh, M. Analysis of Genetic Variation in Sorghum (Sorghum bicolor (L.) Moench) Genotypes with Various agronomical Traits Using SPAR Methods. *Gene* **2016**, *576* (1), 581−585.
(9) Hussien Ibrahim, M. E.; Adam Ali, A. Y.; Zhou, G.; Ibrahim Elsiddig, A. M.; Zhu, G.; Ahmed Nimir, N. E.; Ahmad, I. Biochar Application Affects Forage Sorghum under Salinity Stress. *Chil. J. Agric. Res.* **2020**, *80* (3), 317−325.
(10) Ali, Z.; Khan, D.; Ahmed, N. Salt Tolerance of Three Sorghum Cultivars during Germination and Early Seedling Growth. *Int. J. Biol. Biotechnol.* **2013**, *10* (2), 193−202.
(11) Almodares, A.; Sharif, M. E. Effect of Water Quality on Yield of Sugar Beet and Sweet Sorghum. *J. Environ. Biol.* **2005**, *26* (3), 487−493.
(12) Ali, Z.; Khan, D.; Ahmed, N. Physiological Parameters of Salt Tolerance in Three Cultivars of Sorghum bicolor (L.) Moench. at Seedling Stage under Single Salt (NaCl) Salinity. *Int. J. Biol. Biotechol.* **2013**, *10* (1), 125−142.
(13) Javid, M. G.; Sorooshzadeh, A.; Moradi, F.; Modarres, S. S. A. M.; Allahdadi, I. The Role of Phytohormones in Alleviating Salt Stress in Crop Plants. *Aust. J. Crop Sci.* **2011**, *5* (6), 726−734.
(14) Kumar, K.; Gambhir, G.; Dass, A.; Tripathi, A. K.; Singh, A.; Jha, A. K.; Yadava, P.; Choudhary, M.; Rakshit, S. Genetically Modified Crops: Current Status and Future Prospects. *Planta* **2020**, *251* (4), 91.
(15) Hossain, A.; Bhatt, R.; Sarkar, S.; Barman, M.; Majumder, D.; Saha, S.; Islam Mst, T.; Maitra, S.; Meena, R. S. Cost-Effective and Eco-Friendly Agricultural Technologies in Rice-Wheat Cropping Systems for Food and Environmental Security. In *Sustainable Intensification for Agroecosystem Services and Management*; Jhariya, M. K., Banerjee, A., Meena, R. S., Kumar, S., Raj, A., Eds.; Springer Singapore: Singapore, 2021; pp 69−96. .
(16) Kumawat, K. C.; Sharma, B.; Nagpal, S.; Kumar, A.; Tiwari, S.; Nair, R. M. Plant Growth-Promoting Rhizobacteria: Salt Stress Alleviators to Improve Crop Productivity for Sustainable Agriculture Development. *Front. Plant Sci.* **2023**, *13*, 1101862.
(17) Aloo, B. N.; Dessureault-Rompré, J.; Tripathi, V.; Nyongesa, B. O.; Were, B. A. Signaling and Crosstalk of Rhizobacterial and Plant Hormones That Mediate Abiotic Stress Tolerance in Plants. *Front. Microbiol.* **2023**, *14*, 1171104.
(18) Giannelli, G.; Potestio, S.; Visioli, G. The Contribution of PGPR in Salt Stress Tolerance in Crops: Unravelling the Molecular Mechanisms of Cross-Talk between Plant and Bacteria. *Plants* **2023**, *12* (11), 2197.
(19) Abbasi, M. W.; Ahmed, N.; Zaki, M. J.; Shuakat, S. S.; Khan, D. Potential of Bacillus Species against Meloidogyne Javanica Parasitizing Eggplant (Solanum Melongena L.) and Induced Biochemical Changes. *Plant Soil* **2014**, *375* (1−2), 159−173.
(20) Forni, C.; Duca, D.; Glick, B. R. Mechanisms of Plant Response to Salt and Drought Stress and Their Alteration by Rhizobacteria. *Plant Soil* **2017**, *410* (1−2), 335−356.
(21) Bharti, N.; Barnawal, D. Amelioration of Salinity Stress by PGPR. In *PGPR Amelioration in Sustainable Agriculture*; Elsevier, 2019; pp 85−106. .
(22) Shah, G.; Jan, M.; Afreen, M.; Anees, M.; Rehman, S.; Daud, M. K.; Malook, I.; Jamil, M. Halophilic Bacteria Mediated Phytoremediation of Salt-Affected Soils Cultivated with Rice. *J. Geochem. Explor.* **2017**, *174*, 59−65.
(23) Gul, S.; Javed, S.; Azeem, M.; Aftab, A.; Anwaar, N.; Mehmood, T.; Zeshan, B. Application of Bacillus Subtilis for the Alleviation of Salinity Stress in Different Cultivars of Wheat (Tritium Aestivum L.). *Agronomy* **2023**, *13* (2), 437.
(24) Dong, H.; Wang, Y.; Di, Y.; Qiu, Y.; Ji, Z.; Zhou, T.; Shen, S.; Du, N.; Zhang, T.; Dong, X.; Guo, Z.; Piao, F.; Li, Y. Plant Growth-Promoting Rhizobacteria Pseudomonas Aeruginosa HG28−5 Improves Salt Tolerance by Regulating Na+/K+ Homeostasis and ABA Signaling Pathway in Tomato. *Microbiol. Res.* **2024**, *283*, No. 127707.

(25) Korejo, F.; Ali, S. A.; Humayun, F.; Rahman, A.; Sultana, V.; Ara, J.; Ehteshamul-Haque, S. Management of Root Rotting Fungi and Root Knot Nematode with Endophytic Fluorescent Pseudomonas Associated with Salvadora Species. *Pak. J. Bot.* **2019**, *51* (4), 1507.
(26) Khan, A.; Singh, A. V. Multifarious Effect of ACC Deaminase and EPS Producing Pseudomonas Sp. and Serratia Marcescens to Augment Drought Stress Tolerance and Nutrient Status of Wheat. *World J. Microbiol. Biotechnol.* **2021**, *37* (12), 198.
(27) Kour, D.; Rana, K. L.; Sheikh, I.; Kumar, V.; Yadav, A. N.; Dhaliwal, H. S.; Saxena, A. K. Alleviation of Drought Stress and Plant Growth Promotion by Pseudomonas Libanensis EU-LWNA-33, a Drought-Adaptive Phosphorus-Solubilizing Bacterium. *Proc. Natl. Acad. Sci. India Sect. B Biol. Sci.* **2020**, *90* (4), 785−795.
(28) Zhang, M.; Yang, L.; Hao, R.; Bai, X.; Wang, Y.; Yu, X. Drought-Tolerant Plant Growth-Promoting Rhizobacteria Isolated from Jujube (Ziziphus Jujuba) and Their Potential to Enhance Drought Tolerance. *Plant Soil* **2020**, *452* (1−2), 423−440.
(29) Chu, T. N.; Tran, B. T. H.; Van Bui, L.; Hoang, M. T. T. Plant Growth-Promoting Rhizobacterium Pseudomonas PS01 Induces Salt Tolerance in Arabidopsis Thaliana. *BMC Res. Notes* **2019**, *12* (1), 11.
(30) Samaddar, S.; Chatterjee, P.; Roy Choudhury, A.; Ahmed, S.; Sa, T. Interactions between Pseudomonas Spp. and Their Role in Improving the Red Pepper Plant Growth under Salinity Stress. *Microbiol. Res.* **2019**, *219*, 66−73.
(31) Pandey, S.; Gupta, S. Evaluation of Pseudomonas Sp. for Its Multifarious Plant Growth Promoting Potential and Its Ability to Alleviate Biotic and Abiotic Stress in Tomato (Solanum Lycopersicum) Plants. *Sci. Rep.* **2020**, *10* (1), 20951.
(32) Costa-Gutierrez, S. B.; Lami, M. J.; Santo, M. C. C.-D.; Zenoff, A. M.; Vincent, P. A.; Molina-Henares, M. A.; Espinosa-Urgel, M.; De Cristóbal, R. E. Plant Growth Promotion by Pseudomonas Putida KT2440 under Saline Stress: Role of eptA. *Appl. Microbiol. Biotechnol.* **2020**, *104* (10), 4577−4592.
(33) Singh, P.; Singh, R. K.; Zhou, Y.; Wang, J.; Jiang, Y.; Shen, N.; Wang, Y.; Yang, L.; Jiang, M. Unlocking the Strength of Plant Growth Promoting *Pseudomonas* in Improving Crop Productivity in Normal and Challenging Environments: A Review. *J. Plant Interact.* **2022**, *17* (1), 220−238.
(34) Hashem, A.; Alarjani, K. M.; Almutariri, K. F.; Parray, J. A.; Sharma, S. K.; Kumar, A.; Dawoud, T. M.; Almaary, K. S.; Shameem, N.; Abd-Allah, E. F. Impact of Microbiomes to Counter Abiotic Stresses in Medicinal Plants- A Review. In *Core Microbiome: Improving Crop Quality and Productivity*; Parray, J. A., Shameem, N., Abd-Allah, E. F., Mir, M. Y., Eds.; Wiley, 2022; pp 30−50. .
(35) Rajabi Dehnavi, A.; Piernik, A.; Ludwiczak, A.; Szymańska, S.; Ciarkowska, A.; Cárdenas Pérez, S.; Hrynkiewicz, K. Mitigation of Salt Stress in Sorghum bicolor L. by the halotolerant Endophyte Pseudomonas stutzeri ISE12. *Front. Plant Sci.* **2024**, *15*, 1458540.
(36) Habiba, S.; Noreen, R.; Ali, S. A.; Hasan, K. A.; Ara, J.; Sultana, V.; Ehteshamul-Haque, S. Protective Role of Epiphytic Fluorescent Pseudomonas on Natural Postharvest Decay of Tomato at Room Temperature. *J. Appl. Bot. Food Qual.* **2017**, 288−297.
(37) Weatherley, P. E. Studies in the Water Relations of the Cotton Plant. I. The Field Measurement of Water Deficits in Leaves. *New Phytol.* **1950**, *49* (1), 81−97.
(38) Dionisio-Sese, M. L.; Tobita, S. Antioxidant Responses of Rice Seedlings to Salinity Stress. *Plant Sci.* **1998**, *135* (1), 1−9.
(39) Lichtenthaler, H. K.; Wellburn, A. R. Determinations of Total Carotenoids and chlorophylls *a* and *b* of Leaf Extracts in Different Solvents. *Biochem. Soc. Trans.* **1983**, *11* (5), 591−592.
(40) Yemm, E. W.; Willis, A. J. The Estimation of Carbohydrates in Plant Extracts by Anthrone. *Biochem. J.* **1954**, *57* (3), 508−514.
(41) Bates, L. S.; Waldren, R. P.; Teare, I. D. Rapid Determination of Free Proline for Water-Stress Studies. *Plant Soil* **1973**, *39* (1), 205−207.
(42) Singleton, V. L.; Rossi, J. A. Colorimetry of Total Phenolics with Phosphomolybdic-Phosphotungstic Acid Reagents. *Am. J. Enol. Vitic.* **1965**, *16* (3), 144−158.
(43) Guri, A. VARIATION IN GLUTATHIONE AND ASCORBIC ACID CONTENT AMONG SELECTED CULTIVARS OF *Phaseolus Vulgaris* PRIOR TO AND AFTER EXPOSURE TO OZONE. *Can. J. Plant Sci.* **1983**, *63* (3), 733−737.
(44) Bradford, M. M. A Rapid and Sensitive Method for the Quantitation of Microgram Quantities of Protein Utilizing the Principle of Protein-Dye Binding. *Anal. Biochem.* **1976**, *72* (1−2), 248−254.
(45) Aebi, H. [13] Catalase in Vitro. *Methods Enzymol.* **1984**, *105*, 121−126.
(46) Nakano, Y.; Asada, K. Hydrogen Peroxide Is Scavenged by Ascorbate-Specific Peroxidase in Spinach Chloroplasts. *Plant Cell Physiol.* **1981**, *22* (5), 867−880.
(47) Polle, A.; Otter, T.; Seifert, F. Apoplastic Peroxidases and Lignification in Needles of Norway Spruce (Picea Abies L.). *Plant Physiol.* **1994**, *106* (1), 53−60.
(48) Beyer, W. F.; Fridovich, I. Assaying for Superoxide Dismutase Activity: Some Large Consequences of Minor Changes in Conditions. *Anal. Biochem.* **1987**, *161* (2), 559−566.
(49) Velikova, V.; Yordanov, I.; Edreva, A. Oxidative Stress and Some Antioxidant Systems in Acid Rain-Treated Bean Plants. *Plant Sci.* **2000**, *151* (1), 59−66.
(50) Heath, R. L.; Packer, L. Photoperoxidation in Isolated Chloroplasts. *Arch. Biochem. Biophys.* **1968**, *125* (1), 189−198.
(51) Khan, M. A.; Ungar, I. A.; Showalter, A. M. The Effect of Salinity on the Growth, Water Status, and Ion Content of a Leaf Succulent Perennial Halophyte, Suaeda Fruticosa (L.) Forssk. *J. Arid Environ.* **2000**, *45* (1), 73−84.
(52) Kumar, V.; Raghuvanshi, N.; Pandey, A. K.; Kumar, A.; Thoday-Kennedy, E.; Kant, S. Role of halotolerant Plant Growth-Promoting Rhizobacteria in Mitigating Salinity Stress: Recent Advances and Possibilities. *Agriculture* **2023**, *13* (1), 168.
(53) Rabiei, Z.; Hosseini, S. J.; Pirdashti, H.; Hazrati, S. Physiological and Biochemical Traits in Coriander Affected by Plant Growth-Promoting Rhizobacteria under Salt Stress. *Heliyon* **2020**, *6* (10), No. e05321.
(54) Jangra, M.; Devi, S.; Satpal; Kumar, N.; Goyal, V.; Mehrotra, S. Amelioration Effect of Salicylic Acid Under Salt Stress in Sorghum bicolor L. *Appl. Biochem. Biotechnol.* **2022**, *194* (10), 4400−4423.
(55) Li, J.; Lei, S.; Gong, H.; Liu, Z.; Zhang, Y.; Ouyang, Z. Field Performance of Sweet Sorghum in Salt-Affected Soils in China: A Quantitative Synthesis. *Environ. Res.* **2023**, *222*, No. 115362.
(56) Azeem, M.; Sultana, R.; Mahmood, A.; Qasim, M.; Siddiqui, Z. S.; Mumtaz, S.; Javed, T.; Umar, M.; Adnan, M. Y.; Siddiqui, M. H. Ascorbic and Salicylic Acids Vitalized Growth, Biochemical Responses, Antioxidant Enzymes, Photosynthetic Efficiency, and Ionic Regulation to Alleviate Salinity Stress in Sorghum bicolor. *J. Plant Growth Regul.* **2023**, *42* (8), 5266−5279.
(57) Ullah, A.; Bano, A. Role of PGPR in the Reclamation and Revegetation of Saline Land. *Pak. J. Bot.* **2019**, *51* (1), 27−35.
(58) Timofeeva, A. M.; Galyamova, M. R.; Sedykh, S. E. Plant Growth-Promoting Soil Bacteria: Nitrogen Fixation, Phosphate Solubilization, Siderophore Production, and Other Biological Activities. *Plants* **2023**, *12* (24), 4074.
(59) Alori, E. T.; Glick, B. R.; Babalola, O. O. Microbial Phosphorus Solubilization and Its Potential for Use in Sustainable Agriculture. *Front. Microbiol.* **2017**, *8*, 971.
(60) Glick, B. R.; Nascimento, F. X. Pseudomonas 1-Amino-cyclopropane-1-Carboxylate (ACC) Deaminase and Its Role in Beneficial Plant-Microbe Interactions. *Microorganisms* **2021**, *9* (12), 2467.
(61) Shahid, M.; Singh, U. B.; Khan, M. S.; Singh, P.; Kumar, R.; Singh, R. N.; Kumar, A.; Singh, H. V. Bacterial ACC Deaminase: Insights into Enzymology, Biochemistry, Genetics, and Potential Role in Amelioration of Environmental Stress in Crop Plants. *Front. Microbiol.* **2023**, *14*, 1132770.
(62) Liu, X.; Yao, T.; Chai, J.; Han, J. Adsorption of Sodium Ions by Exopolysaccharides from *Pseudomonas Simiae* MHR6 and Its

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954

Improvement of Na $^{+}$ /K $^{+}$ Homeostasis in Maize under Salt Stress. *J. Agric. Food Chem.* **2023**, *71* (50), 19949−19957.
(63) Ansari, M.; Shekari, F.; Mohammadi, M. H.; Juhos, K.; Végvári, G.; Biró, B. Salt-Tolerant Plant Growth-Promoting Bacteria Enhanced Salinity Tolerance of Salt-Tolerant Alfalfa (Medicago Sativa L.) Cultivars at High Salinity. *Acta Physiol. Plant.* **2019**, *41* (12), 195.
(64) Wungrampha, S.; Joshi, R.; Singla-Pareek, S. L.; Pareek, A. Photosynthesis and Salinity: Are These Mutually Exclusive? *Photosynthetica* **2018**, *56*, 366−381.
(65) Hameed, A.; Ahmed, M. Z.; Hussain, T.; Aziz, I.; Ahmad, N.; Gul, B.; Nielsen, B. L. Effects of Salinity Stress on Chloroplast Structure and Function. *Cells* **2021**, *10* (8), 2023.
(66) Pan, T.; Liu, M.; Kreslavski, V. D.; Zharmukhamedov, S. K.; Nie, C.; Yu, M.; Kuznetsov, V. V.; Allakhverdiev, S. I.; Shabala, S. Non-Stomatal Limitation of Photosynthesis by Soil Salinity. *Crit. Rev. Environ. Sci. Technol.* **2021**, *51* (8), 791−825.
(67) Dias, A. S.; de Lima, G. S.; da Silva, S. S.; dos Anjos Soares, L. A.; Chaves, L. H. G.; Gheyi, H. R.; de Lacerda, C. N.; Fernandes, P. D. Gas Exchange, Photosynthetic Pigments, and Photochemical Efficiency of Sesame under Salt Stress and Phosphate Fertilization. *Semina: Cienc. Agrár.* **2022**, 1237.
(68) Khan, T. A.; Saleem, M.; Fariduddin, Q. Melatonin Influences Stomatal Behavior, Root Morphology, Cell Viability, Photosynthetic Responses, Fruit Yield, and Fruit Quality of Tomato Plants Exposed to Salt Stress. *J. Plant Growth Regul.* **2023**, *42* (4), 2408−2432.
(69) He, Y.; Wu, Z.; Wang, W.; Ye, B.-C.; Zhang, F.; Liu, X. Different Responses of Capsicum Annuum L. Root and Shoot to Salt Stress with Pseudomonas Putida Rs-198 Inoculation. *J. Plant Growth Regul.* **2019**, *38* (3), 799−811.
(70) Arefian, M.; Vessal, S.; Malekzadeh-Shafaroudi, S.; Siddique, K. H. M.; Bagheri, A. Comparative Proteomics and Gene Expression Analyses Revealed Responsive Proteins and Mechanisms for Salt Tolerance in Chickpea Genotypes. *BMC Plant Biol.* **2019**, *19* (1), 300.
(71) Cheng, Z.; Woody, O. Z.; McConkey, B. J.; Glick, B. R. Combined Effects of the Plant Growth-Promoting Bacterium Pseudomonas Putida UW4 and Salinity Stress on the Brassica Napus Proteome. *Appl. Soil Ecol.* **2012**, *61*, 255−263.
(72) Zhang, Y.; Yang, F.; Ren, H.; Liu, J.; Mu, W.; Wang, Y. Pseudomonas Monteilii PN1: A Great Potential P-Nitrophenol Degrader with Plant Growth Promoting Traits under Drought and Saline−Alkali Stresses. *Biotechnol. Lett.* **2019**, *41* (6−7), 801−811.
(73) Kumudini, B. S.; Patil, S. V. Role of Plant Hormones in Improving Photosynthesis. In *Photosynthesis, Productivity and Environmental Stress*; Ahmad, P., Ahanger, M. A., Alyemeni, M. N., Alam, P., Eds.; Wiley, 2019; pp 215−240. .
(74) Sharma, N.; Muthamilarasan, M.; Dulani, P.; Prasad, M. Genomic Dissection of ROS Detoxifying Enzyme Encoding Genes for Their Role in Antioxidative Defense Mechanism against Tomato Leaf Curl New Delhi Virus Infection in Tomato. *Genomics* **2021**, *113* (3), 889−899.
(75) Szymańska, S.; Dąbrowska, G. B.; Tyburski, J.; Niedojadło, K.; Piernik, A.; Hrynkiewicz, K. Boosting the Brassica Napus L. Tolerance to Salinity by the halotolerant Strain Pseudomonas stutzeri ISE12. *Environ. Exp. Bot.* **2019**, *163*, 55−68.
(76) Farhangi-Abriz, S.; Tavasolee, A.; Ghassemi-Golezani, K.; Torabian, S.; Monirifar, H.; Rahmani, H. A. Growth-Promoting Bacteria and Natural Regulators Mitigate Salt Toxicity and Improve Rapeseed Plant Performance. *Protoplasma* **2020**, *257* (4), 1035−1047.
(77) Gamalero, E.; Glick, B. R. Recent Advances in Bacterial Amelioration of Plant Drought and Salt Stress. *Biology* **2022**, *11* (3), 437.
(78) Khan, V.; Princi; Mubashshir, M.; Umar, S.; Iqbal, N. Methyl Jasmonate and Pseudomonas Fluorescens Synergistically Boost Antioxidative Defense, Secondary Metabolites, and Osmolyte Production to Enhance Drought Resilience in Mustard. *J. Plant Growth Regul.* **2024**.
(79) Abbas, R.; Rasul, S.; Aslam, K.; Baber, M.; Shahid, M.; Mubeen, F.; Naqqash, T. halotolerant PGPR: A Hope for Cultivation of Saline Soils. *J. King Saud Univ. - Sci.* **2019**, *31* (4), 1195−1201.
(80) Islam, F.; Yasmeen, T.; Ali, Q.; Ali, S.; Arif, M. S.; Hussain, S.; Rizvi, H. Influence of Pseudomonas Aeruginosa as PGPR on Oxidative Stress Tolerance in Wheat under Zn Stress. *Ecotoxicol. Environ. Saf.* **2014**, *104*, 285−293.
(81) Mahadik, S.; Kumudini, B. S. Enhancement of Salinity Stress Tolerance and Plant Growth in Finger Millet Using Fluorescent Pseudomonads. *Rhizosphere* **2020**, *15*, No. 100226.
(82) Santander, C.; Vidal, G.; Ruiz, A.; Vidal, C.; Cornejo, P. Salinity Eustress Increases the Biosynthesis and Accumulation of Phenolic Compounds That Improve the Functional and Antioxidant Quality of Red Lettuce. *Agronomy* **2022**, *12* (3), 598.
(83) Khan, V.; Umar, S.; Iqbal, N. Synergistic Action of *Pseudomonas Fluorescens* with Melatonin Attenuates Salt Toxicity in Mustard by Regulating Antioxidant System and Flavonoid Profile. *Physiol. Plant.* **2023**, *175* (6), No. e14092.
(84) Kang, S.-M.; Radhakrishnan, R.; Khan, A. L.; Kim, M.-J.; Park, J.-M.; Kim, B.-R.; Shin, D.-H.; Lee, I.-J. Gibberellin Secreting Rhizobacterium, Pseudomonas Putida H-2−3 Modulates the Hormonal and Stress Physiology of Soybean to Improve the Plant Growth under Saline and Drought Conditions. *Plant Physiol. Biochem.* **2014**, *84*, 115−124.
(85) Tayyab, N.; Naz, R.; Yasmin, H.; Nosheen, A.; Keyani, R.; Sajjad, M.; Hassan, M. N.; Roberts, T. H. Combined Seed and Foliar Pre-Treatments with Exogenous Methyl Jasmonate and Salicylic Acid Mitigate Drought-Induced Stress in Maize. *PLoS One* **2020**, *15* (5), No. e0232269.
(86) Hussain, S.; Hussain, S.; Ali, B.; Ren, X.; Chen, X.; Li, Q.; Saqib, M.; Ahmad, N. Recent Progress in Understanding Salinity Tolerance in Plants: Story of Na+/K+ Balance and Beyond. *Plant Physiol. Biochem.* **2021**, *160*, 239−256.
(87) Sahito, Z. A.; Khan, D.; Ahmed, N. Some Parameters of Growth of River Cooba Seedlings under Salt Stress. *Int. J. Biol. Biotechnol.* **2013**, *10* (3), 339−352.
(88) Isayenkov, S. V. Genetic Sources for the Development of Salt Tolerance in Crops. *Plant Growth Regul.* **2019**, *89* (1), 1−17.
(89) Ashraf, M.; Hasnain, S.; Berge, O.; Mahmood, T. Inoculating Wheat Seedlings with Exopolysaccharide-Producing Bacteria Restricts Sodium Uptake and Stimulates Plant Growth under Salt Stress. *Biol. Fertil. Soils* **2004**, *40* (3), 157.
(90) Ilangumaran, G.; Smith, D. L. Plant Growth Promoting Rhizobacteria in Amelioration of Salinity Stress: A Systems Biology Perspective. *Front. Plant Sci.* **2017**, *8*, 1768.
(91) Kumari, S.; Chhillar, H.; Chopra, P.; Khanna, R. R.; Khan, M. I. R. Potassium: A Track to Develop Salinity Tolerant Plants. *Plant Physiol. Biochem.* **2021**, *167*, 1011−1023.
(92) Santoyo, G. How Plants Recruit Their Microbiome? New Insights into Beneficial Interactions. *J. Adv. Res.* **2022**, *40*, 45−58.
(93) Ge, J.; Li, D.; Ding, J.; Xiao, X.; Liang, Y. Microbial Coexistence in the Rhizosphere and the Promotion of Plant Stress Resistance: A Review. *Environ. Res.* **2023**, *222*, No. 115298.
(94) Zhang, Y.; Gao, X.; Shen, Z.; Zhu, C.; Jiao, Z.; Li, R.; Shen, Q. Pre-Colonization of PGPR Triggers Rhizosphere Microbiota Succession Associated with Crop Yield Enhancement. *Plant Soil* **2019**, *439* (1−2), 553−567.
(95) Cunha, I. D. C. M. D.; Silva, A. V. R. D.; Boleta, E. H. M.; Pellegrinetti, T. A.; Zagatto, L. F. G.; Zagatto, S. D. S. S.; Chaves, M. G. D.; Mendes, R.; Patreze, C. M.; Tsai, S. M.; Mendes, L. W. The Interplay between the Inoculation of Plant Growth-Promoting Rhizobacteria and the Rhizosphere Microbiome and Their Impact on Plant Phenotype. *Microbiol. Res.* **2024**, *283*, No. 127706.

https://doi.org/10.1021/acsomega.4c08110
ACS Omega 2025, 10, 940−954