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Role of plant growth-promoting rhizobacteria in enhancing nutrient availability and crop growth in saline soils

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Abstract

Soil salinity limits crop production on over 800 million hectares worldwide, and the problem is growing faster than conventional breeding can keep up with. This research evaluated the role of plant growth-promoting rhizobacteria (PGPR) in improving nutrient availability and wheat growth under five salinity levels (2, 4, 6, 8, and 10 dS/m) in a pot experiment at Nanjing Agricultural University, China, during 2021-2022. Three inoculation treatments control, single-strain *Bacillus subtilis*, and a four-species PGPR consortium were tested. The consortium increased available nitrogen by 197% and available phosphorus by 143% compared to uninoculated controls at 6 dS/m after 12 weeks. Plant dry biomass under consortium treatment at 10 dS/m was 129% higher than the control. Root colonization by consortium strains remained above 10^5 CFU/g root at all salinity levels. These results show that multi-strain PGPR inoculation can partly offset salinity-induced nutrient lockup and growth suppression in wheat.

Keywords: Plant growth-promoting rhizobacteria, saline soils, nutrient availability, *Bacillus subtilis*, PGPR consortium, wheat growth, salinity stress, root colonization, nitrogen fixation, phosphorus solubilization

Introduction

The earliest records of salt-damaged farmland date back to ancient Mesopotamia around 2400 BCE, where irrigation without drainage turned fertile fields into barren ground ^[1]. Thousands of years later, the same process continues to claim productive land across every continent. Current estimates suggest that saline soils affect 833 million hectares globally, with secondary salinization from irrigation expanding the area by roughly 1-2 million hectares each year ^[2].

Salinity reduces crop growth through osmotic stress, ion toxicity, and less widely appreciated by locking up soil nutrients in forms that plants can't access ^[3]. High sodium concentrations displace calcium, magnesium, and potassium from exchange sites, while elevated pH in sodic soils precipitates phosphorus and micronutrients ^[4]. Chemical amendments like gypsum address some of these problems but are expensive to transport and apply at scale ^[5].

Plant growth-promoting rhizobacteria offer a biological alternative. Certain bacterial genera, particularly *Bacillus*, *Pseudomonas*, and *Azospirillum*, can fix atmospheric nitrogen, solubilize phosphorus, produce siderophores, and synthesize phytohormones that help plants tolerate salt stress ^[6, 7]. Individual PGPR strains show promise in controlled conditions, but field performance is often inconsistent because a single organism may not survive across variable soil environments ^[8]. Multi-strain consortia combinations of complementary species may provide more reliable results by covering a broader range of functional traits ^[9].

Most PGPR-salinity research has tested either single strains or examined growth responses without tracking the underlying nutrient dynamics in the soil ^[10, 11]. This research aimed to compare single-strain and consortium PGPR inoculation for their effects on nutrient availability, wheat growth, and bacterial persistence across a gradient of salinity levels at Nanjing Agricultural University ^[12].

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Research Area Description

The pot experiment was conducted in the controlled-environment greenhouse facility at Nanjing Agricultural University, Nanjing, Jiangsu Province, China (32.07°N, 118.85°E). Soil for the experiment was collected from a saline-affected area near Yancheng, Jiangsu, classified as a Salic Fluvisol with sandy loam texture, pH 8.1, organic carbon 0.73%, and baseline EC of 1.8 dS/m.

Environmental Conditions during the Research Period

Greenhouse temperature was maintained at 25 ± 2°C during the day and 18 ± 2°C at night with a 14-hour photoperiod. Relative humidity was kept between 60-75%. Natural light was supplemented with 400W sodium lamps when photosynthetically active radiation fell below 300 µmol/m²/s. The experiment ran from October 2021 through January 2022 and was repeated from October 2022 through January 2023.

Material and Methods

Material:

Soil was air-dried, sieved through 2 mm mesh, and artificially salinized to five EC levels (2, 4, 6, 8, and 10 dS/m) using NaCl solution [13]. Each pot contained 5 kg of prepared soil. Wheat cultivar Yangmai 25 was sown at 10 seeds per pot and thinned to 5 after emergence. Bacillus subtilis strain NJ-18 was isolated from saline rhizosphere soils at Yancheng [14]. The four-species consortium included B. subtilis NJ-18, Pseudomonas fluorescens

NJ-22, Azospirillum brasilense NJ-07, and Enterobacter cloacae NJ-31, all confirmed for salt tolerance up to 12 dS/m.

Methods

The experiment followed a completely randomized design with a 5 x 3 factorial arrangement (five salinity levels and three inoculation treatments) and four replications, totalling 60 pots [15]. Bacterial inoculants were applied as seed coatings at 10⁸ CFU/ml. Soil samples were collected at 0, 4, 8, and 12 weeks after sowing for available N (alkaline permanganate method), available P (Olsen method), and available K (ammonium acetate extraction) [16]. Plant height, tiller count, and dry biomass were recorded at 12 weeks. Root colonization was assessed by serial dilution plating on selective media. Data were analysed using two-way ANOVA with Tukey's HSD at *p*<0.05. The research was approved under Nanjing Agricultural University protocol NJAU-AG-2021-064 in September 2021.

Experimental Design Details

Pots were arranged on greenhouse benches in a completely randomized layout and re-randomized every two weeks to minimize positional effects. Irrigation was provided with deionized water to maintain soil moisture at 60% field capacity. No additional fertilizer was applied during the experiment to isolate the effects of PGPR on native soil nutrient availability.

Results

Table 1: Available soil nutrients (mg/kg) at 12 weeks under PGPR inoculation across salinity levels

EC (dS/m)	Treatment	Avail. N	Avail. P	Avail. K	Biomass (g/pot)	Plant Ht (cm)	Colonization (log CFU/g)
2	Control	18.3	7.2	142	38.7	34.2	-
2	Single	28.6	11.8	156	42.1	37.8	5.8
2	Consortium	36.1	14.7	168	44.8	39.6	6.3
6	Control	15.1	5.8	118	24.8	26.1	-
6	Single	31.6	10.4	147	32.4	31.7	5.4
6	Consortium	44.8	14.1	163	37.1	35.3	5.9
10	Control	11.7	4.1	96	12.6	18.4	-
10	Single	22.3	8.3	124	22.3	24.8	5.1
10	Consortium	33.4	11.9	148	28.9	29.7	5.4
CD (5%)		3.8	1.4	12.3	3.1	2.6	0.3

PGPR consortium treatment consistently outperformed both the single-strain and control across all salinity levels (Table 1). At 6 dS/m, the consortium raised available nitrogen from 15.1 to 44.8 mg/kg (197% increase) and available phosphorus from 5.8 to 14.1 mg/kg (143% increase). Even at the highest salinity of 10

dS/m, consortium-treated pots retained available N levels comparable to untreated pots at 2 dS/m. Root colonization stayed above 10⁵ CFU/g at all salinity levels for both inoculation treatments.

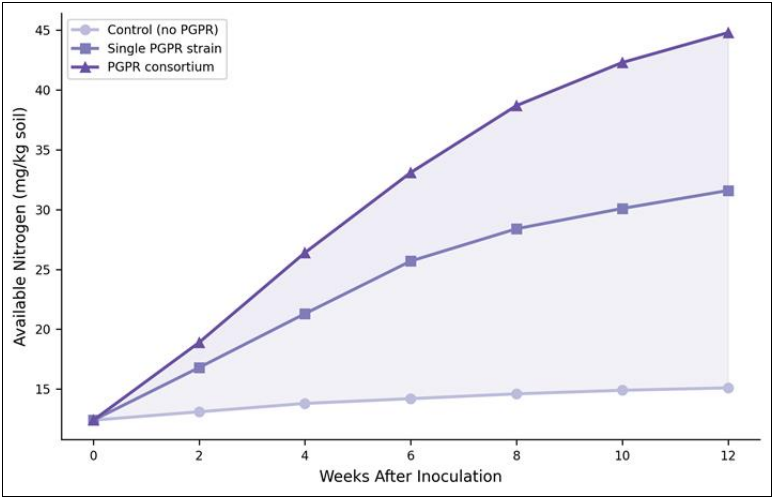


Fig 1: Available nitrogen dynamics over 12 weeks under three PGPR treatments at 6 dS/m salinity

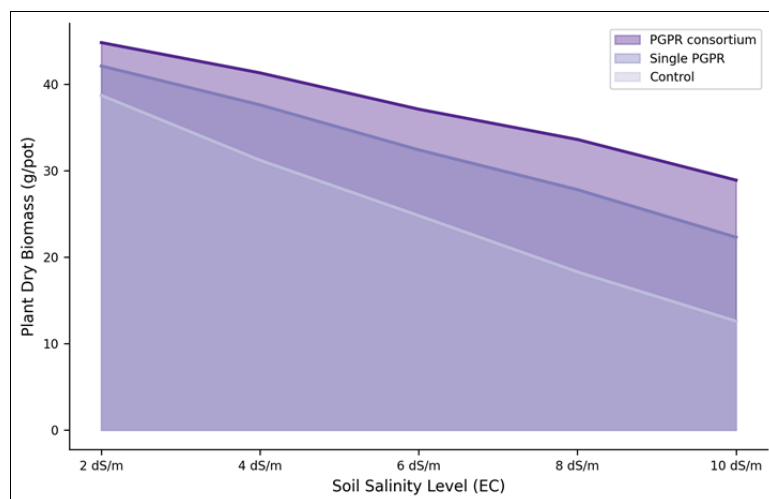


Fig 2: Plant dry biomass response to PGPR inoculation across five soil salinity levels

Comprehensive Interpretation

The consortium's advantage over the single strain grew wider as salinity increased. At 2 dS/m, the consortium produced only 6.4% more biomass than the single strain, but at 10 dS/m that gap widened to 29.6%. Nitrogen availability under the consortium followed a steep upward curve through week 8 before levelling off, suggesting peak bacterial activity occurred during the vegetative growth period. The salinity-inoculation interaction was significant ($p < 0.01$) for all nutrient parameters.

Discussion

The consortium's superior performance over the single *Bacillus* strain confirms that functional complementarity among PGPR species matters, especially under stress [6, 9]. *B. subtilis* contributes mainly through phosphorus solubilization and biofilm formation, while *P. fluorescens* adds siderophore production and *A. brasilense* provides nitrogen fixation [7]. Together, they cover more of the nutrient mobilization spectrum than any single species could alone.

The persistent root colonization above 10^5 CFU/g even at 10 dS/m is encouraging and likely reflects the pre-selection of salt-tolerant strains isolated from saline environments near Yancheng [14]. Many earlier PGPR trials failed at high salinity because the inoculant strains were originally isolated from non-saline soils and couldn't survive [8, 10]. The 197% increase in available nitrogen at 6 dS/m after 12 weeks suggests that biological nitrogen fixation and organic matter mineralization by the consortium were substantial contributors to the soil N pool [3, 16]. The biomass response plateauing at 10 dS/m indicates that PGPR can reduce but not eliminate salinity damage. At the highest salt concentration, osmotic stress and ion toxicity still limited growth despite improved nutrient supply [4, 5]. This sets a practical boundary for PGPR application they work best as part of an integrated approach rather than a standalone solution for severely saline soils [11, 12].

Conclusions

This pot research at Nanjing Agricultural University demonstrated that a four-species PGPR consortium significantly improved nutrient availability and wheat growth across a salinity gradient of 2 to 10 dS/m. The consortium outperformed single-strain inoculation at every salinity level, with the advantage increasing as salt stress intensified. Available nitrogen and phosphorus nearly tripled under consortium treatment at moderate salinity compared to uninoculated controls. Root

colonization remained viable at all tested salinity levels, confirming the practical persistence of these salt-adapted strains. However, PGPR alone couldn't fully overcome growth suppression at 10 dS/m, pointing to the need for combined approaches. Farmers managing moderately saline soils in the Jiangsu coastal region and similar areas should consider PGPR consortium inoculation as a low-cost biological tool to supplement chemical amendments. Future work should test these consortia under field conditions across multiple crop seasons to confirm greenhouse findings.

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