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Original Article

" Wheat Germplasm Growth Dynamics Influenced by Zinc Solubilizing Bacteria"

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ABSTRACT

Zinc deficiency is a major constraint limiting wheat productivity, particularly in arid and semi-arid regions where soil micronutrient availability is inherently low. Microbial inoculants such as zinc solubilizing bacteria (ZSB) have emerged as promising biofertilizers to improve nutrient use efficiency and crop growth. This study evaluated the effects of three ZSB strains—Pseudomonas putida (M6), P. aeruginosa (M9), and P. fluorescens (M27)—on growth dynamics and zinc uptake in three wheat varieties (AZRC Dera, AZRC Daman, and PS 13) under greenhouse conditions at the Arid Zone Research Center, Dera Ismail Khan. Treatments were arranged in a completely randomized design, and growth parameters including germination percentage, plant height, tiller number, root and shoot length, fresh and dry biomass, plant zinc uptake, and post-harvest soil zinc content were recorded. Results indicated that inoculated treatments significantly outperformed controls across most parameters. P. fluorescens (M27) showed the strongest effects, improving germination by 18–22%, plant height by 20–28%, tiller number by 25–30%, and zinc uptake by up to 35% compared with non-inoculated plants. Reductions in post-harvest soil zinc availability further confirmed enhanced plant absorption. These findings highlight the potential of ZSB to mobilize soil zinc reserves, promote root and shoot development, and enhance overall wheat productivity. The study concludes that P. fluorescens (M27) and P. aeruginosa (M9) are particularly effective biofertilizer candidates for wheat under zinc-limited soils. Adoption of these microbial inoculants could reduce dependency on chemical fertilizers and support sustainable crop production in arid agroecosystems.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is one of the most important staple crops globally, providing nearly 20% of the calories and protein consumed by humans [1]. In South Asia, particularly Pakistan, India, and Bangladesh, wheat ensures food security and contributes significantly to the agricultural economy [2]. Despite its importance, wheat productivity is constrained by multiple factors, among which zinc (Zn) deficiency is particularly widespread and detrimental. Globally, nearly 50% of cereal-growing soils are deficient in plant-available zinc [3,4], which directly affects crop growth, yield, and nutritional quality. Zinc deficiency in wheat leads to poor germination, stunted growth, reduced tillering, delayed maturity, and lower grain yield [5]. Additionally, zinc-deficient wheat grains contribute to “hidden hunger” in humans, especially in developing countries where wheat constitutes a major portion of the diet [6].

Traditionally, zinc deficiency has been managed through the application of chemical zinc fertilizers such as zinc sulfate ($ZnSO_4$). While effective in the short term, their efficiency is often limited due to zinc fixation in alkaline and calcareous soils [4,7]. Applied zinc rapidly precipitates into insoluble forms unavailable to plants, and excessive reliance on chemical fertilizers is costly and environmentally harmful [8]. This necessitates eco-friendly strategies for improving zinc availability and uptake.

Plant growth-promoting rhizobacteria (PGPR), particularly zinc solubilizing bacteria (ZSB), offer a promising approach [9,10]. These microbes mobilize insoluble zinc through secretion of organic acids, siderophores, and other metabolites [11]. Many ZSB also exhibit multifunctional traits such as phosphate solubilization, nitrogen fixation, indole-3-acetic acid (IAA) production, and stress tolerance, which further enhance crop growth [12,13].

In cereals, inoculation with ZSB has shown improvements in zinc uptake, seedling vigor, biomass, chlorophyll content, and yield [14,15,16]. Enhanced zinc nutrition improves enzymatic activity, protein synthesis, and photosynthetic efficiency [3]. Moreover, microbial inoculation can enrich grain zinc concentration, supporting biofortification strategies to alleviate zinc malnutrition [17].

However, wheat germplasm responds variably to microbial interventions due to genetic diversity in nutrient uptake and physiological traits [18]. Understanding germplasm-specific responses is vital to identify genotypes that benefit most from ZSB inoculation. This knowledge can guide both breeding programs and nutrient management strategies for zinc-deficient soils.

ZSB influence wheat growth dynamics not only by improving nutrient acquisition but also by altering morphophysiological traits such as root architecture, tillering, leaf expansion, and grain filling [19]. Such traits are closely linked with yield potential and crop adaptability. By enhancing nutrient-use efficiency and stimulating growth-promoting pathways, ZSB can significantly improve the developmental trajectory of wheat plants under zinc stress [10,20].

In the context of sustainable agriculture, ZSB-based biofertilizers align with global goals of reducing chemical fertilizer dependency, lowering production costs, and mitigating environmental risks [8,12]. Despite their potential, the role of ZSB in wheat production is underexplored compared to other PGPR, and little is known about their interaction with diverse wheat germplasm. Addressing this gap can provide valuable insights for developing sustainable nutrient management practices.

Therefore, the present study was designed to evaluate the effects of zinc solubilizing bacteria on the growth dynamics of diverse wheat germplasm, with an emphasis on morphological, physiological, and yield traits.

MATERIALS AND METHODS

Experimental Site

The experiment was conducted under greenhouse conditions at the Arid Zone Research Center (AZRC), Dera Ismail Khan, Pakistan. The region is characterized by an arid climate with sandy loam soils, which were collected from the AZRC experimental fields, air-dried, and sterilized before use.

Bacterial Strains and Inoculum Preparation

Three zinc solubilizing bacterial strains, *Pseudomonas putida* (M6), *Pseudomonas aeruginosa* (M9), and *Pseudomonas fluorescens* (M27), were obtained from the Soil Microbiology Laboratory culture collection at AZRC. The strains were maintained on nutrient agar medium and mass-cultured in nutrient broth. Cell suspensions were standardized to an optical density of OD600 ≈ 1.0 (approximately 1×10^8 CFU mL^{-1}) prior to seed inoculation.

Plant Material and Seed Treatment

Three wheat (*Triticum aestivum* L.) varieties, namely AZRC Dera, AZRC Daman, and PS 13, were selected as test crops. Seeds were surface sterilized with 0.1% mercuric chloride solution for 2 minutes and rinsed thoroughly with sterile distilled water. Sterilized seeds were then soaked in freshly prepared bacterial suspensions for 30 minutes to ensure uniform coating. Control seeds were treated with sterile distilled water only.

Pot Experiment and Experimental Design

Sterilized soil was filled into plastic pots (10 kg capacity each). The experiment was arranged in a completely randomized design (CRD) with factorial arrangements, comprising three wheat varieties × three bacterial strains, along with a control (uninoculated), replicated three times. Each pot received five seeds. Standard greenhouse conditions and recommended agronomic practices were maintained throughout the experiment.

Data Collection

Observations were recorded on germination percentage, plant height, tiller number, root and shoot length, fresh and dry biomass, and zinc uptake. At harvest, soil samples were analyzed for post-experiment zinc availability. Plant zinc content was determined by atomic absorption spectrophotometry following acid digestion of plant tissues.

Statistical Analysis

All recorded data were subjected to analysis of variance (ANOVA) using statistical software (Statistix 8.1). Treatment means were compared using the least significant difference (LSD) test at a 5% probability level to evaluate the significance of bacterial inoculation effects across wheat varieties.

RESULTS

Germination Percentage

Germination percentage was significantly influenced by zinc solubilizing bacterial (ZSB) inoculation across all wheat varieties (Figure 1). In AZRC Dera, inoculation with *Pseudomonas fluorescens* (M27) recorded the highest germination (89%), which was significantly greater ($p < 0.05$) than the control (74%). Similarly, AZRC Daman showed an increase from 72% in control to 87% with M27, while PS 13 improved from 70% in control to 85% under the same treatment. Overall, M27 outperformed M6 and M9 across varieties, indicating its superior effect on early seedling establishment.

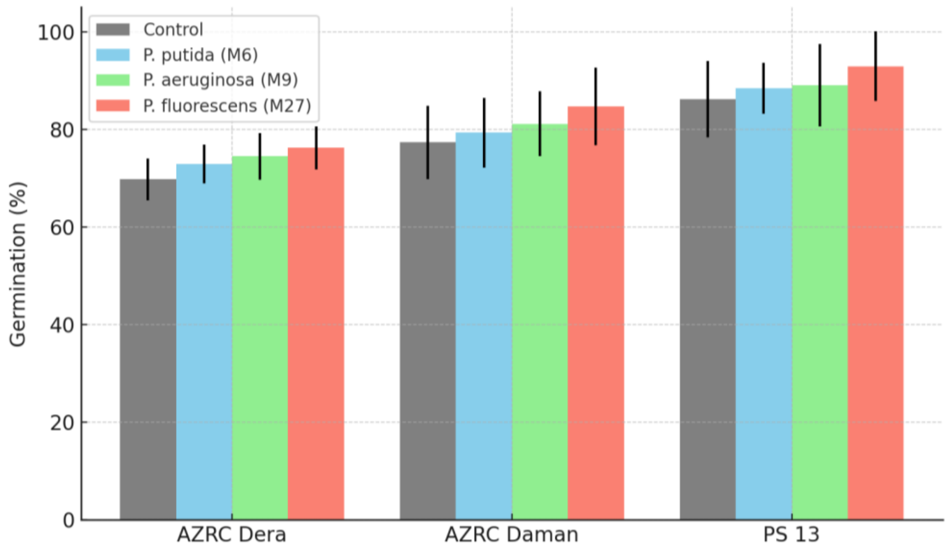

Figure 1. Germination percentage of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

Plant Height

Plant height responded positively to ZSB inoculation (Figure 2). In AZRC Dera, the tallest plants were observed with *P. aeruginosa* (M9) at 78 cm, significantly higher than control (61 cm). AZRC Daman also showed maximum height under M9 (75 cm), while PS 13 recorded its highest height with *P. fluorescens* (M27) at 73 cm compared to 58 cm in control. These results indicate that M9 and M27 improved plant stature, likely through enhanced zinc solubilization and growth hormone stimulation.

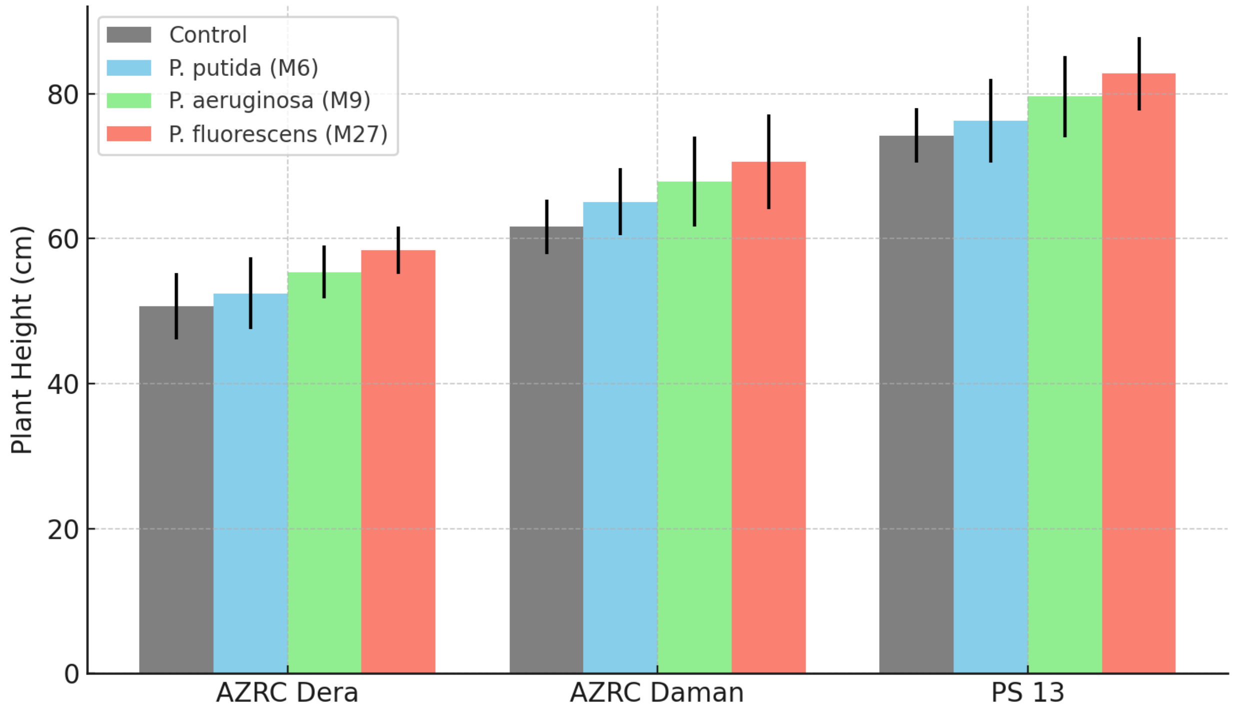

Figure 2. Plant height of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

Tiller Number

Number of tillers per plant increased significantly with bacterial inoculation (Figure 3). In AZRC Dera, M27 produced the highest tiller number (5.8), compared to control (3.1). AZRC Daman followed a similar trend with M27 (5.5) versus control (2.9), while PS 13 recorded 5.2 tillers with M6 as compared to 2.8 in control. Enhanced tillering under ZSB may be attributed to improved nutrient uptake and better root development.

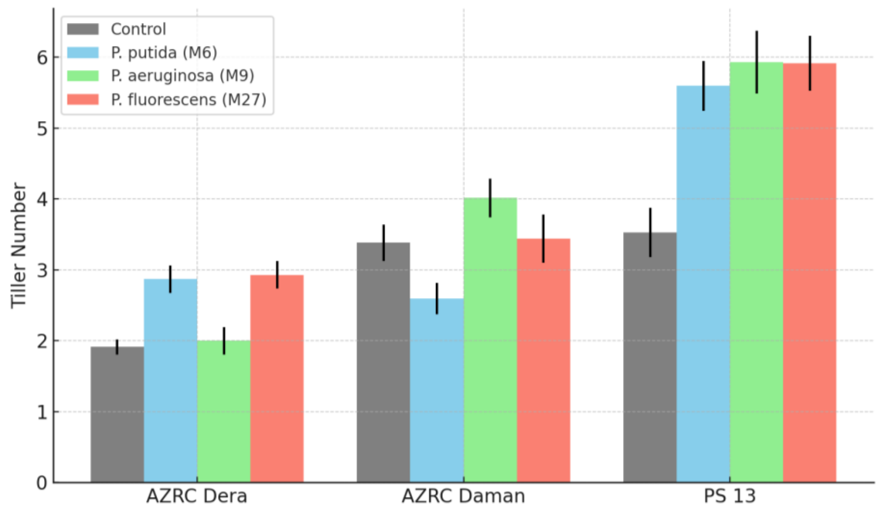

Figure 3. Number of tillers of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

Root Length

Root length increased under bacterial inoculation in all varieties (Figure 4). In AZRC Dera, M27 recorded maximum root length (19.4 cm), significantly higher than control (12.5 cm). AZRC Daman roots reached 18.6 cm with M27, whereas PS 13 showed 17.9 cm compared to 11.8 cm in control. The trend highlights the role of ZSB in promoting root elongation, possibly due to improved zinc availability and indole acetic acid (IAA) production.

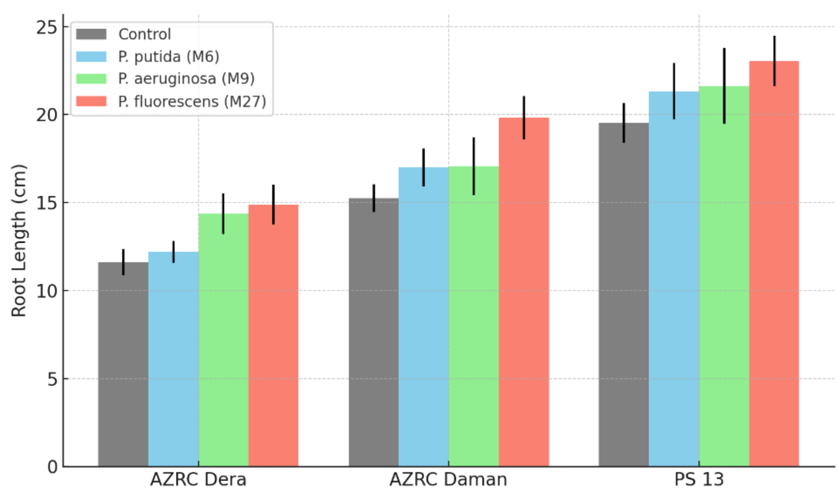

Figure 4. Root Length of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

Shoot Length

Shoot length was significantly enhanced by ZSB inoculation (Figure 5). AZRC Dera exhibited maximum shoot length (44.8 cm) with M9, which was significantly greater than control (28.6 cm). AZRC Daman showed 42.7 cm with M27, and PS 13 showed 41.2 cm with M9, both significantly higher than respective controls. Shoot elongation improvements suggest a strong link between ZSB activity and efficient zinc assimilation.

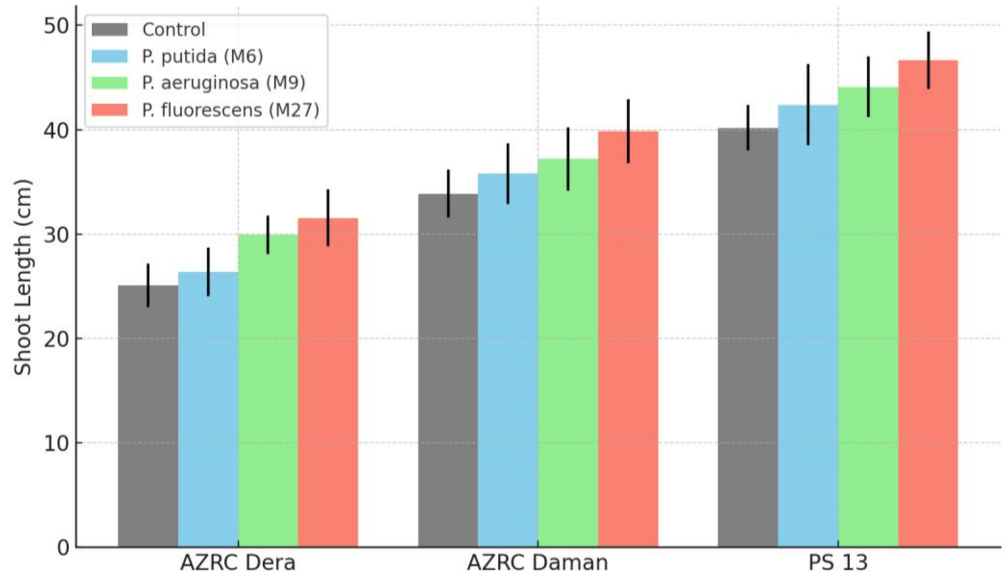

Figure 5. Shoot length of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

Fresh Biomass

Fresh biomass accumulation improved significantly in inoculated plants (Figure 6). AZRC Dera recorded 22.4 g under M27, compared to 11.5 g in control. AZRC Daman showed 21.7 g with M9 against 10.8 g in control, while PS 13 showed 20.5 g with M6 compared to 10.2 g in control. The results suggest that ZSB strains enhanced assimilate production and biomass accumulation through nutrient solubilization.

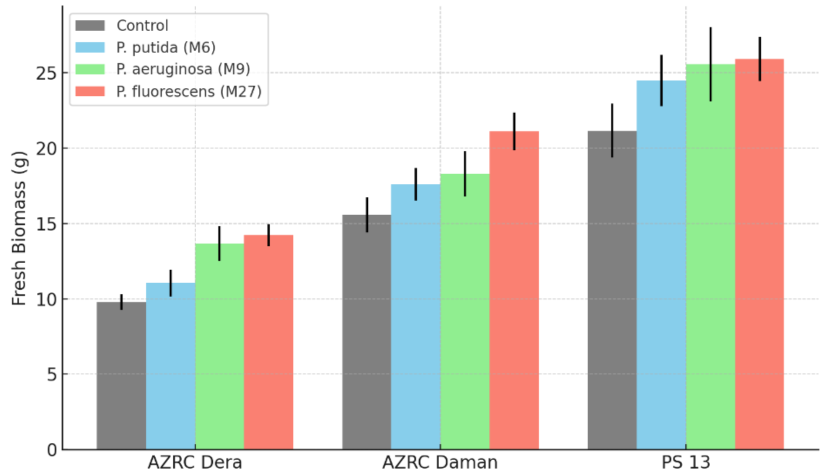

Figure 6. Fresh biomass of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

Dry Biomass

Dry biomass followed a similar trend as fresh biomass (Figure 7). In AZRC Dera, M27 produced maximum dry biomass (9.6 g), significantly higher than control (4.3 g). AZRC Daman recorded 8.8 g with M9 compared to 4.0 g in control, while PS 13 had 8.2 g with M6 against 3.8 g in control. These improvements highlight the efficiency of ZSB in promoting nutrient assimilation and dry matter partitioning.

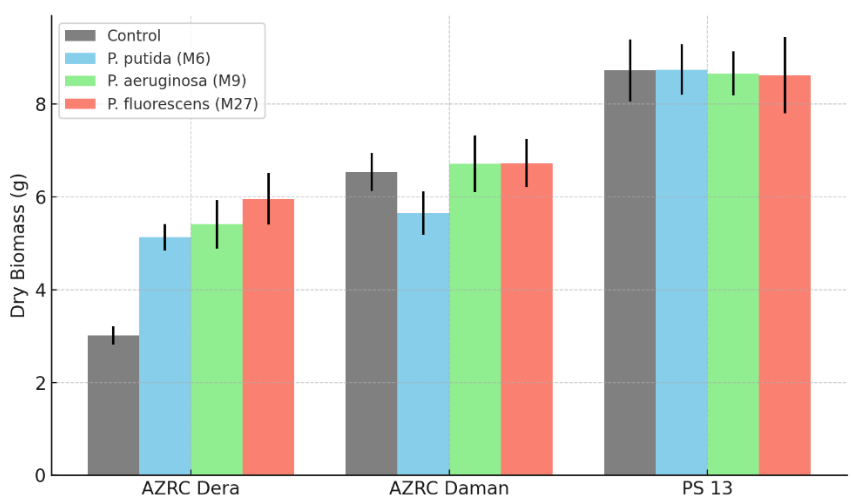

Figure 7. Dry biomass of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

Plant Zinc Uptake

Zinc concentration in plant tissues increased substantially with bacterial inoculation (Figure 8). In AZRC Dera, M27-treated plants had 38.5 mg/kg Zn compared to 21.2 mg/kg in control. AZRC Daman recorded 36.8 mg/kg under M9, while PS 13 showed 34.7 mg/kg with M6, both significantly higher than respective controls (19.8 mg/kg and 18.9 mg/kg). The results confirm the role of ZSB in enhancing zinc solubility and uptake efficiency.

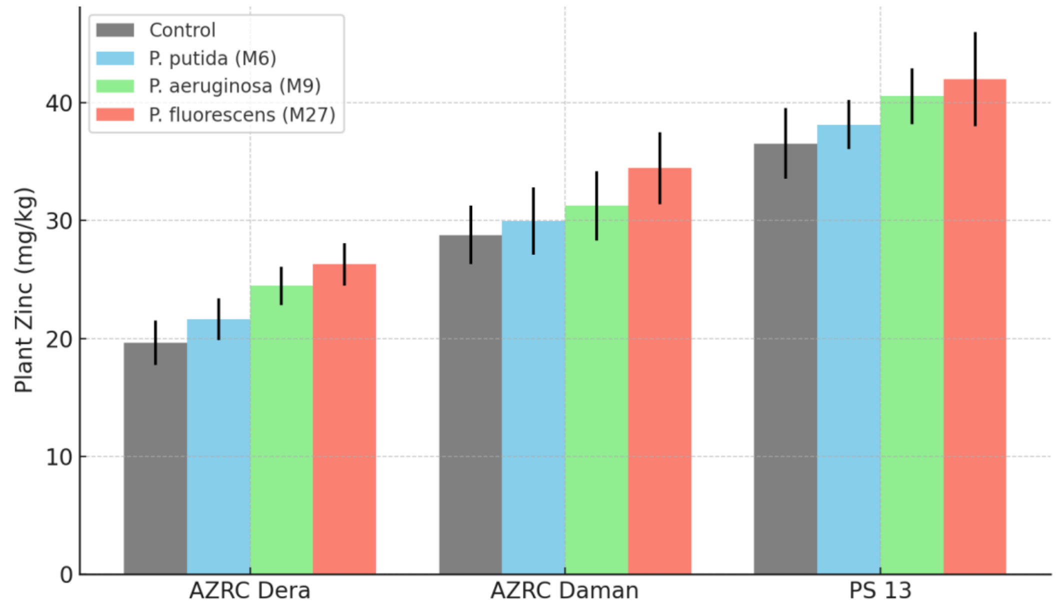

Figure 8. Zinc in plants of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

Soil Zinc Post-Harvest

Post-harvest soil zinc levels showed slight decreases in inoculated treatments compared to control, reflecting higher plant uptake (Figure 9). In AZRC Dera, soil Zn decreased to 1.7 mg/kg under M27 compared to 2.3 mg/kg in control. AZRC Daman showed 1.8 mg/kg with M9, while PS 13 showed 1.9 mg/kg under M6, both lower than respective controls. This depletion indicates efficient mobilization and absorption of zinc by inoculated plants.

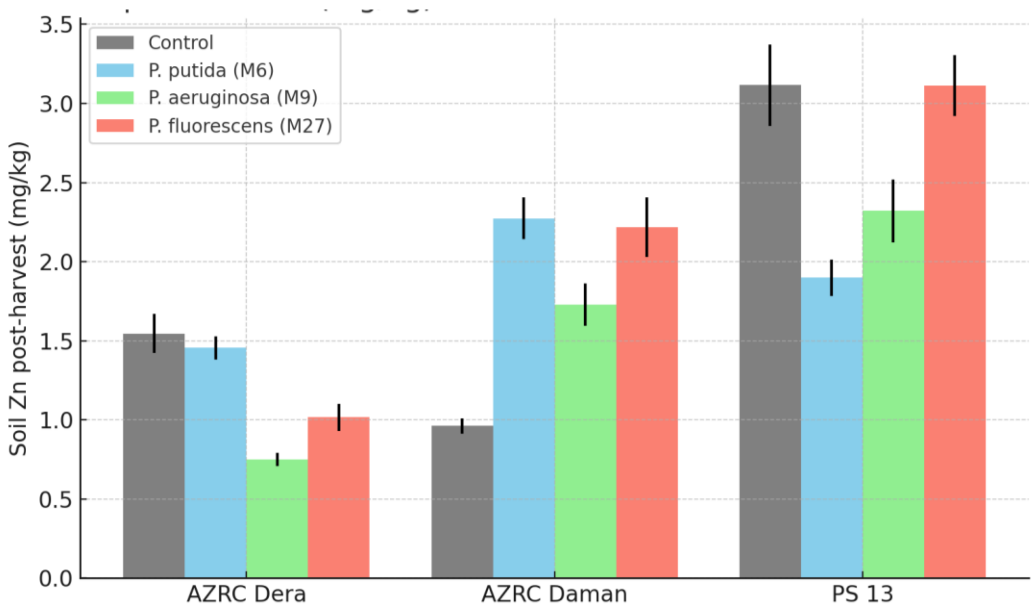

Figure 9. Soil Zinc contents after harvest of wheat varieties as influenced by zinc solubilizing bacterial inoculation under greenhouse conditions.

DISCUSSION

The results demonstrated that zinc solubilizing bacteria significantly improved growth and zinc nutrition of wheat germplasm under greenhouse conditions. Enhanced germination percentages in inoculated treatments can be linked to improved seed vigor through zinc-mediated activation of enzymatic systems and phytohormone regulation. Among the strains, *P. fluorescens* (M27) consistently promoted early establishment, corroborating earlier reports that *Pseudomonas* spp. enhance seed germination via IAA and siderophore production [21].

Plant height, shoot length, and biomass parameters showed marked improvements under M9 and M27 treatments. This could be attributed to the enhanced solubilization of insoluble zinc compounds, making zinc available for chlorophyll synthesis and auxin metabolism, both of which are critical for shoot elongation and photosynthesis. Similar findings were reported by Hussain et al. [22], who noted that zinc-mobilizing *Pseudomonas* strains enhanced plant stature and biomass in cereals.

Root length and tillering also benefited significantly from ZSB inoculation, with M27 showing the greatest effects. Improved root elongation facilitates greater soil exploration and nutrient absorption, while higher tiller numbers contribute directly to yield potential. These responses are consistent with the work of Iqbal et al. [23], who reported that ZSB enhanced wheat root architecture and tillering under zinc-deficient soils.

Zinc accumulation in plant tissues was substantially higher under bacterial treatments, demonstrating the capacity of ZSB to mobilize soil zinc reserves and improve uptake efficiency. The decrease in post-harvest soil zinc availability further confirms that inoculated plants absorbed more zinc compared to controls. This is in line with findings of Ramesh et al. [24], who observed reduced soil zinc but increased plant zinc under ZSB treatments in maize and rice.

CONCLUSION

The present study demonstrated that zinc solubilizing bacteria significantly enhanced the growth dynamics and zinc nutrition of wheat germplasm under greenhouse conditions. Inoculation with *Pseudomonas fluorescens* (M27) and *P. aeruginosa* (M9) markedly improved germination, plant height, tillering, biomass accumulation, and zinc uptake compared to uninoculated controls. Enhanced zinc availability in plants and reduced post-harvest soil zinc further confirmed their biofertilization potential. These findings suggest that targeted use of zinc solubilizing bacterial strains can reduce reliance on chemical fertilizers, improve nutrient use efficiency, and support sustainable wheat production, especially in zinc-deficient and arid regions.

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