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Molecular Characterization of Phosphate Solubilizing Bacteria from Cereal Crop Rhizospheres: A Review

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Abstract

Phosphorus (P) is an essential macronutrient for plant growth, yet a large fraction of soil phosphorus remains unavailable due to its fixation in insoluble forms. Excessive reliance on chemical fertilizers to overcome this deficiency has raised serious economic and environmental concerns. Phosphate solubilizing bacteria (PSB) represent an eco-friendly and sustainable alternative, as they mobilize insoluble phosphates through mechanisms such as organic acid secretion, phosphatase activity, and siderophore production. Cereal crops, including rice, wheat, and maize, are particularly dependent on phosphorus for optimum productivity, making the rhizosphere of these crops an important niche for PSB research. Recent advances in molecular techniques such as 16S rRNA gene sequencing, functional gene analysis, metagenomics, and phylogenetic studies have enabled precise characterization of novel PSB strains and their functional attributes. This review highlights the diversity and mechanisms of PSB in cereal rhizospheres, with emphasis on molecular characterization under field conditions. Furthermore, it discusses their role in sustainable crop production, current challenges in large-scale application, and future prospects for integrating molecularly characterized PSB strains into biofertilizer technology. Overall, PSB hold significant promise for reducing chemical fertilizer dependency and improving global food security.

Keywords: *Phosphate solubilizing bacteria, Cereal crops, Rhizosphere, 16S rRNA, Metagenomics, Biofertilizer.*

1. Introduction

Phosphorus (P) is one of the most vital macronutrients required by plants and plays a central role in several biochemical and physiological processes, including energy transfer through ATP, photosynthesis, nucleic acid synthesis, signal transduction, and the regulation of metabolic pathways. Despite being the eleventh most abundant element in the Earth's crust, phosphorus is often a limiting nutrient in agricultural soils because more than 80–90% of total soil phosphorus exists in insoluble or poorly soluble forms bound with calcium, iron, or aluminum complexes. These immobile forms cannot be readily absorbed by plants, resulting in phosphorus deficiency that significantly restricts plant growth, root development, and ultimately crop yield [1]. To compensate for this deficiency, farmers worldwide depend heavily on chemical phosphate fertilizers. While these inputs can temporarily replenish plant-available phosphorus, their long-term use has raised several concerns. Excessive application not only escalates the cost of crop production but also contributes to nutrient leaching, eutrophication of aquatic systems, and degradation of soil health [2]. Furthermore, the global reserves of rock phosphate, the primary raw material for synthetic fertilizers, are finite and projected to decline in the coming decades. These challenges highlight the urgent need for sustainable alternatives to ensure long-term agricultural productivity and environmental protection.

One promising strategy is the utilization of phosphate solubilizing bacteria (PSB) as biofertilizers. PSB are a diverse group of soil microorganisms that have the natural ability to mobilize insoluble phosphorus into plant-available forms. They achieve this primarily through the secretion of organic acids such as gluconic acid, oxalic acid, and citric acid, which lower the soil pH and chelate cations bound to phosphate. In addition, PSB produce extracellular enzymes like phosphatases and phytases that hydrolyze organic phosphorus compounds, thereby expanding the pool of available phosphorus in the rhizosphere [3]. Other mechanisms include the release of siderophores and protons, which further contribute to solubilization processes.

By increasing phosphorus bioavailability, PSB enhance root proliferation, nutrient uptake, and crop productivity while simultaneously reducing dependence on chemical fertilizers.

The rhizosphere of cereal crops represents a particularly important niche for isolating and characterizing PSB. Cereal crops such as rice, wheat, maize, barley, and sorghum are staple foods for the majority of the global population and account for a large proportion of the world's phosphorus fertilizer demand. These crops require high levels of phosphorus to support processes like tillering, grain filling, and energy metabolism [4]. As a result, their rhizospheres harbor diverse microbial communities that have adapted to phosphorus-limiting conditions. Studying and characterizing PSB from cereal rhizospheres under field conditions not only improves our understanding of microbial ecology but also offers practical solutions for developing targeted biofertilizers to enhance sustainable cereal production, molecular characterization techniques have revolutionized the study of PSB diversity and functionality. Traditional culture-dependent methods, while useful for isolating individual strains, capture only a small fraction of microbial diversity. Many microorganisms remain uncultivable under laboratory conditions, making it difficult to fully understand the complexity of PSB communities. Molecular tools such as 16S rRNA gene sequencing, DNA fingerprinting techniques (RAPD, RFLP, and AFLP), and metagenomic approaches now allow precise identification and classification of bacterial taxa. Functional gene markers such as *pqq*, *gcd* (gluconic acid dehydrogenase), and *phoD* (alkaline phosphatase) have been employed to confirm phosphate solubilizing traits at the genetic level [5]. Metagenomics and next-generation sequencing have further enabled the exploration of uncultured microbial populations, revealing novel PSB taxa with unique solubilization mechanisms.

Molecular characterization not only enhances taxonomic resolution but also provides insight into the metabolic potential, ecological adaptation, and symbiotic interactions of PSB with host plants. For example, PSB belonging to genera such as *Pseudomonas*, *Bacillus*, *Rhizobium*, *Enterobacter*, and *Burkholderia* have been frequently reported from cereal rhizospheres and have shown significant variation in their phosphate solubilization efficiency. Molecular tools allow researchers to compare these strains, identify novel isolates, and assess their stability and efficiency under diverse field conditions [6]. Field-based studies are particularly important because laboratory performance of PSB often does not translate directly into field success due to variations in soil properties, climatic conditions, and microbial competition. Evaluating PSB under natural conditions ensures the selection of robust strains capable of maintaining their phosphate solubilization activity in real agricultural systems. Molecular markers combined with ecological assessments can help identify strains with consistent performance, paving the way for their integration into biofertilizer formulations.Moreover, PSB offer additional plant growth-promoting benefits beyond phosphorus solubilization. Many isolates produce phytohormones such as indole-3-acetic acid (IAA), gibberellins, and cytokinins, which stimulate root elongation and improve nutrient acquisition. Some also exhibit biocontrol properties by suppressing plant pathogens through the production of antibiotics, siderophores, or lytic enzymes. These multifunctional attributes make PSB valuable candidates for inclusion in integrated nutrient management strategies, challenges remain in harnessing the full potential of PSB. The variability in field performance, lack of standardized molecular markers for functional characterization, and limited understanding of microbial interactions in the rhizosphere are significant bottlenecks. Furthermore, the scalability of PSB-based biofertilizers requires rigorous quality control, formulation stability, and farmer acceptance [7]. To address these gaps, interdisciplinary research combining molecular microbiology, soil science, plant physiology, and biotechnology is essential, phosphorus deficiency is a major constraint in global cereal production, and phosphate solubilizing bacteria offer a sustainable and eco-friendly alternative to chemical fertilizers. The molecular characterization of PSB isolated from cereal crop rhizospheres provides critical insights into their diversity, genetic traits, and ecological functions [8]. Advances in molecular tools have opened new avenues for discovering novel PSB strains with enhanced efficiency under field conditions. Harnessing these microbial resources can significantly contribute to sustainable agriculture, reduce environmental impacts, and ensure food security for the growing global population.

Table 1. Mechanisms of Phosphate Solubilization by PSB

Mechanism	Example Compounds/Enzymes	Effect on Phosphate Availability	Representative Genera
Organic acid production	Gluconic acid, citric acid, oxalic acid	Chelation of Ca^{2+}, Fe^{3+}, Al^{3+} ions; release of soluble P	*Bacillus*, *Pseudomonas*, *Enterobacter*
Enzyme activity	Acid phosphatase, alkaline phosphatase	Hydrolysis of organic P compounds	*Rhizobium*, *Serratia*
Proton extrusion	H^+ release via respiration	Decrease in rhizosphere pH; enhanced solubilization	*Azotobacter*, *Klebsiella*
Siderophore production	Catecholate and hydroxamate siderophores	Chelation of Fe^{3+}, indirectly releasing bound P	*Pseudomonas*, *Burkholderia*

Table 2. Molecular Tools for Characterizing PSB

Technique	Purpose	Advantages	Limitations
16S rRNA sequencing	Taxonomic identification (genus/species)	High accuracy; universal marker	Cannot resolve strain-level diversity
Functional gene analysis	Detect genes (e.g., *gcd*, *ppx*)	Confirms P-solubilization traits	Limited to known genes
Metagenomics (NGS)	Culture-independent community profiling	Reveals rare/unculturable taxa	Expensive, requires bioinformatics
qPCR and RT-PCR	Quantify expression of functional genes	Monitors gene activity under field conditions	Requires prior gene knowledge

Table 3. Dominant PSB Reported in Cereal Crop Rhizospheres

Cereal Crop	Dominant PSB Genera	Functional Traits	Reported Impact on Crop Growth
Rice (*Oryza sativa*)	*Pseudomonas*, *Bacillus*	P solubilization, N fixation, siderophore production	↑ Yield by 20–25%
Wheat (*Triticum aestivum*)	*Enterobacter*, *Rhizobium*	Dual PGPR traits, enzyme secretion	↑ Grain weight, ↑ root biomass
Maize (*Zea mays*)	*Bacillus*, *Klebsiella*	Organic acid production, strong acidification	↑ P availability, ↑ shoot length

Table 4. Applications and Challenges of PSB in Agriculture

Applications	Examples	Challenges
Biofertilizers as P-supplements	PSB inoculants for rice, wheat, maize	Strain specificity, limited shelf life
Bioremediation of P-deficient soils	Mixed microbial consortia	Soil pH and temperature sensitivity
Synergistic use with other PGPR (e.g., N-fixers)	*Rhizobium* + *Bacillus* consortia	Variability in field efficacy
Reduction of chemical fertilizer dependency	PSB formulations in India & China	Regulatory approval, commercialization hurdles

2. Importance of Phosphorus in Cereal Crop Productivity

Phosphorus (P) is one of the most essential macronutrients required for the growth and productivity of plants. It is a structural component of nucleic acids, phospholipids, and ATP, and it plays a central role in energy transfer, signal transduction, and metabolic regulation. Among field crops, cereals such as rice (*Oryza sativa*), wheat (*Triticum aestivum*), and maize (*Zea mays*) are the most important as they contribute over 60% of the global calorie intake [9]. Ensuring an adequate phosphorus supply is therefore critical not only for crop yield but also for global food and nutritional security.

Role of Phosphorus in Plant Growth and Cereal Physiology

Phosphorus is indispensable during several stages of cereal crop growth. In the early vegetative phase, it promotes vigorous root development, which is essential for water and nutrient acquisition. Adequate phosphorus availability in this stage improves tillering in rice and wheat and enhances canopy expansion, which in turn contributes to higher photosynthetic efficiency. During the grain filling stage, phosphorus supports carbohydrate metabolism, ATP synthesis, and translocation of sugars from source to sink tissues. As a result, cereals with sufficient phosphorus supply develop fuller grains with higher starch content, thereby increasing both yield and quality [10]. Phosphorus deficiency, on the other hand, results in characteristic symptoms such as stunted growth, purpling of leaves due to anthocyanin accumulation, delayed maturity, poor seed set, and reduced biomass. In cereals like maize, deficiency leads to weak root systems and increased susceptibility to drought stress. In rice, phosphorus deficiency hampers tillering and panicle initiation, leading to sharp declines in grain yield.

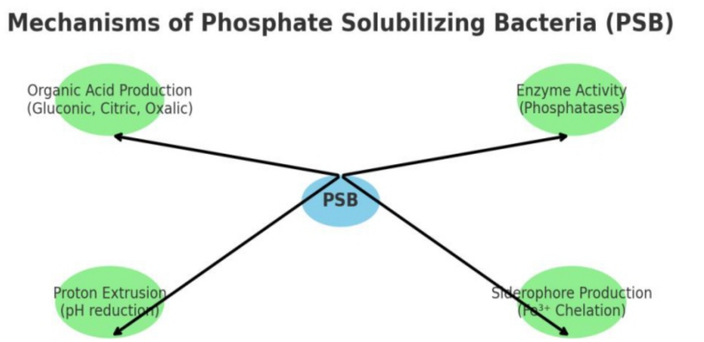

Fig 1: *Phosphate solubilizing bacteria (PSB) enhance phosphorus availability to plants through multiple mechanisms. Organic acid secretion (e.g., gluconic, citric, and oxalic acids) chelates metal cations (Ca^{2+}, Fe^{3+}, Al^{3+}) bound to insoluble phosphates, releasing soluble forms.*

Global Demand and Fertilizer Dependency

The intensification of agriculture over the past five decades has drastically increased the demand for phosphorus fertilizers. According to global projections, fertilizer requirements will continue to rise due to population growth, dietary shifts, and the need for higher crop productivity. Asia and Africa are projected to experience the steepest increases in phosphorus demand because they harbor some of the most rapidly growing populations and depend heavily on cereal-based diets [11]. However, the use of chemical phosphate fertilizers comes with multiple challenges. First, phosphorus fertilizers are derived from finite rock phosphate reserves, which are unevenly distributed globally and concentrated in only a few countries. This raises concerns about long-term sustainability and price volatility. Second, phosphate fertilizers are highly inefficient in soils; nearly 70–80% of applied phosphorus becomes fixed due to interactions with calcium, aluminum, or iron compounds, making it unavailable for plant uptake. This not only increases fertilizer costs for farmers but also results in environmental issues such as eutrophication of water bodies due to phosphorus runoff.

Regional Context: India and Other Developing Nations

In countries like **India**, where cereal production underpins national food security, phosphorus deficiency is a major constraint to crop productivity. Many Indian soils are characterized by low available phosphorus due to high fixation rates, particularly in acidic and alkaline soils. Studies have reported that in intensive rice-wheat cropping systems, phosphorus depletion rates are extremely high because of continuous removal through grain harvests. To sustain yields, farmers often resort to higher fertilizer application, which escalates production costs and increases environmental burden [12]. Similarly, in African nations where fertilizer use is already low due to high prices and limited access, phosphorus deficiency contributes significantly to yield gaps. Enhancing phosphorus availability through alternative approaches is thus essential for both continents.

PSB-Based Biofertilizers as a Sustainable Alternative

Phosphate solubilizing bacteria (PSB) offer a promising eco-friendly and cost-effective solution to the phosphorus challenge in cereal production. Unlike chemical fertilizers, which add external phosphorus to the system, PSB mobilize native soil phosphorus that is otherwise locked in insoluble forms. These bacteria release organic acids, phosphatases, and other metabolites that transform unavailable phosphates into soluble forms readily absorbed by plant roots [13]. Field studies in rice, wheat, and maize have shown that inoculation with PSB enhances phosphorus uptake, promotes better root growth, and increases grain yield. For instance, *Pseudomonas* and *Bacillus* strains have been reported to improve phosphorus acquisition in wheat under both irrigated and rainfed conditions. In rice ecosystems, PSB consortia applied along with reduced fertilizer doses have resulted in yields comparable to or higher than those achieved with full chemical fertilizer doses, thereby lowering input costs for farmers [14]. PSB inoculation has multifunctional benefits beyond phosphorus solubilization. Many strains produce phytohormones, fix atmospheric nitrogen, or enhance tolerance against abiotic stresses such as drought and salinity. In maize, certain PSB strains have been shown to enhance root hair density and length, thereby improving overall nutrient and water uptake efficiency.

Food Security and Future Perspectives

With global population expected to exceed 9 billion by 2050, cereal production must increase substantially to meet food demands. However, achieving this solely through chemical fertilizers is neither economically feasible nor environmentally sustainable. Incorporating PSB-based biofertilizers into integrated nutrient management systems can help bridge the gap between productivity and sustainability.PSB application can also contribute to soil health restoration. Unlike chemical fertilizers that often degrade soil microbial diversity, PSB enrich the microbial community and improve nutrient cycling [15]. This is particularly important in regions where intensive cereal cultivation has led to declining soil fertility and reduced responsiveness to fertilizers, the use of molecular techniques to characterize and select high-performing PSB strains from cereal rhizospheres opens new opportunities for crop-specific biofertilizer formulations. Such targeted approaches can enhance adoption by farmers and provide consistent results under field conditions.Phosphorus is indispensable for cereal crop growth and productivity, yet its availability in soils remains a major constraint due to fixation and inefficient use of chemical fertilizers. Rising global demand for cereals, coupled with limitations in phosphorus fertilizer supply, underscores the urgent need for sustainable alternatives. Phosphate solubilizing bacteria represent a promising strategy to improve phosphorus availability, reduce fertilizer dependency, and ensure long-term food security, particularly in phosphorus-deficient regions of Asia and Africa [16]. By integrating PSB into agricultural systems, we can move toward more resilient, productive, and environmentally friendly cereal production systems.

3. Phosphate Solubilizing Bacteria: Mechanisms of Action

Phosphate solubilizing bacteria (PSB) are among the most important plant growth-promoting rhizobacteria (PGPR) that enhance nutrient availability, particularly phosphorus, in agricultural soils. Their ability to mobilize insoluble forms of phosphorus into bioavailable orthophosphate makes them invaluable as biofertilizers. Several interrelated mechanisms contribute to their solubilization potential, ranging from organic acid secretion to enzyme production [17]. Understanding these processes is crucial for developing efficient microbial formulations for field application.

3.1 Organic Acid Production

The primary mechanism by which PSB solubilize phosphate is through the secretion of low molecular weight organic acids such as **gluconic acid, citric acid, oxalic acid, lactic acid, malic acid, and succinic acid**. These acids act in two ways: they reduce soil pH by releasing protons (H^+), and they chelate cations such as calcium (Ca^{2+}), iron (Fe^{3+}), and aluminum (Al^{3+}) that are bound to phosphate molecules. This process converts insoluble phosphate compounds (e.g., tricalcium phosphate, iron phosphate, and aluminum phosphate) into soluble orthophosphate ions readily absorbed by plants. Among these acids, gluconic acid and citric acid are most commonly reported as dominant in phosphate solubilization by genera such as *Pseudomonas* and *Bacillus*[18].

3.2 Enzyme Activity

In addition to mineral phosphate solubilization, PSB play a vital role in mobilizing organic forms of phosphorus present in the soil, such as phytates, nucleic acids, and phospholipids. This is achieved through the secretion of phosphatase enzymes, primarily acid phosphatases, alkaline phosphatases, and phytases. These enzymes hydrolyze complex organic phosphorus compounds into simpler inorganic forms that plants can utilize. For example, *Rhizobium* and *Enterobacter* strains have been reported to produce high levels of phytase, which is particularly significant in soils rich in phytate-bound phosphorus [2]. The dual capacity of PSB to act on both inorganic and organic phosphorus pools enhances their efficiency as biofertilizers.

3.3 Proton Extrusion and Rhizosphere Acidification

Another important mechanism is proton extrusion associated with bacterial respiration and metabolism. Many PSB release H^+ ions during the assimilation of ammonium or organic carbon sources [3]. This acidification of the rhizosphere microenvironment enhances the solubilization of insoluble phosphates, complementing organic acid activity. Studies on *Serratia* and *Pseudomonas* strains have shown that significant decreases in rhizosphere pH correlate directly with increases in soluble phosphorus content, confirming the importance of this process.

3.4 Siderophore Production

Siderophores are high-affinity iron-chelating compounds secreted by many soil bacteria, including *Pseudomonas*, *Bacillus*, and *Serratia*. While their primary role is in iron acquisition, siderophores also indirectly contribute to phosphorus solubilization. By binding Fe^{3+} ions in the soil, siderophores reduce the formation of insoluble iron-phosphate complexes, thereby releasing phosphate ions into the soil solution [4]. This dual functionality highlights the multifaceted benefits of PSB in nutrient cycling.

3.5 Prominent PSB Genera

Several bacterial genera have been identified as efficient phosphate solubilizers in cereal crop rhizospheres. Among them, Bacillus and Pseudomonas are the most extensively studied due to their robust adaptability, high metabolic activity, and ability to survive under diverse soil conditions. *Enterobacter*, *Rhizobium*, and *Serratia* also demonstrate strong solubilization capacities and additional plant growth-promoting traits such as nitrogen fixation, phytohormone production, and biocontrol properties 5]. The use of these genera in consortia often provides synergistic effects, leading to higher phosphate availability and improved crop performance.

4. Molecular Characterization of Phosphate Solubilizing Bacteria (PSB)

The characterization of phosphate solubilizing bacteria (PSB) is central to understanding their ecological diversity, metabolic potential, and application as biofertilizers. Traditional microbiological methods such as morphological, biochemical, and physiological assays have provided valuable insights, but they are insufficient to resolve the full spectrum of bacterial diversity in complex ecosystems such as cereal rhizospheres. Many PSB remain unculturable under standard laboratory conditions, making molecular approaches essential for precise identification and functional assessment [6]. Modern molecular biology and high-throughput sequencing techniques have therefore revolutionized PSB research, enabling deeper insights into their taxonomy, functional genes, and ecological dynamics.

4.1 16S rRNA Gene Sequencing

The 16S ribosomal RNA (rRNA) gene has long been considered the gold standard for bacterial taxonomy and phylogenetic analysis. Its conserved regions facilitate the design of universal primers, while hypervariable regions provide species- and genus-level resolution. Through 16S rRNA sequencing, researchers have identified diverse PSB genera such as *Bacillus*, *Pseudomonas*, *Enterobacter*, *Rhizobium*, and *Serratia* in cereal rhizospheres.In cereal crop studies, 16S sequencing not only enables species-level identification but also provides phylogenetic relationships, allowing scientists to cluster isolates into operational taxonomic units (OTUs).

For instance, comparative analysis of PSB communities in rice versus maize rhizospheres has shown that while *Bacillus* dominates in maize soils, *Pseudomonas* is often more abundant in rice rhizospheres. Such findings highlight the crop-specific microbial recruitment strategies driven by root exudates [7].The growing availability of curated databases such as SILVA and RDP has further strengthened the accuracy of PSB classification, while tools like QIIME and Mothur have facilitated community-level analyses [8]. Nevertheless, while 16S rRNA provides excellent taxonomic resolution, it cannot alone confirm functional traits related to phosphate solubilization.

4.2 Functional Gene Analysis

To complement taxonomic profiling, functional gene analysis is critical for confirming the **mechanistic basis of phosphate solubilization** in PSB. Several key genes have been identified and used as molecular markers:

- **gcd (glucose dehydrogenase):** This gene encodes a pivotal enzyme in the direct oxidation pathway, responsible for converting glucose into gluconic acid—a major organic acid linked to phosphate solubilization. The presence of gcd has been strongly correlated with high phosphate solubilization potential in genera such as *Enterobacter* and *Pseudomonas*.
- **ppx (exopolyphosphatase):** This gene regulates the degradation of polyphosphates into orthophosphate, directly enhancing the availability of soluble phosphorus. PSB strains harboringppx demonstrate superior mineral phosphate mobilization.
- **phoD, phoA, and phytase genes:** These encode alkaline phosphatases and phytases that hydrolyze organic phosphorus compounds, making them essential in soils rich in phytate-bound P.

PCR-based screening of these genes provides a rapid diagnostic tool for selecting potent PSB strains. Moreover, quantitative PCR (qPCR) enables the estimation of functional gene abundance in rhizosphere communities, linking molecular traits with field-level outcomes in phosphorus mobilization and cereal crop growth.

4.3 Metagenomics and Next-Generation Sequencing (NGS)

Culture-independent methods such as metagenomics and next-generation sequencing (NGS) have transformed PSB research by enabling the exploration of microbial communities beyond the limitations of cultivation [9]. High-throughput sequencing of environmental DNA provides a comprehensive picture of microbial diversity, functional capacity, and ecological interactions.

- **Metagenomics:** By sequencing total DNA from cereal rhizospheres, researchers can detect both culturable and unculturable PSB taxa. This approach reveals rare or novel PSB species that might otherwise be overlooked in culture-based studies.

Metagenomic data also provide insights into metabolic pathways associated with phosphate solubilization, such as organic acid biosynthesis and phosphatase activity.

- **Amplicon sequencing (16S rRNA + functional markers):** Targeted sequencing approaches allow for detailed community profiling across different soil types, crop varieties, and management practices. For example, NGS-based studies have shown how fertilizer regimes shift the relative abundance of PSB in wheat and rice rhizospheres.
- **Shotgun metagenomics:** Beyond taxonomy, this approach uncovers the **functional gene repertoire** of PSB communities. By mapping sequences to metabolic databases such as KEGG, researchers can identify the presence of gcd, ppx, phoD, and other genes at the community level.
- **Metatranscriptomics and proteomics:** Although still emerging, these approaches allow the study of **gene expression** and **protein profiles** in situ, offering dynamic insights into how PSB respond to environmental conditions in the rhizosphere.

5. PSB from Cereal Crop Rhizospheres under Field Conditions

The rhizosphere of cereal crops represents one of the most dynamic microbial niches, enriched with diverse microbial populations that directly influence plant growth and nutrient cycling [10]. Phosphate solubilizing bacteria (PSB) are among the most significant functional groups in these ecosystems, as they enhance phosphorus availability in soils where the majority of phosphorus exists in insoluble forms. Several field-based studies across agroecological zones have demonstrated the diversity, ecological significance, and plant growth-promoting effects of PSB in rice, wheat, and maize systems.

Rice Rhizosphere

Rice (*Oryza sativa*) is a staple food crop for more than half of the global population and has been a focal point of PSB research. Numerous studies have reported that rice rhizospheres are dominated by *Pseudomonas* and *Bacillus* species, which not only solubilize insoluble phosphate but also contribute additional benefits such as nitrogen fixation, siderophore production, and phytohormone (indole-3-acetic acid, IAA) synthesis. Field trials with *Pseudomonas fluorescens* strains have demonstrated significant increases in available phosphorus, which translated into improved root biomass and higher grain yields. Similarly, *Bacillus subtilis* inoculants have been shown to enhance phosphorus mobilization and reduce reliance on chemical fertilizers in paddy soils [11]. The anaerobic and waterlogged conditions typical of rice fields create unique selective pressures, favoring PSB that are both facultatively anaerobic and metabolically versatile.

Wheat Rhizosphere

Wheat (*Triticum aestivum*), a major cereal crop in temperate regions, has been associated with a highly diverse PSB community, particularly species belonging to *Enterobacter* and *Rhizobium*. These bacteria not only solubilize mineral and organic phosphates but also display dual plant growth-promoting traits such as nitrogen fixation, phytohormone production, and stress tolerance induction. Studies from India and the Middle East have reported that inoculation of wheat with *Enterobacter cloacae* and *Rhizobium leguminosarum* strains increased phosphorus uptake efficiency by over 25%, while simultaneously improving grain protein content [12]. The synergistic role of PSB with arbuscular mycorrhizal fungi (AMF) has also been highlighted in wheat systems, suggesting that microbial consortia could provide more consistent benefits than single-strain inoculants.

Maize Rhizosphere

In maize (*Zea mays*), PSB communities are characterized by the predominance of *Bacillus* and *Klebsiella* species. These bacteria exhibit strong acidification potential through the secretion of organic acids such as gluconic acid and citric acid, which effectively mobilize calcium-bound phosphates [13]. Field studies in sub-Saharan Africa and South America have reported that *Bacillus megaterium* inoculants increased maize grain yield by 15–20% compared to untreated controls, while *Klebsiella pneumoniae* strains enhanced phosphorus uptake under nutrient-deficient soils. The maize rhizosphere is particularly favorable for PSB proliferation due to its rich exudation of sugars, amino acids, and organic acids that serve as microbial substrates.

Field Performance and Variability

Several large-scale field trials have confirmed that PSB inoculation can increase available phosphorus in soils by 20–30% and significantly boost crop yields. However, the performance of PSB in the field remains inconsistent. Factors such as soil pH, temperature, moisture availability, and the composition of native microbial communities strongly influence inoculant success. For example, while *Bacillus*-based formulations have shown promising results in neutral to alkaline soils, their efficacy declines in acidic environments. Similarly, competition with native microbial populations often reduces the colonization efficiency of introduced PSB strains [14]. These challenges highlight the need for site-specific strain selection and formulation strategies to maximize field performance.

6. Applications and Challenges of PSB in Agriculture

The increasing global demand for sustainable agricultural practices has placed PSB at the forefront of biofertilizer development [16]. Their ability to solubilize phosphorus, coupled with other plant growth-promoting traits, makes them attractive candidates for multiple agricultural applications.

However, their widespread adoption is constrained by several challenges that must be addressed to ensure consistency and scalability.

6.1 Applications of PSB in Agriculture

1.Biofertilizer Development: PSB-based biofertilizers represent a cost-effective and eco-friendly alternative to chemical phosphate fertilizers. When applied as seed treatments, soil amendments, or root inoculants, PSB enhance the bioavailability of phosphorus, reduce input costs, and minimize the environmental risks associated with phosphate runoff. Several commercial formulations, such as *Bacillus subtilis* and *Pseudomonas putida*-based biofertilizers, are already marketed in Asia and Latin America, though their use is still limited compared to chemical fertilizers.

2. Bioremediation of Phosphorus-Deficient Soils: In nutrient-poor soils, PSB can mobilize residual phosphorus pools, thereby rehabilitating degraded lands. This application is particularly relevant in tropical and subtropical regions where phosphorus deficiency is a major constraint to cereal productivity.

3. Synergistic Use in Microbial Consortia: Recent research emphasizes the advantages of microbial consortia, where PSB are combined with nitrogen-fixing bacteria, potassium solubilizers, and mycorrhizal fungi. These consortia provide holistic plant growth support, balancing multiple nutrient cycles while enhancing resilience against biotic and abiotic stresses. For instance, PSB co-inoculated with *Azotobacter* and *Glomus* spp. have demonstrated improved nutrient uptake efficiency and yield stability in rice–wheat cropping systems [17].

4. Sustainable Intensification and Climate-Smart Agriculture: By reducing dependence on synthetic fertilizers, PSB inoculants contribute to lowering greenhouse gas emissions and promoting carbon-efficient farming practices. This aligns with global strategies for climate-smart agriculture aimed at ensuring food security while mitigating environmental degradation [18].

6.2 Challenges in PSB Application

1. Variability in Field Efficacy: A major limitation of PSB inoculants is their inconsistent performance under real-world conditions. Soil heterogeneity, fluctuations in temperature and moisture, and interactions with native microbiota all influence the survival and activity of inoculated strains. Unlike controlled laboratory conditions, field environments often pose stresses that reduce PSB effectiveness.

2. Strain-Specificity and Crop Compatibility: Not all PSB strains perform equally across different cereal crops or soil types. A strain effective in rice rhizospheres may not thrive in wheat or maize systems due to differences in root exudation and soil microenvironments. This necessitates the development of crop-specific or region-specific formulations to ensure consistent benefits.

3. Formulation and Shelf-Life Limitations: Commercial PSB biofertilizers often suffer from short shelf life and reduced viability during storage and transport. Advances in carrier materials (peat, lignite, biochar, or polymer-based encapsulation) are required to enhance inoculant stability and field performance.

4. Regulatory and Commercial Barriers: The commercialization of PSB inoculants is hindered by insufficient molecular data for regulatory approval, lack of standardized protocols for strain validation, and limited awareness among farmers. Policies promoting the integration of microbial biofertilizers into mainstream agricultural practices are necessary to bridge this gap.

PSB play a pivotal role in enhancing phosphorus availability in cereal crop systems, with field-based studies consistently demonstrating yield improvements and reductions in chemical fertilizer dependency [19]. Their applications extend beyond phosphorus solubilization to include bioremediation, microbial consortia development, and contributions to climate-smart agriculture. However, widespread adoption faces hurdles related to field variability, formulation challenges, and regulatory barriers. Future research must focus on developing robust, crop-specific, and site-specific PSB inoculants, while integrating advanced molecular tools for strain characterization and quality assurance. Strengthening the link between laboratory findings and on-farm performance will be crucial for realizing the full potential of PSB in sustainable agriculture.

8. Conclusion

Phosphate solubilizing bacteria (PSB) have emerged as a cornerstone of sustainable agricultural practices, offering an eco-friendly alternative to chemical phosphate fertilizers. Their ability to mobilize insoluble phosphorus reserves through organic acid secretion, enzymatic activity, and synergistic interactions with plants makes them highly valuable for improving nutrient use efficiency in cereal crops. Given that cereals such as rice, wheat, and maize constitute the backbone of global food security, the role of PSB in enhancing phosphorus availability cannot be overstated.

Advances in molecular characterization techniques have significantly expanded our understanding of PSB diversity, functionality, and ecological adaptability. Tools such as 16S rRNA gene sequencing, functional gene analysis, and next-generation sequencing (NGS) approaches have revealed the presence of both culturable and unculturable PSB within cereal rhizospheres, providing insights into their mechanisms of action and potential for targeted application. These molecular tools not only facilitate accurate identification and strain selection but also support the development of robust biofertilizer formulations, several challenges remain in translating laboratory success into consistent field performance. Soil heterogeneity, environmental variability, and competition with native microbiota often limit the efficacy of introduced inoculants. Furthermore, issues related to formulation stability, strain specificity, and regulatory approval hinder widespread adoption.Nevertheless, the integration of molecularly characterized PSB into biofertilizer technology presents a promising pathway for reducing fertilizer dependency, enhancing crop productivity, and promoting climate-smart agriculture. With continued research, policy support, and farmer adoption, PSB can play a transformative role in ensuring sustainable cereal production and global food security.

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