

Exploring the role of plant associating bacteria as bioinoculants and their beneficial effects in phytostimulation: A review

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With an increase in global demand for food without unwanted environmental issues stresses a need for sustainable agriculture. Up till now, conventional agricultural methods focused on obtaining great crop yields from the use of chemical fertilizers but overlooked the hazardous concerns that are leading to soil depletion. These chemical fertilizers adversely affect soil structure, decrease fertility, damage soil flora, and lead to soil erosion. In this scenario, understanding the natural mechanisms of plant-microbe interactions in the rhizospheric environment can potentially lead a way towards eco-friendly agriculture, as the plant associating bacteria prompting phytostimulation can be the key players in unlocking sustainable alternative for conventional fertilizers. Plant growth-promoting bacteria (PGPB) are a distinct class of soil microorganisms that promote plant growth and yields by enhancing nutrient delivery and shielding the plants against diseases. Nitrogen-fixing bacteria such as *Rhizobium* and *Azotobacter*, for instance, fix atmospheric nitrogen into a usable form for plants, which minimizes synthetic fertilizers' requirement. Some other PGPB genera such as *Pseudomonas* and *Bacillus* induce root and shoot elongation by synthesizing phytohormones. These bacteria also provide protection to plants by synthesizing antimicrobial substances and increasing the competitive nature of the rhizosphere. Bacteria like *Azospirillum*, *Enterobacter*, and *Flavobacterium* also stimulate plant growth by producing phytohormones under specific environmental conditions. Utilization of PGPB as bio-stimulants in agriculture is a promising method for sustainable agriculture, minimizing dependence on chemical fertilizers and maintaining soil health. This approach would play an important role in sustaining a balanced ecosystem along with increasing agricultural productivity.

Keywords: Bacteroids, Bio-stimulant, Chemotaxis, Diazotropicus, Fertilizers, PGP bacteria, Phytohormone, Plant health, Rhizosphere

Introduction

Plants, throughout their life cycle, maintain dynamic relationship with micro-organisms which aids their growth and development. Among such bacteria, plant growth promoting bacteria are one of their kind species that take charge in boosting plant health and productivity by residing in their close proximities. These bacteria benefit the plants in numerous ways – both direct and indirect, starting from improving nutrient uptake to conferring disease resistance and decreasing environmental stress¹. Eventually these contribute to them in thriving at challenging environments without any need for synthetic fertilizers². Direct effects of PGPB on plants involves nutrient cycling, plant hormone production, and antagonism against pathogens. For example, certain PGP organisms ability to degrade organic

matter in the soil, results in generation of reduced forms of essential nutrients like nitrogen, phosphorus, and potassium that can be more consumed by plants. Additionally, some bacteria produce plant growth hormones, such as auxins, cytokinins, and gibberellins, which primarily involves in stimulating root elongation and growth metabolism. But indirect impacts are frequently seen by the elimination of pathogens from plant roots or aerial portions *via* competence, or by facilitating immuno-modulation in plants. These bacteria attain nutrition from the chemical secretions and root exudates, which foster an environment friendly for those bacteria which can outcompete dangerous pathogens³. PGPB can also provide durability and resilience to plants stress and drought conditions. In addition to the rhizosphere, another population of helpful bacteria that shield plants from diseases and environmental stresses is found in the phyllosphere, which is the surface of plant leaves, stems, and fruits. Apart of the

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forementioned, there are endophytic bacteria, which confer the stimulatory effects on plants colonizing the internal tissues of plants⁴.

However, a transition happened in research on PGPB where the recent studies suggested employing mixed-strain treatments over single strains for phytostimulation, due to the synergistic potential among microbial communities that benefits the plant growth^{2,5}. These bacteria offer an authentic application to limit the use of synthetic pest control agents and chemical fertilizers, thereby putting forward more eco-friendly agriculture practices. Thus, understanding how plant growth promoting (PGP) bacteria contribute to the development of robust and healthy plants and their mechanisms, is essential with a stressing demand for sustainable agriculture practices⁵. They are an essential part of contemporary agricultural methods because of the increasing amount of research on PGPB, which highlights their ability to improve plant production as well as preserve and repair soil health.

Association of PGP bacteria with plants

In the rhizosphere, plants obtain a systemic relationship with plant growth-promoting bacteria (PGPB) through complex molecular signaling, where they actively exchange a variety of compounds under a dynamic environment supporting vegetation of both plants and bacteria. For these bacteria to colonize the root surfaces, root exudates, such as sugars, amino acids, organic acids, vitamins, phenolics and diverse secondary metabolites acts as key players in this phenomenon^{6,7}. These exudates acting as pre-communication signals, drives bacteria to recognize their plant host and establish symbiosis⁶. Along with these compounds, certain other volatile substances such as carbon dioxide, secondary metabolites, alcohols, and aldehydes also shapes the microflora around rhizosphere, upon their release into soil⁷. However, a set of factors are studied to be influencing the composition of root exudates, such as environmental conditions, plant genotype, and the microbial population within the rhizosphere, collectively playing a role in facilitating interactions between plants and their microbial partners⁶. Eventually, this ongoing rapport of plants with PGPB offers a great support to plant health and productivity.

Also, the carbon level fluctuations in the rhizosphere is considerably an influencing factor of microbial growth and development. Sugars exuded from plants, like glucose and galactose, are often important for

regulating genes related to uptake and breakdown of these compounds. Other compounds exuded from plants, plant-derived polyphenols, phenolic acids are equally important. Since they involve in development of plant stress resistance, as well as in maintaining mutualistic associations between plants and microbes. These interactions are signaled by molecules that help establish symbioses, such as the relationship between legumes and rhizobia, or the formation of arbuscular mycorrhizal symbioses. Additionally, phenolic compounds can trigger redox reactions that affect microbial community composition by influencing hormonal balance and nutrient dynamics, either supporting or competing with plant and microbial growth. Flavonoids, in particular, act as key signaling molecules in plant-microbe interactions, like attracting rhizobia and activating genes for nodulation, thereby influencing the balance between plant hosts and pathogens through phytohormonal regulation^{6,7}.

In some cases, alterations in composition of root exudates happen, as a result of presence of toxic substances in the rhizosphere which eventually drives a hypersecretion of organic acids like malic acid, oxalic acid and citric acids. These organic acids can either attract microbes or provide a carbon source for their nutrition. Indeed, the concentration of hydrocarbon substrates decide the diversity and abundance of microbial population to be entertained around the plant proximities. Furthermore, environmental factors such as nutrient deficiencies, temperature extremes, soil pH, light intensity, soil type, oxygen levels, and microbial presence can all influence the efficiency of exudation processes⁷.

Proteins facilitating the Plant-rhizobacteria interactions⁶

Earlier, it was studied that Rhizobia as a bio-inoculant has shown significant effect on the protein profile in various plants, which induces the expression of several key proteins, particularly in the root hairs, roots, and bacteroids. Proteins, which takes up active role in signal transduction, oxidative stress responses and carbohydrate metabolism, such as calcium/calmodulin kinase, lipoxygenases, phospholipase D, ascorbate peroxidase, phosphoglucosyltransferase, and lectins are upregulated. In the roots, a drastic increase in the level of proteins linked to metabolize carbohydrates, amino acids, and flavonoids, in turn reflects the complex biochemical interactions between the plant and rhizobia. Bacteroids are symbiotic nitrogen-fixing bacteria that possess a set of unique

proteins that include transport and detoxification proteins, as well as proteins related to stress reactions and carbon and nitrogen metabolism. These proteins play a crucial role in the proper fixation of nitrogen and for the survival of rhizobia in the plant root system. The focus of research has thus shifted to the proteins associated with the outer membrane of the bacterium or secreted into the rhizosphere through the changes induced by the plant.

The rhizobial proteins are secreted via many cytoplasmic routes, mainly through the general secretion (Sec) and two-arginine (Tat) pathways as well as specialized systems like types III, IV, V, and VI secretion⁸. Among the many notable secreted proteins of rhizobia are adhesins such as rhicadhesin, which helps in adhering rhizobium to root hairs, and hydrolytic enzymes such as cellulase, which breaks down the root hair cell walls, allowing penetration of the bacteria⁹. Also, glycanases modify extracellular polysaccharides (EPS), play an important role in biofilm formation and nodulation. Significantly, these proteins include PlyA and PlyB in *R. leguminosarum*, or ExoK and ExsH in *S. meliloti*. Another important protein released by *R. leguminosarum* in response to flavonoids generated by the plant is Nod-O, a calcium-binding protein that presumably aids cation transport across root hair membranes. This protein collaborates with nodulation factor (NF) signaling to enhance nodulation.

This association between rhizobia and plant roots induces the secretion of plant-derived proteins in the rhizosphere. These include hydrolases such as chitinases, glycosidases, and peptidases to degrade microbial structures. In addition, there are pathogenesis-related proteins such as thaumatin-like proteins and lectins from plant cells, along with superoxide dismutase, glycine betaine-binding ABC transporters, and outer membrane lipoproteins from bacteria. Proteins that manage plant immune responses and serve in nutrient exchange contribute to the establishment of beneficial symbiotic relationships with the rhizobia. In summary, proteins involved in plant-rhizobia interactions are highly diversified: both plant- and bacterial-derived factors signal transduction, adhesion, metabolism and immune response, helping with the establishment of good symbionts and fixation of nitrogen⁶.

Root colonization of PGP bacteria

Rhizospheric bacteria colonize plants through a distinct mechanism that involves several sequential

steps, that often differentiates with bacterial lifestyle. Bacteria can colonize the rhizosphere soil, the rhizoplane (root surface), or even enter plant tissues as endophytes. The beginning of colonization is marked by chemotaxis, followed by adhering to the root surface, and in some cases, biofilm formation on the rhizoplane or endophytic colonization. These bacteria migrate and attach to target roots, when guided by a conserved intracellular signal transduction pathway and various signal sensors¹¹. Upon reaching the rhizosphere, bacteria relieve from further migration and adhere to the root surface, the process is coined as root attachment. At this stage, bacteria must also overcome the plant's immune response to continue colonizing the root¹². Occasionally, distinct species of rhizobacteria form biofilms on the rhizoplane. During this process, competency between bacteria emerges for uptake of essential nutrients that support their growth and biofilm development. While some endophytes stay still on the root surface and eventually penetrate into plant tissues.

Endophytic bacterial colonization is a multi-step process that includes (a) chemotaxis, (b) adhesion on the root surface, (c) entry and distribution inside the root, and (d) development and survival of the population as micro-colonies¹¹.

a) Chemotaxis is the ability where certain chemicals released from plants guide the bacteria to move along, which is crucial for rhizosphere colonization and establishing primary bacteria-root interactions. Root exudates activate chemosensory pathways, causing motile bacteria to move towards the root and through means of flagellar swimming, swarming, twitching, and gliding, motility occurs. These processes determine the initial contact site on the root, which determines the root colonization efficiency.

b) The second step of symbiosis between bacteria and plant host comprises root surface colonization, a crucial process for rhizoplane and endophytic colonization. This process is driven by; an initial attachment, where rhizobacteria embeds onto root surface reversibly, and secondary attachment graduates to an irreversible attachment. Initial attachment is characterized by weak, reversible, and nonspecific binding, allowing single cell attachment. Physicochemical and electrostatic forces influence the initial interactions between the root and bacterial cell envelope. Rhizobacteria uses their locomotory equipment *i.e.* pili, flagella, polar

flagellum, and fimbriae to pass through these forces. In the secondary attachment stages, only a small percentage of rhizobacteria switch to a stronger binding mode.

c) Endophytic bacteria find entry into plant roots through various pathways, including unintended openings from wounds, cracks or adventitious roots, emerging lateral roots, root hairs, undamaged epidermal cells, and hydathodes in the stems. However, establishment of successful colonization occurs in around a week. Once established, they can expand into the aerial parts of the plant by producing degradative enzymes such as pectinases and cellulases. Physiological state and immune-responsive state of target plant influences the success of root colonization.

Mechanism of action of PGPB on plants

Notable studies about mechanism of action of Plant Growth-Promoting Bacteria (PGPB) have demonstrated an improvement in plant growth and nutritional status, even under adverse environmental conditions such as drought and salinity¹³. As their direct mechanisms benefits plant by, synthesis of phytohormones (such as IAA, gibberellins, cytokinins, ABA, and ethylene), biological nitrogen fixation, and the solubilization of phosphate. Indirectly, they contribute to plant health by producing substances like hydrogen cyanide, antibiotics, volatile organic compounds, siderophores, and ammonia, which act to suppress pathogenic organisms. Additionally, PGPB enhance plant water dynamics and also involves in regulating ion balance, and mitigate abiotic stress^{13,14}. In terms of inducing the plant resilience and fitness, these bacteria acts primarily through direct mechanisms such as hormone production, nitrogen fixation, and phosphate solubilization, as well as indirect mechanisms that involve pathogen inhibition, including the production of antibiotics, cell wall degrading enzymes, antioxidants, quorum sensing interference, induced systemic resistance, and iron sequestration through siderophores.

Production of phytohormones by PGP bacteria

PGP bacteria produce various phytohormones that are chemical signals operating in regulation of cellular processes in plants at low concentrations. The most notable groups of phytohormones include auxins, cytokinins, gibberellins, abscisic acid, ethylene, brassinosteroids, salicylic acid, jasmonates, polyamines, nitric oxide, and strigolactones¹⁵. Microorganisms like *Pseudomonas*, *Azospirillum*, *Rhizobium*, *Bacillus*,

Delftia and *Klebsiella* are recognized as producers of phytohormones¹⁶. Bacterial-secreted hormones, particularly cytokinins (CKs) and auxins, they act as signaling agents in controlling plant cell division and differentiation, eventually they are involved in influencing root and shoot development¹⁷.

Cytokinins are the purine derivatives synthesized in root tips and developing seeds. These compounds serve as secondary messengers in detection of nodulation factors (NF) facilitating nodule formation in legumes. There are two proposed pathways for the biosynthesis of cytokinins: the direct pathway, which involves the synthesis of dimethylallyl pyrophosphate (DMAPP) and N⁶-isopentenyladenosine monophosphate (i6 AMP) from AMP, and the indirect pathway, which involves the turnover of tRNA containing cis-zeatin to form zeatin-type compounds. Auxins, aromatic compounds with carboxylic acid groups, play a significant role in plant growth, modifying root structure, and controlling apical dominance. Indole-3-acetic acid (IAA) is the most abundant auxin in plants, responsible for cell enlargement, division, tissue differentiation, and responding to light and gravity. It is produced through de novo synthesis from tryptophan, which passes through oxidative deamination or decarboxylation. Over 80% of Rhizospheric bacteria isolated in different crop regions have shown to produce and release IAA. Regulation of these physiological processes by auxin involves auxin-induced changes in gene expression. Gibberellins and brassinosteroids are also crucial in processes like nodule formation. Gibberellins are synthesized from geranyl diphosphate through a series of enzymatic transformations. The biosynthetic pathway involves the conversion of geranyl diphosphate into early gibberellin intermediates, followed by the production of active gibberellin compounds. Gibberellin regulation is tightly controlled by DELLA proteins, whose C-terminal GRAS domain forms the structural core essential for their function. DELLA proteins act as negative regulators of gibberellin signaling, and their accumulation in seeds triggers the expression of genes responsible for producing F-box proteins, which play a role in the degradation of DELLA proteins. This degradation is facilitated by the SCF complex (containing GID2/SLY1), which targets DELLA proteins for ubiquitination and subsequent proteasomal destruction, thus modulating gibberellin levels and activity within the plant. Ethylene is a

crucial metabolite in plant growth and development, acting as a growth regulator and stress hormone. Certain environmental parameters influence the production of ethylene, that includes: salinity of soil, high temperature, drought, wounding of plant, water logging, heavy metal deposition. PGPRs with enzyme ACC deaminase support growth and development by declining ethylene levels, promoting salt tolerance, and decreasing drought stress. Various bacterial strains containing ACC deaminase enzymes show good effects on plant growth and development, making them potential candidates for biofertilizer preparation^{6,18}.

Production of volatile organic compounds by PGP bacteria

Bacterial strains associated with plants have the ability to produce and actively emit a wide variety of volatile organic compounds (VOCs). Volatile organic compounds, owing to their low molecular weight (less than 300 Da), low boiling temperatures, and high vapour pressure, are able to seamlessly pass through biological membranes and permeate the surrounding environment. There are now 450 bacterial and fungal strains from which over 1500 distinct VOCs have been identified¹⁹. Bacterial volatile organic compounds (VOCs) have been divided into six chemical classes based on their structure: acids, hydrocarbons, ketones and alcohols, chemicals containing nitrogen, sulfur compounds, and terpenes²⁰.

Volatile organic compounds (VOCs) are a diverse group of chemical substances produced by microorganisms through various biosynthetic pathways. Hydrocarbons, which are organic compounds composed solely of hydrogen and carbon, can be generated by either the elongation-decarboxylation mechanism or head-to-head condensation, both of which are derived from biosynthetic fatty acid pathways. Cyanobacteria, for instance, are known for their ability to produce longer hydrocarbons through the consolidation of extended hydrocarbons²¹. Additionally, ketones and alcohols, such as acetoin and 2, 3-butanedione, are formed through the disintegration of fatty acids during anaerobic pyruvate fermentation. In comparison to ketones and alcohols, acids are present in lower concentrations, and typically include compounds like propionic or butyric acids, which are released from unsaturated fatty acid chains. Another important metabolite is glyoxylic acid, which is produced through specific metabolic reactions and serves as a crucial intermediary in various biosynthetic processes²⁰.

Sulfur-containing compounds also play a significant role in the production of volatile organic compounds, particularly in the aroma of fermented products such as cheese and wine. These compounds are largely derived from the biogenesis of volatiles linked to methionine metabolism. For example, dimethyl sulfide and 1(methyl thio)-3-pentanone are key sulfur-based volatiles produced by bacteria²². The cleavage of 3-dimethylsulfoniopropionate, derived from L-methionine, is often observed in plants and marine algae. This process underscores the critical role of sulfur compounds in shaping the scents and flavors of microbial and plant-associated environments. In addition, nitrogen-containing VOCs such as trimethylamine (TMA) and trimethylamine oxide (TMAO) are produced by bacteria through the reduction of TMAO²³. These compounds are not only involved in the enhancement of plant growth but also contribute to the characteristic smell of spoiled fish. Trimethylamine and its derivatives are common in both fish and the intestines of animals, including humans, where they play a role in digestion and microbial processes.

Terpenes, another class of VOCs, are synthesized from the precursor molecules dimethylallyl pyrophosphate and isopentenyl pyrophosphate. These compounds can be produced through two primary biosynthetic pathways: the mevalonate pathway or the deoxy-xylulose phosphate pathway. Notable terpenoids include geosmine, known for its earthy odor, and albaflavone, an antibiotic compound. Both of these terpenoids are produced by bacteria and contribute to the unique chemical profiles of microbial environments, influencing both ecological interactions and the aromas associated with certain bacterial species²⁴.

Plant growth promoting (PGP) bacterial genera Azospirillum

Azospirillum, a genus of gram-negative plant growth-promoting bacteria (PGPB), plays a crucial role in enhancing plant growth and development through various mechanisms. Its ability to colonize over 100 plant species highlights its widespread significance in agriculture. *Azospirillum* contributes to plant growth both directly and indirectly. Directly, it promotes plant metabolism by producing essential compounds such as polyamines and cadaverine, which regulate root growth and stress responses. For instance, cadaverine, synthesized from lysine, has been linked to increased root growth in species like

pine and soybean, while also helping plants like rice cope with osmotic stress by improving water retention and reducing abscisic acid (ABA) levels. Indirectly, *Azospirillum* enhances plant growth through several key processes such as nitrogen fixation, phosphate solubilization, hormone regulation, and pathogen control. Its ability to fix atmospheric nitrogen into ammonia, via the enzyme nitrogenase, is particularly significant, especially in nitrogen-deficient soils. The bacterium also produces 1-aminocyclopropane-1-carboxylate (ACC) deaminase, which decreases ethylene levels by breaking down ACC, the precursor to ethylene, a hormone known to inhibit plant growth under high concentrations. Additionally, impact of *Azospirillum* on improving root systems, mineral uptake, and water absorption has been demonstrated in various species, such as *Sorghum bicolor*, where inoculation increased adventitious root growth and improved foliage characteristics. Overall, diverse biological functions of *Azospirillum* significantly contribute to improved plant health, resilience, and productivity in agricultural systems²⁴.

Azotobacter

Azotobacter is another genus of bacteria, a beneficial rhizobacterium that enhances plant growth through direct and indirect mechanisms. It plays a crucial role in nitrogen fixation by synthesizing the enzyme nitrogenase in nitrogen-deficient soils. *Azotobacter* also promotes plant growth by producing phytohormones like indole acetic acid, cytokinins, and gibberellins, leading to increased crop productivity and environmental resistance. It also provides indirect benefits as a bioprotectant, controlling plant diseases and contributing to nutrient cycling in soil. Application of *Azotobacter* as an inoculant has been widely adopted in regions like India, China, and Indonesia due to its positive impact on crop yield and plant quality. *Azotobacter* also produces siderophores, these are iron chelating compounds that shows high affinity to ferric ion. Siderophores binds with iron (Fe^{3+}) available in the rhizosphere, leaving the soil pathogens scarce of iron. Since pathogens are not built with ferri-siderophore receptors, prevent them from uptake of the iron-siderophore complex²⁵.

Azotobacter inoculation has been shown to significantly improve crop production across a variety of crops, including wheat, beans, corn, potatoes, oats, and cloves²⁶. Inoculating wheat seeds with *A. chroococcum* resulted in a 42% increase in dry matter compared to the control. Additionally, a

10-18% increase in yields has been reported for crops such as beans, corn, and potatoes, while a foliar spray of *Azotobacter* boosted both grain and straw yields of rice. *Azotobacter* inoculation holds great potential for improving yields in oilseeds and other crops, with notable increases in mustard, sunflower, sugarcane, fruit trees, pearl millet, sorghum, jute, and cotton. Several vegetable crops, including tomato, brinjal, cabbage, onion, radish, chilies, and sweet potato, have also shown positive responses to *Azotobacter* inoculation. Furthermore, a synergistic effect was observed when *Azotobacter* was co-inoculated with *Rhizobium* in crops like pea, chickpea, and groundnut, further enhancing crop productivity²⁷.

Bacillus

Members of the *Bacillus* genus are among the most abundant and widely distributed soil microorganisms in the rhizosphere, exhibiting numerous plant growth-promoting (PGP) characteristics. *Bacillus* species play a vital role in enhancing soil nutrient availability by solubilizing and mobilizing nutrients like phosphate, potassium, and zinc, converting them into forms that plants can absorb. Endophytic *Bacillus* species such as *B. amyloliquefaciens*, *B. megaterium*, *B. subtilis*, and *Brevibacillus agri* are particularly effective at solubilizing insoluble phosphates, increasing the bioavailability of zinc²⁸. Potassium and zinc deficiencies in soil have been reported recently, and certain potassium-solubilizing plant growth-promoting rhizobacteria (PGPRs) such as *B. edaphicus*, *Acido thiobacillus ferrooxidans*, *B. mucilaginosus*, and *Paenibacillus* can release potassium in a form accessible to plants. Species like *Bacillus aryabhatai* and *B. subtilis* also solubilize insoluble zinc, making it more available for plant uptake. In addition, various *Bacillus* and *Paenibacillus* species contribute to improved iron absorption by producing siderophores, which bind to iron and other metals, creating soluble forms that plants can use. Siderophores can also help remediate heavy metals, such as lead (Pb), by aiding in their phytoextraction from soil²⁹.

Pseudomonas

Pseudomonas are another set of bacterial species including fluorescent *Pseudomonads*, are gram-negative, aerobic rod shaped, which are known for acclimatizing rhizosphere and rhizoplane, showing rapid growth rate and the ability to breakdown various organic substrates, including root exudates. They

mostly employ diverse biocontrol mechanisms, including antibiosis, HCN production, siderophore production, competency in nutrient uptake, and induced systemic resistance. *Pseudomonas* release growth factors that aids in the plant's rhizosphere competition, efficient microbial colonization, and collaborative synergy. Numerous *Pseudomonas* species, including *Pseudomonas aeruginosa*, *Pseudomonas fluorescens*, *Pseudomonas jessenii*, *Pseudomonas putida*, and *Pseudomonas vancouverensis*, are recognized for their functions in the bio-suppression of plant infections. Prior to their treatment with plant growth promoting *Pseudomonas* strains, various plant species have been reported to have shown increased root and shoot mass and suppression of pathogenic microflora, indicating the phytostimulation potential of *Pseudomonas*³⁰. *Pseudomonas* do not form a symbiosis similar to that formed by *Rhizobia* with plants, but they can penetrate plant tissues and establish themselves as endophytes. *Pseudomonas* and arbuscular mycorrhizal (AM) combined as an inoculant boost plant growth, harvest index, mineral absorption (including nitrogen), high soluble phosphate content, and root colonization ability under stressful circumstances. Both *Pseudomonas aeruginosa* and *Pseudomonas fluorescens* are capable of producing cyanide as a secondary metabolite, which contributes significantly to the effectiveness of plant growth-promoting bacteria (PGPR) by suppressing phytopathogens. *Pseudomonas fluorescens* is another species that has been shown to improve groundnut plant germination by 29–30%, resulting in a notable increase in grain yield of over 70%³¹.

Flavobacterium

Flavobacterium, a genus of bacteria found in soil and the plant rhizosphere, have been studied as bio-stimulants in various plants. Plant-wide microbiota analysis marked the prevalence of *Bacteroidota*, including *Flavobacteriaceae* family members often observed in specific plant-beneficial interactions. *Flavobacterium* species, relatively abundant in the rhizosphere and are recognized as a core taxonomic group in the rhizospheric population. Studies have been conducted showing the benefitting actions of *Flavobacterium* species in plant growth stimulation in various plants, such as tomato, maize, rice, and ryegrass³². The effects of PGP on plants may be linked to the production of substances that help plants grow, such as auxin and nitrogen resources.

Flavobacterium are also known for providing abiotic stress tolerance, including drought and salt stress, to both monocot and dicot plants. *Flavobacterium* species like IG 15, IR29-16, IC27-25, and IC31-28 are reported in enhancing tolerance against drought and stress conditions in monocot plants like wheat and rice. Another strain, *F. crocinum* HYN0056 activates stress tolerance to drought and salt stress in *Arabidopsis thaliana*. As a result, these are mainly involved in activating the molecular pathway associated with abscisic acid (ABA). *Flavobacterium* species also regulate diverse stress-inducible genes, including WRKY transcription factor and antioxidant enzyme-encoding genes, in addition to ABA signaling³².

Acetobacter

Acetobacter diazotrophicus, a nitrogen-fixing bacterium from the *Acetobacteriaceae* family, plays a significant role in enhancing plant growth and development, particularly in sugarcane. First identified in Brazil, it has since been found in various regions worldwide, including Australia, India, Mexico, Uruguay, Canada, and Cuba. This bacterium is primarily associated with the roots and stems of sugarcane, suggesting it acts as a systemic endophyte, colonizing different parts of the plant while remaining absent in the surrounding soil. Beyond sugarcane, *A. diazotrophicus* has also been isolated from other sugar-rich plants such as *Pennisetum purpureum* (elephant grass) and sweet potato, as well as from certain insects like mealybugs and leafhoppers. Additionally, it has been shown to colonize coffee plants, both through seed and vegetative propagation. In agricultural practices, particularly in India, *A. diazotrophicus* has been effectively used to colonize sugarcane varieties, especially where the use of chemical nitrogen fertilizers is replaced by organic manures over extended periods. This bacterium contributes to plant growth by fixing atmospheric nitrogen, thus reducing the need for chemical nitrogen inputs and promoting healthier, more sustainable crop development. As a result, *Acetobacter* is increasingly utilized as a beneficial inoculant in sugarcane cultivation, helping to support plant nutrition and overall growth³⁵.

Other genera

Apart of the forementioned species, others too such as *Enterobacter*, *Micrococcus*, *Klebsiella* have been shown to promote plant growth, especially under

stress conditions such as phosphorus deficiency and heavy metal contamination. These benefits are attributed to *Enterobacter* species, being able to solubilize inorganic phosphorous influence plant metabolism in phosphorus-limited environments³⁴. For instance, a study by Alzate *et al.* (2021) found a significant participation of *Enterobacter* in root development and increased phosphorus uptake in maize and cucumber plants. In cucumber, this helped the plants to withstand the effects of phosphorus deficiency³⁵. In another study by Badawy *et al.* (2022), they explored the role of *Micrococcus luteus* and *Enterobacter cloacae*, in helping tomato plants to withstand heavy metal stress, specifically from cadmium (Cd) and nickel (Ni) contamination. These bacteria are known to develop resilience in plants against heavy metals by strengthening their antioxidant systems and promoting osmoregulation³⁶. On the other hand, *Klebsiella variicola* has also been studied for its ability to provide tolerance to salinity, particularly in wheat and maize crops under saline conditions, indirectly aiding their growth. This bacterium produces key proteins that aid in salinity tolerance, such as ACC deaminase, indole-3-acetic acid (IAA), exopolysaccharides (EPS), and osmolytes³⁷. *K. variicola* as a inoculant for plants seemed to have enhanced seed germination, root length, shoot height, chlorophyll content, and nutrient uptake (N, P, K, Na, Mg). The bacterium also alleviated salinity stress through osmolyte production and pH regulation, leading to improved soil properties and overall plant health under stressful conditions³⁸.

Conflict of interest

All authors declare no conflict of interest.

References

- Raj R, Dhiman S & Shakya L, Nanotechnological interventions in sustainable agriculture. *Indian J Biochem Biophys*, 59 (2022) 1153.
- Santoyo G, Guzmán-Guzmán P, Parra-Cota FI, Santos-Villalobos Sdl, Orozco-Mosqueda MdC & Glick BR, Plant growth stimulation by microbial consortia. *Agronomy*, 11 (2021) 219.
- Wang H, Liu R, You MP, Barbetti MJ & Chen Y, Pathogen biocontrol using plant growth-promoting bacteria (PGPR): Role of bacterial diversity. *Microorganisms*, 9 (2021) 1988.
- Orozco-Mosqueda MdC, Flores A, Rojas-Sánchez B, Urtis-Flores CA, Morales-Cedeño LR, Valencia-Marin MF, Chávez-Avila S, Rojas-Solis D & Santoyo G, Plant growth-promoting bacteria as bioinoculants: Attributes and challenges for sustainable crop improvement. *Agronomy*, 11 (2021) 1167.
- Kathalia R, Checker VG, Jha BS & Saxena T, Environmental nanotechnological applications for sustainable agriculture. *Indian J Biochem Biophys*, 59 (2022) 1056.
- Morel MA & Castro-Sowinski S, The complex molecular signaling network in microbe-plant interaction. In: Arora N, ed, *Plant Microbe Symbiosis: Fundamentals and Advances*. Springer, New Delhi (2013) 1.
- Khare E, Tyagi S & Patil KS, Language of plant-microbe-microbe interactions in rhizospheric ecosystems. In: Maheshwari D, ed, *Molecular aspects of plant beneficial microbes in agriculture*. Academic Press (2020) 59.
- Tseng TT, Tyler BM & Setubal JC, Protein secretion systems in bacterial-host associations, and their description in the Gene Ontology. *BMC Microbiol*, 9 (2009) S2.
- Gupta G, Chauhan PS, Jha PN, Verma RK, Singh S, Yadav VK, Sahoo DK & Patel A, Secretory molecules from secretion systems fine-tune the host-beneficial bacteria (PGPRs) interaction. *Front Microbiol*, 15 (2024) 1355750.
- Subudhi S, Sethi D & Kumar Pattanayak S, Characterization of *Rhizobium* sp. (SAR-5) isolated from root nodule of *Acacia mangium* L. *Indian J Biochem Biophys*, 57 (2020) 327.
- Liu Y, Xu Z, Chen L, Xun W, Shu X, Chen Y, Sun X, Wang Z, Ren Y, Shen Q & Zhang R, Root colonization by beneficial rhizobacteria. *FEMS Microbiol Rev*, 48 (2024) fuad066.
- Santoyo G, Urtis-Flores CA, Loeza-Lara PD, Orozco-Mosqueda MdC & Glick BR, Rhizosphere colonization determinants by plant growth-promoting rhizobacteria (PGPR). *Biology*, 10 (2021) 475.
- Mazumdar M, Sultana T, Sinha S, Sadhukhan S, Datta A, Mandal M & Mitra AK, Comparison of a novel combination of bio-organic fertilizers vis-à-vis a chemical fertilizer. *Indian J Biochem Biophys*, 59 (2022) 197.
- Gupta A, Mishra R, Rai S, Bano A, Pathak N, Fujita M & Kumar M, Mechanistic insights of plant growth-promoting bacteria mediated drought and salt stress tolerance in plants for sustainable agriculture. *Int J Mol Sci*, 23 (2022) 3741.
- ElSabagh A, Islam MS, Hossain A, Iqbal MA, Mubeen M, Waleed M & Abdelhamid MT, Phytohormones as growth regulators during abiotic stress tolerance in plants. *Front Agron*, 4 (2022) 765068.
- Niranjan V, Sureshkumar P, Shankara L, Khedkar G & Kumar J, Insights on mechanism of plant-related bacteria producing phytohormones. In: New insights into phytohormones. *Intech Open* (2024).
- Orozco-Mosqueda MdC, Santoyo G & Glick BR, Recent advances in the bacterial phytohormone modulation of plant growth. *Plants*, 12 (2023) 606.
- Amara U, Khalid R & Hayat R, Soil bacteria and phytohormones for sustainable crop production. In: Maheshwari D, ed, *Bacterial Metabolites in Sustainable Agroecosystem*. *Sustainable Development and Biodiversity, vol 12*. Springer, Cham (2015) 1.
- Matilla MA & Krell T, Plant growth promotion and biocontrol mediated by plant-associated bacteria. In: Egamberdieva D & Ahmad P, eds, *Plant Microbiome: Stress Response*. *Microorganisms for Sustainability, vol 5*. Springer, Singapore (2018) 1.
- Weisskopf L, Schulz S & Garbeva P, Microbial volatile organic compounds in intra-kingdom and inter-kingdom interactions. *Nat Rev Microbiol*, 19 (2021) 391.

- 21 Coates RC, Podell S, Korobeynikov A, Lapidus A, Pevzner P, Sherman DH, Allen EE, Gerwick L & Gerwick WH, Characterization of cyanobacterial hydrocarbon composition and distribution of biosynthetic pathways. *PLoS One*, 9 (2014) e85140.
- 22 Veselova MA, Plyuta VA & Khmell A, Volatile compounds of bacterial origin: Structure, biosynthesis, and biological activity. *Microbiology*, 88 (2019) 261.
- 23 Calcagnile M, Tredici SM, Talà A & Alifano P, Bacterial semiochemicals and transkingdom interactions with insects and plants. *Insects*, 10 (2019) 441.
- 24 Pedraza R, Filippone M, Fontana C, Salazar S, Ramirez A, Sierra-Cacho D & Baca BE, Azospirillum. In: Pedraza R, ed, *Azospirillum. 1st ed. Elsevier, Amsterdam* (2020) 85.
- 25 Sethi SK & Adhikary SP, *Azotobacter*: A plant growth-promoting rhizobacteria used as biofertilizer. *Dyn Biochem Process Biotech Mol Biol*, 6 (2012) 68.
- 26 Sharma S, Tuladhar R, Basnet Y, Manandhar S, Bhattarai S, Singh A & Varma A, Effect of substrates on Azotobacter chroococcum-enriched vermicompost for growth of Phaseolus. In: Sayyed R, ed, *Plant Growth Promoting Rhizobacteria for Sustainable Stress Management. Microorganisms for Sustainability, vol 13. Springer, Singapore* (2019) 1.
- 27 Das HK, *Azotobacters* as biofertilizer. In: *Advances in Applied Microbiology, vol 108. Academic Press, San Diego* (2019) 1.
- 28 Etesami H, Jeong BR & Glick BR, Potential use of *Bacillus* spp. as an effective biostimulant against abiotic stresses in crops—a review. *Curr Res Biotechnol*, 5 (2023) 100128.
- 29 Jamwal VL, Ntemafack A, Qayum A, Kapoor N, Gulfam S, Singh SK & Gandhi SG, Estimation of bioactive potential of culturable bacterial endophytes from *Coleus*. *Indian J Biochem Biophys*, 59 (2022) 331.
- 30 Ren J, Wang J, Karthikeyan S, Liu H & Cai J, Natural anti-phytopathogenic fungi compound phenol, 2, 4-bis (1, 1-dimethylethyl) from *Pseudomonas fluorescens* TL-1. *Indian J Biochem Biophys*, 56 (2019) 162.
- 31 Sah S, Krishnani S & Singh R, *Pseudomonas* mediated nutritional and growth promotional activities for sustainable food security. *Curr Res Microbiol Sci*, 2 (2021) 100084.
- 32 Seo H, Kim JH, Lee SM & Lee SW, The plant-associated *Flavobacterium*: A hidden helper for improving plant health. *Plant Pathol J*, 40 (2024) 251.
- 33 Tilak K, Nandanavanam R, Pal K, De R, Saxena A, Nautiyal C, Mittal S, Tripathi A & Johri BN, Diversity of plant growth and soil health supporting bacteria. *Curr Sci*, 89 (2005) 1.
- 34 Ranawat B, Mishra S & Singh A, *Enterobacter hormaechei* (MF957335) enhanced yield, disease and salinity tolerance in tomato. *Arch Microbiol*, 203 (2021) 2659.
- 35 Pradhan M, Das R, Sahoo R, Adak T, Pradhan C & Mohanty S, Comparative P solubilizing efficiencies of five acid soil bacteria incubated with calcium, aluminium and iron phosphates. *Indian J Biochem Biophys*, 59 (2022) 947.
- 36 Badawy IH, Hmed AA, Sofy MR & Al-Mokadem AZ, Alleviation of cadmium and nickel toxicity and phyto-stimulation of tomato plant L. by endophytic *Micrococcus luteus* and *Enterobacter cloacae*. *Plants*, 11 (2022) 2018.
- 37 Kusale SP, Attar YC, Sayyed RZ, El Enshasy H, Hanapi SZ, Ilyas N, Elgorban AM, Bahkali AH & Marraiki N, Inoculation of *Klebsiella variicola* alleviated salt stress and improved growth and nutrients in wheat and maize. *Agronomy*, 11 (2021) 927.
- 38 Ghazi A, Atia E & Elsakhawy T, Evaluation of an endophytic plant growth-promoting bacterium, *Klebsiella variicola*, in mitigation of salt stress in tuberose (*Polianthes tuberosa* L.). *J Horticult Sci Biotech*, 96 (2021) 770.