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Research Article

Plant Growth Promoting Rhizobacteria: A Boon to Agriculture

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Abstract

Plant Growth Promoting Rhizobacteria (PGPR) are a group of bacteria that enhances plant growth and yield. PGPRs as biofertilizers, are the living micro-organisms which are applied to surface of plant, seed, or soil. They colonize the plant rhizosphere or inside the plant body (as endophyte) and promotes plant growth by providing fundamental nutrients to crop plants. The rapidly increasing population is exerting immense pressure on agricultural lands for higher crop yields, which results in increasing use of chemical fertilizers. But due to negative environmental impact of artificial fertilizers and their increasing costs, microorganisms have a vital role in agriculture as they promote the exchange of plant nutrients and reduce application of chemical fertilizers as much as possible. Substances produced by PGPRs protect the plants against various pathogens by direct antagonistic interactions between the biocontrol agent and the pathogen, as well as by induction of host resistance. Resistance-inducing and antagonistic rhizobacteria might be useful in formulating new inoculants with combinations of different mechanisms of action, leading to a more efficient use for biocontrol strategies to improve cropping systems.

Keywords: *Plant growth promoting rhizobacteria; biofertilizers; biocontrol; rhizosphere*

1. Introduction

Sustainable agriculture is fundamentally important in today's world because it offers the potential to meet our future agricultural needs because of a fast growing population, something that conventional agriculture will not be able to do. In modern cultivation process, there is a wide indiscriminate use of chemical fertilizers, which has led to pollution of soil, air and water. Excess use of these chemicals exerts deadly effects on soil microorganisms and also contribute in deteriorating the fertility status of soil and polluting the atmosphere. Despite of being costly, the manufacturing of chemical fertilizers depletes non-renewable resources, the oil and natural gas used to generate these fertilizers, and poses human and environmental hazards. The use of these fertilizers on a long term basis leads to decrease in pH as well as exchangeable bases, therefore making them out of stock to crops, as a result of which, the productivity of crop declines (Gupta, 2015). Thus, the negative environmental impact of artificial fertilizers and their ever-increasing costs, the use of beneficial soil microorganisms such as Plant Growth Promoting Rhizobacteria (PGPR) for sustainable and secure agriculture has increased worldwide during the last couple of decades. Therefore, agricultural biotechnology and microbiology aims to develop microbial inoculants which can increase plant growth and suppress plant diseases, so that focus can be on the use of reduced quantity of chemical fertilizers and pesticides. In this, we review different mechanisms commonly used by the PGPR to influence plant growth and health in the natural environment.

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2. Rhizosphere

The rhizosphere is coined more than hundred years ago by Hiltner (1904). It is a nutrient-rich habitat for microorganisms, where severe, intense interactions take place between the plant, soil, and microfauna (Antoun, 2005). They may have positive, negative or no visible effect on plant growth. Plant growth and productivity is highly affected by these interactions. Different type of microorganisms such as bacteria, fungi, protozoa, algae coexist among them. Bacteria contribute most to plant health by releasing many organic exudates, thus creating a selectively sensitive environment where diversity is less (Sivasakthi, *et al.,* 2014). Out of them, plant growth promoting bacteria (PGPR) are most abundant among all others in the rhizosphere.

3. Role of PGPR

PGPR are free living bacteria that resides in soil. They either directly or indirectly assist rooting (Mayak, *et al.,* 1999). They play different roles in the soil which proves beneficial for plant health and productivity. They colonize the rhizosphere and protect plant from its pathogens, produce secondary metabolites such as antibiotics that suppress harmful rhizobacteria, produce siderophores, and phytohormones, can fix atmospheric nitrogen, and help in providing nutrition uptake by solubilizing phosphate and produce biologically active substances which influence the plant growth and development (Arshad and Frankenberger, 1998). PGPR can also clean environment by detoxifing pollutants like, heavy metals and pesticides. Several researches are still going on to understand the diversity and importance of soil PGPR communities and their roles in betterment of agricultural productivity.

4. Occurrence and forms of PGPR

The mechanism by which PGPR exerts their beneficial effect on plants can be very diverse. They can establish themselves on root's suface or inside the roots. PGPR can be classified into extracellular plant growth promoting rhizobacteria (ePGPR) that may exist in the rhizosphere, on the rhizoplane or in the spaces between the cells of root cortex. The bacterial genera such as *Agrobacterium*, *Arthrobacter*, *Azotobacter*, *Azospirillum*, *Bacillus*, *Flavobacterium*, *Pseudomonas* and *Serratia* belong to ePGPR. The other category is intracellular plant growth promoting rhizobacteria (iPGPR) that locates generally inside the specialized nodular structures of root cells (Figueiredo, *et al.,* 2011). It belongs to the family of Rhizobiaceae includes *Allorhizobium*, *Bradyrhizobium*, *Mesorhizobium* and *Rhizobium*, endophytes and *Frankia* species both of which can symbiotically fix atmospheric nitrogen with the higher plants.

5. Mechanism of action

PGPR affect plant growth in two different ways, indirectly or directly. The direct promotion of plant growth by PGPR is either by providing the plant with a compound that is synthesized by the bacterium, for example phytohormones, or by facilitating the uptake of certain nutrients from the environment (Glick, 1995). Isolates exhibiting two or three plant growth promoting (PGPR) traits, as combination of ammonia excretion, IAA production ability, phosphorous solubilization and siderophore production ability of bacteria may promote plant growth synergistically or individually. They have good prospects to improve plant growth as they have nitrogen fixing ability too, especially in soil which is affected by salts (Kochar and Gera 2015), the tested. Thus, multi-strain inoculant biofertilizers may be particularly beneficial. The indirect promotion of plant growth occurs when PGPR lessen or prevent the deleterious effects of one or more phytopathogenic organisms. This can happen by producing antagonistic substances or by inducing resistance to pathogens (Glick, 1995).

6. PGPR as biofertilizers

A vast amount of artificial fertilizers are used to replenish soil N and P, resulting in high costs and increased environmental pollution. Thus, biofertilizers are microorganisms that improve nutrient availability to plants. They contribute to plant nutrition both by facilitating nutrient uptake and by increasing primary nutrient availability in the rhizosphere by different methods, as fixing atmospheric nirogen, solubilizing mineral nutrients, mineralizing organic compounds and producing phytohormones (Bhardwaj, *et al.,* 2014), (Arora, *et al.,* 2011). They are also used to increase crop growth and yield when applied complementary to, or as substitute for, chemical fertilizers. The plant-PGPR cooperation plays an important role by enhancing growth and health of plants.

Nitrogen fixation: Nitrogen (N) is a vital element for all forms of life. It is the most important nutrient for plant growth and productivity. Even 78 % of the nitrogen is present in the atmosphere, it is not available to the plants due to high losses by emission or leaching. PGPR have the ability to fix atmospheric nitrogen and made it available to plants by two mechanisms: symbiotic and non-symbiotic. Symbiotic nitrogen fixation is a mutualistic relationship between a microbe and the plant. Symbiotic bacteria which act as PGPR are *Rhizobium*, *Bradyrhizobium*, *Sinorhizobium*, and *Mesorhizobium* with leguminous plants, *Frankia* with non-leguminous trees and shrubs (Zahran, 2001). Non-symbiotic nitrogen fixation is carried out by free living diazotrophs. They stimulate the growth of non- leguminous plants as radish and rice. Non-symbiotic Nitrogen fixing rhizospheric bacteria belongs to genera including *Azoarcus*, *Azotobacter*, *Acetobacter*, *Azospirillum*, *Burkholderia*, *Diazotrophicus*, *Enterobacter*, *Gluconacetobacter*, *Pseudomonas* and cyanobacteria (*Anabaena, Nostoc*) (Bhattacharyya and Jha, 2012) (Vessey, 2003). This an integrated approach for management of diseases on crop, promotion of plant growth and for maintaining the nitrogen level in agricultural soil.

Phosphate solubilization: Phosphorus is the most important key element in the nutrition of plants, next to nitrogen. It is involved in metabolic processes of plant, as photosynthesis, energy transfer, signal transduction, macromolecular biosynthesis and respiration (Khan, *et al.,* 2010). It is abundantly available in soils in both organic and inorganic forms, but because 95-99% phosphate present in the insoluble, immobilized, and precipitated form. Microorganisms play an important role in mediating phosphorus available to plants by playing a role in the soil phosphorus cycle. PGPRs either directly solubilise and mineralise inorganic phosphorus or facilitate the mobility of organic phosphorus through microbial turnover and/or increase the root system (Richardson and Simpson, 2011). These bacteria secrete different types of organic acids (e.g., carboxylic acid), which lowers the pH in the rhizosphere and thus release the bound forms of phosphate like Ca_3 $(PO_4)_2$ in the calcareous soils. Apart from providing the availability of accumulated phosphate (by solubilization), phosphorus biofertilizers also help in increasing the efficiency of biological nitrogen fixation and render availability of Fe, Zn, etc., through production of plant growth promoting substances. Among the heterogenous and naturally abundant microbes of rhizosphere, phosphate solublizing microorganisms (PSM) have provided an alternative biotechnological solution in sustainable agriculture to meet the P demands of plants. PSM belongs to genera *Bacillus* and *Pseudomonas*.

Siderophore production: Iron is an essential micronutrient for nearly all organisms in the biosphere. Iron is the fourth most abundant element on earth. In aerobic soils, iron is exists as ferric ion or Fe^{+3}, which is the predominant form in nature. This form of iron is not readily assimilated by either bacteria or plants. It is only sparingly soluble so that the amount of iron available for assimilation by living organisms is extremely low (Ma, 2005). So to provide the iron to plants, PGPR secrete Siderophores under iron limited conditions. They are low molecular weight iron chelators that chelate iron from their surrounding environments and form a complex ferric-siderophore that can move by diffusion and be returned to the cell surface (Andrews, *et al.*, 2003). Siderophores are divided into three main families depending on the characteristic functional group, i.e. hydroxamates, catecholates and carboxylates. At present more than 500 different types of siderophores are known, of which 270 have been structurally characterized (Cornelis, 2010).

7. PGPR as phytohormone producers

PGPRs produce includes indole acetic acid, cytokinins, gibberellins and inhibitors of ethylene production. The most common best characterized and physiologically the most active auxin in plants is indole-3-acetic acid (IAA) which stimulates rapid (e.g. increase in cell elongation) and long-term (e.g. cell division and differentiation) responses in plants (Cleland, 1990). Auxins are quantitatively the most abundant phytohormones secreted by *Azospirillum*. IAA, a phytohormone, produced in young leaves, stems and seeds from transamination and decarboxylation reaction of tryptophan. Tryptophan is an amino acid commonly found in root exudates. It is the main precursor molecule for biosynthesis of IAA in bacteria (Etesami, *et al.,* 2009). The most common mechanism for the biosynthesis of indole acetic acid by PGPRs like *Pseudomonas*, *Rhizobium*, *Bradyrhizobium*, *Agrobacterium*, *Enterobacter* and *Klebsiella is* the formation via indole-3- pyruvic acid and indole-3-acetic aldehyde (Shilev, 2013]. The IAA production ability may allow bacteria to detoxify excess tryptophan/tryptophan analogues that are deleterious to the bacterial cell. Several plant growth promoting rhizobacteria *Azotobacter* sp., *Rhizobium* sp., *Pantoea agglomerans*, *Rhodospirillum rubrum*, *Pseudomonas fluorescens, Bacillus subtilis* and *Paenibacillus polymyxa* can produce cytokinins or gibberellins or both can produce either cytokinins or gibberellins or both. Cytokinins stimulate plant cell division and control root development by inhibiting primary root elongation and lateral root formation and promoting root hair formation (Werner, *et al.,* 2003), (Riefler *et al.,* 2006) while gibberellins promote the development of stem tissue, root elongation and lateral root extension (Yaxley, *et al.,* 2001). Ethylene is a key phytohormone has a wide range of biological activities that can affect plant growth and development. It plays important role in root initiation, inhibits root elongation, promotes fruit ripening, promotes lower wilting, stimulates seed germination, promotes leaf abscission, activates the synthesis of other plant hormones. At low levels, it can promote plant growth in several plant species including *Arabidopsis thaliana*, while it is normally considered as an inhibitor of plant growth and known as a senescence hormone (Pierik, *et al.,* 2006).At high concentration, it induces defoliation and other cellular processes that may lead to reduced crop performance [Bhattacharyya and Jha, 2012]. The enzyme 1-aminocyclopropane-1 carboxylic acid (ACC) is a pre-requisite for ethylene production, catalyzed by ACC oxidase. PGPR assuch as *Acinetobacter, Achromobacter, Agrobacterium, Alcaligenes, Azospirillum, Bacillus, Burkholderia, Enterobacter, Pseudomonas, Ralstonia, Serratia* and *Rhizobium* etc. are able to produce ethylene.

8. PGPR as bioformulations for plant growth promotion

Due to wide use of chemical fertilizers and pesticides, harmful chemicals are accumulating in the environment and thus destroying the ecosystem, causing pollution and spreading diseases. Therefore, plant growth promoting rhizobacterial bioformulations is in great demand now so as to protect our environment. Bioformulations refers to preparations of microorganism that may be partial or complete substitute for chemical fertilization, pesticides, which offers an ecofriendly sustainable approach to increase crop production and health. They are biologically active products used in the agricultural fields. They contain one or more valuable microbial strains which are an easy to use and have economical carrier material which maintains the cell viability under adverse environmental conditions. A good quality formulation promotes survival of bacteria maintaining available population sufficient to show growth promoting effects on plants [Singh, *et al.,* 2014]. While introducing microbial inoculants, it must be taken into consideration that what kind of plant species and soil type shape microbial communities in the rhizosphere is there because there can be the chances of competition between inoculated and resident microflora. As this could rapidly deplete the population of introduced microbes and may account in part for the inconsistencies observed between greenhouse studies and field trials (Martinez-Viveros, *et al.,* 2010).

As compared with chemical fertilizers, microbial inoculants have many advantages. If suitable microbes is selected carefully, there is a reduced risk of environmental damage and potentially human health. They are safer to apply and their activity is more targeted. They are effective in small quantities and they multiply fast if given appropriate conditions are provided (where their population size is controlled by the plant and indigenous microbes) and may survive to the next season. They decompose faster and more effectively and they can be used on their own or in combination with conventional pest management (Berg, 2009). But it is important that ratio between inoculum size and the concentration of fertilizers must be effective. Thus PGPR is a promising sustainable and environmentally friendly approach to obtain sustainable fertility of the soil and plant growth indirectly.

9. PGPR as biocontrol

Phytopathogenic microorganisms are a major and chronic threat to sustainable agriculture and ecosystem stability. Regular use of chemical pesticides and fungicides has led to environmental concerns and even cause pathogen resistance, forcing constant development of new agents (Fernando, *et al.,* 2006). According to Beattie (2006), bacteria that reduce the incidence or severity of plant diseases are often referred to as biocontrol agents whereas those that exhibit antagonistic activity toward a pathogen are defined as antagonists. Biocontrol agent may be considered as alternative to chemical pesticides. PGPRs interact with soil pathogens through several mechanisms such as antibiosis (production of antimicrobial compounds), competition for iron and nutrients or for colonization sites, predation and parasitism and induction of resistance factors.

The production of antibiotics is considered to be one of the most powerful and studied biocontrol mechanisms of plant growth promoting rhizobacteria against phytopathogens (Shilev, 2013). To prevent the proliferation of plant pathogens (generally fungi), a variety of antibiotics have been identified, including compounds such as amphisin, 2,4-diacetylphloroglucinol (DAPG), oomycin A, phenazine, pyoluteorin, pyrrolnitrin, tensin, tropolone, and cyclic lipopeptides produced by pseudomonads [Loper and Gross, 2007] and oligomycin A, kanosamine, zwittermicin A, and xanthobaccin produced by *Bacillus*, *Streptomyces*, and *Stenotrophomonas* sp. (Compant, *et al.,* 2005).

Pathogen suppression can also occur competitively through indirect inhibition. *Bradyrhizobium, Pseudomonas, Rhizobium, Streptomyces, Serratia,* and *Azospirillum* are siderophore producing PGPRs. Siderophore production confers competitive advantages to PGPR that can colonize roots and exclude other microorganisms from this ecological niche (Haas and Defago, 2005). Siderophore producing PGPR that can prevent the proliferation of pathogenic microorganisms by sequestering Fe3+ in the area around the root, thus makes iron unavailable to pathogens (Martinez- Viveros *et al.,* 2010).

Both mechanisms have vital functions in microbial antagonism and these mechanisms leads to elicit induced resistance. Induced resistance is the state of an enhanced defensive ability developed by plants when appropriately stimulated in response to specific environmental stimuli (Van Loon, *et al.*, 1998). Resistance-inducing and antagonistic rhizobacteria might be useful in formulating new inoculants. It is an alternate ecofreiendly biological approach for controlling plant diseases and improving the cropping systems into which it can be most profitably applied (Beneduzi, *et al.,* 2012).

Certain bacteria synthesize a wide spectrum of multifunctional polysaccharides including intracellular polysaccharides, structural polysaccharides, and extracellular polysaccharides. Exopolysaccharides (EPS) helps in biofilm formation; root colonization can affect the interaction of microbes with roots appendages. EPS-producing microbes colonizes the plant roots effectively and further helps to hold the free phosphorous from the insoluble one in soils and circulate essential nutrient to the plant for proper growth and development. They also shield the plant from the attack of foreign pathogens. EPS producing microbes form shield from desiccation, protect against stress (Qurashi and Sabri, 2012).

Many PGPRs can synthesize anti-fungal metabolites such as antibiotics, fungal cell wall- lysing enzymes, or hydrogen cyanide, which suppress the growth of fungal pathogens. PGPR and bacterial endophytes play a vital role in the management of various fungal diseases but one of the major problem faced with biocontrol agents is lack of appropriate delivery system.

The evolution of T3SSs in bacteria with this knowledge will contribute in the development of various biotechnological applications involving the design of efficient strategies to control plant diseases, the exploitation of T3SS as scaffold for protein delivery into eukaryotic cells, the improvement of the symbiotic properties of rhizobial species or even the expansion of the symbiotic potential towards the nonleguminous plants of agriculture importance (Tampakaki, 2014).

Thus PGPRs as biocontrol agents can be commercialized by doing genetic modifications which could further contribute to sustainable development of agriculture. Therefore, the selected bacterial strains can be used practically for the development of plant growth promoting or biocontrol inoculants, together with other plant growth promoting microbes [EvaLaslo, *et al.,* 2012).

Conclusion

PGPR can affect plant growth by different mechanisms. Their ability to produce various compounds (such as phytohormones, organic acids, siderophores), fix atmospheric nitrogen, solubilize phosphate and produce antibiotics that suppress deleterious rhizobacteria, and production of biologically active substances or plant growth regulators (PGRs). Significant growth has been achieved in the area of PGPR production. PGPR are potential microbes for enriching the soil fertility and increasing the agriculture yield. Plant growth promotion is a complex phenomenon. Most PGPR influences plant growth through multiple mechanisms, and in some cases their effect may only occur through interactions with other microbes. Resistance-inducing and antagonistic rhizobacteria might be useful in formulating new inoculants,thus, offering an attractive alternative of environmentally friendly biological control of plant disease and improving the cropping systems into which it can be most profitably applied. It requires a systematic approach and design, so as to utilize their valuable and useful properties, even crop yields can be maintained by using the combinations of different mechanisms of action and reducing the use of chemical fertilizers. Keeping in mind the current and future progress of PGPR diversity, ability to colonze , mechanisms of action, formulation and application could aid in their development for sustainable agriculture. PGPR are excellent model systems which can provide the biotechnologist with novel genetic constituents and bioactive chemicals having diverse uses in agriculture and environmental sustainability.

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