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PGPR: One Step Ahead To Sustainable Agriculture

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ABSTRACT:

In modern agricultural system indiscriminate application of chemical fertilizers has led to numerous hazardous effects on agricultural crop yield, plant health as well as these are the extreme sources of pollutants on soil, water and air. In this study we have focused on the role of Plant Growth Promoting Rhizobacteria (PGPR).Now a days PGPR is an alternative source of plant growth enhancer. PGPR improve plant growth by direct and indirect ways. They can fix nitrogen, produce siderophore, produce IAA and HCN etc. All these roles of PGPR make them sufficient to perform as an alternative of chemical fertilizers. In this study we have discussed about the roles of PGPR i.e. how they work on plants by different mechanisms and why they are substituting the place of chemical fertilizers.

Key words: PGPR, siderophore, crop yield, chemical fertilizer, HCN

INTRODUCTION:

The plants require available nutrient in sufficient and balanced amounts for their optimal growth. Although soils contain natural reserves of plant nutrients, but these reserve nutrients are in complex forms and unavailable to plants. Thus only a minor portion is released through biological activity or chemical processes in each year. But this release is so slow that it can't compensate the deficiency of nutrients by agricultural production and failed to meet crop requirements. Therefore, fertilizers are used as the supplementary of the nutrients already present in the soil. The application of chemical fertilizer exhibit both advantages and disadvantages in the context of nutrient supply, crop growth and environmental quality. (1) Integrated nutrient supply system is essential for increasing the crop productivity as well as the maintenance and possibly improvement of soil fertility for sustaining crop productivity. Results from various cropping systems showed that there are positive interactions of the integrated use of mineral fertilizers, organic manures, biofertilizers and spraying of growth regulator for maintaining the growth throughout the crop duration (2,3,4). Plant growth promoting rhizobacteria (PGPR) is a heterogeneous group of bacteria. They are present in the rhizospheric zone of soil, that is at the root surfaces and in association with roots, which improve the plant and soil health directly or indirectly. The term PGPR first coined by Kloepper and Schroth(5). PGPRs induce plant growth by producing growth regulators that excite the activity of other beneficial microorganisms. PGPRs directly stimulate the plant and help in nodulation, or indirectly stimulate nodulation. However, they also involve in acceleration of minerals and uptake certain nutrients like Fe, P, Mn, Zn and Cu etc. (6) PGPR alter the solubility of mineral nutrient by releasing organic acids and creating acidity via CO_2 during respiration. The rhizosphere is suitable habitat for acid producing bacteria (7). The organic acid production was the major mechanism of action through which insoluble phosphate compound were converted to more soluble forms. One of the activity of the PGPR in the rhizosphere affects rooting patterns and the supply of available nutrients to plants, thereby modifying the quality and quantity of root exudates. Some bacteria are reported to enhance the plant growth including species of *Azospirillum Azotobacter, Pseudomonas, Enterobacter, Arthrobacter, Burkholderia* and *Bacillus* (8,9,10,11,12). The use of PGPR to plant growth has increased dramatically in various part of the world day by day. Beneficial rhizobacteria have ability to reduce the use of chemical fertilizer and provide a sustainable agriculture as part of an overall management system (13). Sulfur dioxide (SO_2) is one of the major air pollutant and its concentrations increasing day by day in many metropolitan and industrial areas. It is a primary product of fossil fuels power plants combustion or from refining of sulfur-containing ores. It affects the human health and the global ecology (14). Plant growth stimulating rhizobacteria are beneficial bacteria that can also resist various stress conditions in plants. One of these stresses is SO_2 air pollution.(15)

WHAT IS PGPR?

Rhizobacteria that are root-colonizing bacteria, form symbiotic relationships with many plants. The name comes from the Greek *rhiza*, which means root. Though there are parasitic varieties of rhizobacteria, the term generally refers when the bacteria show beneficial relationship for both parties i.e. mutualism. PGPRs are an efficient group of microorganisms which are applied as Biofertilizer. Biofertilizers supply about 65% of the nitrogen to crops worldwide. Rhizobacteria are referred to as plant growth-promoting rhizobacteria, or PGPRs.[1]

Plant growth-promoting rhizobacteria (PGPR) were first defined by Kloepper and Schroth[2] which describe the soil bacteria that are involve in plant growth and development by colonizing the roots of plants following inoculation onto seed. (3)

MECHANISM OF PGPR'S ACTION:

PGPRs help in plant growth by both direct and indirect way, but the specific mechanism of PGPRs are not clearly characterized. [4]

Plant growth promoting rhizobacteria have direct mechanisms that is helpful in nutrient uptake or in increasing nutrient availability by the nitrogen fixation, solubulization of mineral nutrients, mineralization of different organic compounds and help in the production of phytohormones[4,11]

In the root hairs and lateral root of plant of Poaceae, PGPR colonize which may express the plant benificial properties. [27,28]

A PGPR strain have several plant-beneficial properties, which may be co-regulated or not. The PGPR strain which is a plant-beneficial properties affected by both abiotic factors (like pH, oxygen, clay mineralogy, heavy metals, etc.) and biotic factors (i.e., compounds produced by plants or the rhizo-microbiome).[28]

Direct mechanisms of plant growth promotion by PGPRs occur in the absence of plant pathogens or other rhizosphere microorganisms, while indirect mechanisms show the ability of PGPRs on the reduction of the effect of harmful pathogen on plant. PGPRs have been reported to directly increase plant growth by different mechanisms: fixation of atmospheric nitrogen transferred to the plant.[5]

Molecular approaches using microbial and plant mutants altered in their ability to synthesize or respond to specific phytohormones which have increased understanding of the role of phytohormone synthesis and it is a direct mechanism of plant growth improvement by PGPRs.[6]

ROLE OF PGPR:

NITROGEN FIXATION:

Nitrogen fixation is one of the most beneficial processes of plant growth which is represented by rhizobacteria. Nitrogen is an important nutrient to plants, as gaseous nitrogen (N_2) is not available to plants due to the high energy required to break the triple bonds between the two atoms.[7]

Rhizobacteria help in the growth of plant by converting gaseous nitrogen to ammonia through nitrogen fixation and make it an available nutrient for the plant. The host plant serves the bacteria with amino acids thats why they do not need to digest ammonia.[8]

Nitrogenase which is an enzyme involved in nitrogen fixation but requires anaerobic conditions and that is available in the membranes within root nodules. The rhizobacteria also require oxygen to metabolize. The oxygen is served by a hemoglobin protein known as leghemoglobin which is found within the nodules.[7]

Nitrogen (N) is the most important nutrient for plant growth and its productivity. Although,78% N_2 is found in the atmosphere which is unavailable for the growing plants. Then the atmospheric N_2 is converted into forms by biological N_2 fixation (BNF) which changes nitrogen to ammonia by nitrogen fixing microorganisms using a complex enzyme system known as nitrogenase.[9]

PHOSPHORUS SOLUBULIZATION:

Phosphorus is the very important key element for the nutrition of plants, next to nitrogen (N). It plays an important role in virtually all major metabolic processes in plant including photosynthesis, energy transfer, signal transduction, macromolecular biosynthesis and respiration [11]. It is present in soils in both organic and inorganic state. Plants are unable to utilize phosphate because 95-99% phosphate present in the insoluble, immobilized, and precipitated form [13]. Plants absorb phosphate only in two soluble forms, the monobasic (H_2PO_4) and the dibasic ($HPO_4^{\ 2-}$) ions [12].

Plant growth promoting rhizobacteria are able to solubulize the part of the bound phosphate of the soil and thereby make them available to the plants. They also help in making phosphorus available for plants to absorb. The main phosphate solubulization mechanisms is supervised by plant growth promoting rhizobacteria include:

(a) release of complexing or mineral dissolving compounds e.g. organic acid anions, protons, hydroxyl ions, CO_2.

(b) release of extracellular enzymes (biochemical phosphate mineralization).

(c) release of phosphate during substrate degradation (biological phosphate mineralization) [14].

Phosphate solubilizing PGPR such as *Arthrobacter, Bacillus, Beijerinckia, Burkholderia, Enterobacter, Erwinia, Flavobacterium, Microbacterium Pseudomonas, Rhizobium, Rhodococcus*, and *Serratia* have shown in the improvementof plant growth and yield as soil inoculam[12].

The phosphate solubilizing bacteria are used either alone or in combination with other rhizospheric microbes which have been also reported [15].

SIDEROPHORE PRODUCTION:

In ecosystem iron is one of the essential micronutrient for almost all organisms. But iron is fourth abundant element in Earth. In aerobic soils, iron is not readily assimilated by bacteria or plants because of ferric ion or Fe^{3+}.Ferric ion or $Fe^{3+}$is in predominant form in nature and it is only sparingly soluble. For this the amount of iron available for assimilation by living organisms is extremely low [16].

Microorganisms have evolved a specialized mechanisms for the assimilation of iron, including the production of low molecular weight iron-chelating compounds known as siderophores, which transport this element into their cells [17,18]. Depending on the characteristic functional group of siderophores are divided into three main families, i.e. 1)hydroxamates, 2)catecholates and 3)carboxylates. At present more than 500 different types of siderophores are known in which 270 have been structurally characterized [19].

The demonstration of direct benefits of bacterial siderophores on the growth and development of plants have been done by applying radio labeled ferric siderophores as a sole source of iron and it showed that plants are able to take up the labelled iron by a large number of plant growth promoting rhizobacteria including Aeromonas, *Azadirachta, Azotobacter, Bacillus, Burkholderia, Pseudomonas, Rhizobium, Serratia and Streptomyces* sp [20] and enhanced chlorophyll level compared to un inoculated plants [21].

IAA PRODUCTION:

Among all plant growth regulators, Indole Acetic Acid (IAA) is the most common natural auxin found in plants. Its positive effect can be found on root growth [23]. Up to 80% of rhizobacteria can synthesize Indole Acetic Acid (IAA).To stimulate the cell proliferation and enhance the host's uptake of minerals and nutrients from the soil colonizing the seed or root surfaces is proposed to act in conjunction with endogenous IAA in plant [22]. Indole Acetic Acid has great impact on plant cell division, extension, and differentiation. It stimulates seed and tuber germination and increases the rate of xylem and root development. It controls the processes of vegetative growth and initiates lateral and adventitious root formation. It mediates responses to light, gravity and florescence and also affects photosynthesis, pigment formation, biosynthesis of various metabolites and resistance to stressful conditions [24].

Tryptophan is an amino acid which is commonly found in root exudates. This has been identified as main precursor molecule for biosynthesis of IAA in bacteria [25]. The biosynthesis of indole acetic acid by plant growth promoting rhizobacteria (PGPR) involves formation via indole-3- pyruvic acid and indole-3-acetic aldehyde. This is the most common mechanism in bacteria like *Pseudomonas,* *Rhizobium, Bradyrhizobium, Agrobacterium, Enterobacter and Klebsiella* [26]. Low level of IAA secretion on root growth promotion by the free living PGPR e.g., *Alkaligenes faecalis, Enterobacter cloacae, Acetobacter dizotrophicous,* species of *Azospirillum, Pseudomonas and Xanthomonas* sp. was proven. However, microbially produced phytohormones are more powerful. Due to these reasons the threshold between inhibitory and stimulatory levels of chemically produced hormones is low, microbial hormones are more effective by virtue of their continuous slow release.

Several plant growth promoting rhizobacteria, *Azotobacter sp., Rhizobium sp., Pantoeaagglomerans, Rhodospirillumrubrum, Pseudomonas fluorescens, Bacillus subtilis* and *Paenibacillus polymyxa* can produce either cytokinins or gibberellins or both for plant growth promotion [30]. Cytokinins can also be synthesized by some phytopathogens. However, it appears that plant growth promoting rhizobacteria (PGPR) produce lower cytokinin levels compared to phytopathogens. This is why the effect of the plant growth promoting rhizobacteria(PGPR) on plant growth is stimulatory while the effect of the cytokinins from pathogens is inhibitory.

PHYTOHORMONE PRODUCTION:

Ethylene is a key phytohormone which has a wide range of biological activities. It can affect plant growth and development in a large number of different ways including promoting root initiation, inhibiting root elongation, promoting fruit ripening, promoting lower wilting, stimulating seed germination, promoting leaf abscission, activating the synthesis of other plant hormones [31]. The higher concentration of ethylene induces defoliation and other cellular processes that may lead to reduced crop performance [12]. The enzyme 1-aminocyclopropane-1 carboxylic acid (ACC) is required before hand for ethylene production. It is catalyzed by ACC oxidase. Iqbal MA et al [32] reported improved nodule number, nodule dry weight, fresh biomass, grain yield, straw yield, and nitrogen content in grains of lentil. As a result lowering of the ethylene production via inoculation with plant growth promoting strains of *Pseudomonas* sp. containing ACC deaminase along with *R.leguminosarum*. Currently, bacterial strains exhibiting ACC deaminase activity have been identified in a wide range of genera such as *Acinetobacter, Achromobacter, Agrobacterium, Alcaligenes, Azospirillum, Bacillus, Burkholderia, Enterobacter, Pseudomonas, Ralstonia, Serratia and Rhizobium* etc. [30].

Plant growth promoting rhizobacteria(PGPR) is a promising, sustainable and environment friendly approach to obtain sustainable fertility of the soil and plant growth indirectly. This approach takes inspire a wide range of occlusion of plant growth promoting rhizobacteria. This will reduce the need for agrochemicals (fertilizers and pesticides) to improve soil fertility by a variety of mechanisms via production of antibiotics, siderophores, HCN, hydrolytic enzymes etc.[33,34]

ANTIBIOTIC PRODUCTION:

The antibiotic production is considered to be one of the most powerful and studied biocontrol mechanisms of plant growth promoting rhizobacteria against phytopathogens. This has become increasingly better understood over the past two decades [35].

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 [36] and oligomycin A, kanosamine, zwittermicin A, and xanthobaccin produced by *Bacillus*, *Streptomyces*, and *Stenotrophomonas* sp. to prevent the proliferation of plant pathogens (Generally fungi) [37].

Pseudomonas sp. which is an antibiotic 2, 4-diacetylphloroglucinol producing bacteria in soil was reported for biocontrol of disease in wheat which caused by the fungus *Gaeumanomyces graminis* var. tritici [38]. Bacterial action on wheat seeds with *P. fluorescens* strains produce the antibiotic phenazine-1- carboxylic acid (PCA) which resulted in significant suppression of take-all in about 60% of field trials [39]. *Bacillus amyloliquefaciens* is known for lipopeptide and polypeptide production for biological control activity and plant growth promotion activity against soil borne pathogens [40].

Hydrogen Cyanide (HCN) PRODUCTION:

Apart from the production of antibiotic, some rhizobacteria are also capable of producing volatile compound known as hydrogen cyanide (HCN) for biocontrol of black root rot of tobacco, caused by *Thielaviopsis basicola* [41]. Lanteigne et al. [42] also reported that the production of DAPG and HCN by *Pseudomonas* contribute to the biological control of bacterial canker of tomato.

Plant growth promoting rhizobacterial strains can produce certain enzymes such as chitinases, dehydrogenase, β-glucanase, lipases, phosphatases, proteases etc. [43,44] They can exhibit hyperparasitic activity, attacking pathogens by excreting cell wall hydrolases. Through the activity of these enzymes, plant growth promoting rhizobacteria play a very significant role in plant growth promotion particularly to protect them from biotic and abiotic stresses by suppression of pathogenic fungi including *Botrytis cinerea, Sclerotium rolfsii, Fusarium oxysporum, Phytophthora sp., Rhizoctonia solani, and Pythium ultimum* [45,46].

Induced resistance may be defined as a physiological state of enhanced defensive capacity elicited in response to specific environmental stimuli and consequently the plant's innate defenses. They are potentiated against subsequent biotic challenges [47]. Some plant growth promoting rhizobacteria with biopriming plants can also provide systemic resistance against a broad spectrum of plant pathogens. Diseases of fungal, bacterial, and viral origin and in some cases even damage caused by insects and nematodes can be reduced after application of plant growth promoting rhizobacteria (PGPR)[48].

CONCLUSION: PGPR AS BIOFERTILIZER:

The increased number of PGPRs for various crops and vegetables has been commercialize as a biofertilizer, recently includes *Pseudomonas*[51], *Azospirillum* [52]*Bacillus* [53], *Stenotrophomonas* [53], *Rhizobium*[57], *Serratia* [55], *Azotobacter, Klebsiella, Enterobacter, Alcaligens, Arthrobacter, Burkholderia* [55] and *Streptomyces* [55]. *Pseudomonas* and *Bacillus* are the most commonly investigated PGPR and often dominate the rhizosphere [56].

There are various effects of plant growth-promoting rhizobacteria (PGPR) such as seed germination, seedling growth and yield of field grown maize were evaluated in three experiments. [58]

Plant growth promoting effects of PGPR strains in different crops were clearly demonstrated [59]. Bacterial inoculants are able to increase plant growth and germination rate, improve seedling emergence, responses to external stress factors and protect plants from disease[60] .

Improvement of seed germination parameters occur by rhizobacteria has been reported in other cereals such as sorghum [61] and pearl millet [62,63]

The improvement in seed germination by PGPR was also found to work with wheat and sunflower[64,65], where it was found that some PGPR induced increases in seed emergence, in some cases achieving increases up to 100% greater than controls. These findings may be due to the increased synthesis of hormones like gibberellins, which would have triggered the activity of specific enzymes that promoted early germination, such as amylase, which have brought an increase in availability of starch assimilation. Beside this, significant increase in seedling vigor would have occurred by better synthesis of auxins [66].

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