**PSEUDOMONAS: UNIQUE PLANT GROWTH PROMOTING RHIZOBACTERIA**

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**Abstract:** Plant growth promoting rhizobacteria promote plant growth and productivity has internationally been accepted. Fluorescent pseudomonas have been extensively studied as a plant growth promoting rhizobacteria (PGPR). Pseudomonads are known to interact with host plant via chemical mediators that develop a symbiotic relationship. During their close association, they influence the growth of host plant by delivering beneficial effects in rhizosphere. Pseudomonads promote the growth of plants either by direct supply of nutrients, synthesis of phytohormones, solubilization of minerals, or indirectly as a biocontrol agent suppressing the pathogens. Cumulative effect of combination of above properties projects it as bacteria of great economic importance. This is usually achieved by either one or blend of several factors released in rhizosphere by symbiont. These include secretive secondary metabolites e.g. Antibiotics, toxins, enzymes, HCN etc. which inhibits the pathogen or chelators like siderophores which generate the microenvironment, a competitive one for pathogen. These bioformulations also enhanced soil fertility and the PGPR activity also increased grain yield.

**Keywords:** Growth promoting, Pseudomonas, Rhizobacteria

**INTRODUCTION**

Soil-born plant pathogenic fungi cause heavy crop losses all over the world. With its variable climate in different ecological zones, major crops in India are susceptible to diseases caused by soil borne fungal pathogen in the light of present day constraints on plant disease control practices. Chemical control is used to control pathogen but Chemical farming disturbs environment, subvert ecology, degrade soil productivity, mismanage water resources (Ayala and Rao, 2002; Deshwal et al., 2011a). The chemical agriculture affected the soil environment is well known. Biofertilizer such as microbial inoculant which can promote plant growth and productivity have internationally been accepted as an alternative source of chemical fertilizer. PGPR represent a wide variety of soil bacteria which, when grown in association with a host plant, result in stimulation of growth of their host (Deshwal et al., 2003; Deshwal et al., 2010; Deshwal et al., 2011a) Biocontrol provides an alternative means of reducing these pathogens, which are otherwise difficult to control due to their survival strategies. The renewed interest in biocontrol is due to its environmental friendliness, long lasting effect and safety feature. Some of the bacterial antagonists have however also been found to show direct growth promoting effect on crop plants as inoculants and hence these effect plant growth by increasing phosphorous and nitrogen uptake and providing roots with compound such as phytohormones and solubilised iron, but also release antibiotics and chelate iron (Glick et al., 1995; Deshwal et al., 2012). Fluorescent pseudomonads make up a dominant population in soil along with other bacteria like Serratia, Azotobacter, Clostridium, Bacillus, Arthrobacter, Alcaligenes, Agrobacterium, Bradyrhizobium and Rhizobium. Collectively these bacteria have been termed “Rhizobacteria”. Certain fluorescent pseudomonads increase yield or control biologically one or more soil-borne plant pathogens when applied as seed or seed piece inoculants to agriculture crops (Burr and Caesar, 1984; Deshwal et al., 2011b). When PGPR strains are applied in field, the root exudates also enhanced which attract the rhizosphere microbes (Prinkyl and Vancura, 1980) and *Pseudomonas* (biological control) by production of siderophore that sequester iron in the root environment, making it less available to competing deleterious microflora (Kloepper et al., 1980, Bagnasco et al., 1998; Deshwal et al., 2012) Some strains of *Pseudomonas fluorescense* and *P.putida* were reported to be biological control agent against various soil borne fungi (Weller, 1988). Lim and Kim (1995) reported that *Pseudomonas* controlled *Fusarium solani* by production of metabolites which decompose the fungal wall. Bagnasco et al. (1998) reported that fluorescent pseudomonads suppressed the growth of phytopathogenic fungi *Pythium ultimum* and *Rhizoctonia solani*. Similarly Lim et al. (1998) noted the role of siderophore in biological control of *Fusarium solani*. Gupta et al. (1999) observed that fluorescent pseudomonas strain GRC inhibited growth of *Macrophomina phaseolina* and *Fusarium oxysporum* in vitro. Recently, Deshwal et al. (2012) reported that *Pseudomonas aeruginosa* strains inhibited the growth of *Sclerotina sclerotiorum* by 62-83% inhibition zone as compared to control. *Pseudomonas* MR-18 strains showed maximum inhibition. *In vitro* study revealed that *Pseudomonas* strains effectively reduced the growth of *Sclerotina sclerotiorum*. The production of antimicrobial compounds by some pseudomonas strains has been recognized as a major factor in the suppression of many root pathogens (O’Sullivan and O’ Gara, 1992). *Pseudomonas* improves the root colonization and higher population control the growth of deleterious pathogens (Expert...
Fluorescent pseudomonads have another merit on controlling several phytopathogenic fungi besides simultaneously enhancing the growth and yield of various vegetables, mustard or rape seed and cereals (Iswandi et al., 1987; Bagnasco et al., 1998). Lim and Kim (1995) reported that Pseudomonas reduced disease incidence and controlled Fusarium solani by production of metabolites which decompose the fungal wall. Bagnasco et al. (1998) reported that fluorescent pseudomonads suppressed the growth of phytopathogenic fungi Pythium ultimum and Rhizoctonia solani. Similarly Lim et al. (1999) noted the role of siderophore in biological control of Fusarium solani. Deshwal et al. (2012) reported that Pseudomonas aeruginosa MR-18 showed maximum inhibition zone against Sclerotinia sclerotiorum.

(ii) Improve root colonization

Beneficial rhizobacteria are known to colonize rapidly and aggressively the root system, suppress pathogenic microorganism, and enhance plant growth and development (Weller, 1988). A successful antagonist should colonize the rhizosphere at the time of seed germination itself or another word, the antagonist should move from spermosphere to rhizosphere and establish there (Weller, 1983). Gu and Mazzola (2001) suggested the application of procedures to enhance colonization of the rhizosphere by biocontrol. Fluorescent pseudomonas can lead to improved management schemes to control soil borne plant pathogens as observed by large number of workers (Seong and Shin 1996; Lim and Kim, 1997; Bagnasco et al., 1998; Gupta et al., 1999). Poor colonization of the distal parts of the root has been frequently observed in seed inoculation treatments (Hatzinger and Alexander, 1994).

(iii) Beneficial to other microbes

Polonenko et al. (1987) demonstrated that some pseudomonads stimulated nodulations of legumes by Rhizobium spp. and Bradyrhizobium spp. Furmann and Wollum (1989) found that three fluorescent pseudomonads that consistently increased nodule occupancy of the more efficient B. japonicum USDA 123 and USDA 31 (now B. elkanii) in soybean and presumably because of siderophore production. Deshwal and Vig (2011) mentioned that Pseudomonas PVK-23 improved Rhizobium RVK-21 and enhanced plant growth activity of Peanut.

(iv) Enhancement of plant by uptake of several nutrients

Pseudomonads enhance the plant growth productivity by production of metabolites such as siderophore, HCN, chitin and β-1,3-glucanase, antibiotic production which control the pathogenic

Field application

The distribution of the rhizobacteria on hybrids of maize was of Pseudomonas spp. This was observed to be to be most prominent group. The effect of PGPR on crop plants has been evaluated under field condition (Lim et al., 1999). PGPR enhanced plant yield in a wide range of crop plants such as vegetables, rapeseed (Brown, 1974; Iswandi et al., 1987), spring and winter wheat by 8-16% (Brown, 1974), sugar beet to 7-10% (Suslow and Schroth, 1982) and in radish 60-144% (Schroth and Hancock, 1981). Deshwal et al. (2011a) mentioned that Pseudomonas aeruginosa improved plant growth in Mucuna plant.

(i) Reduction of phytopathogens
microorganism in rhizosphere. Pseudomonads produces IAA production, phosphate solubilization, vitamins by which plant root elongation occurs, rhizosphere bacterial population is also improved. Such PGPR activities are given below:

(a) Siderophore production

Competition for iron is another mechanism by which fluorescent pseudomonads may inhibit the growth of pathogens. Siderophores of PGPR in the rhizosphere under iron deficient environment could efficiently chelate environment iron and inhibit the growth of native microflora including root pathogen (Lim and Kim, 1990). The fluorescent pseudomonads are characterized by their production of yellow green pigments, termed pyoverdines or pseudobactins, that fluorescence under UV irradiation and function as siderophore (Abdallah, 1991). Pyoverdines produced by fluorescent pseudomonads inhibit the growth of variety of microorganisms in iron–depleting culture media (Buyer et al., 1989). In most biocontrol strains in which siderophore production has been described as an important mechanism but may contribute to control of pathogen when soil condition are favorable for production (Thomshow and Weller, 1995).

(b) HCN production

Pseudomonads produce HCN which control the root rot pathogen. Thomshow and Weller (1995) observed same observation that pseudomonads exert there beneficial effect on plants by the production of diverse microbial metabolites like HCN. Bagnasco et al. (1998) reported that Pseudomonas strains produced HCN. Gupta et al. (1999) reported that fluorescent pseudomonas GRC1 produced HCN and resulted in inhibition of growth of pathogenic fungus M. phaseolina and Fusarium oxysporum. Deshwal et al. (2011a, b) mentioned that Pseudomonas strains isolated from Mucuna produced HCN. Recently, Deshwal et al. (2012) mentioned same observation.

(c) Chitin and β-1, 3-glucanase production

Hyperparasitism and lysis are the most important forms of biological control of soilborne plant pathogens by microbial antagonists. The mechanism appears to involve the enzymatic hydrolysis of chitin and glucan component of fungal hyphal walls. The lytic activity of bacterial and fungal antagonists is mainly due to the extracellular lytic enzymes chitinase and β-1, 3-glucanase (Henis and Chat, 1975). An antifungal Pseudomonas stutzeri YPL-1 produced extracellular chitinase and β-1, 3-glucanase that were key enzyme in the decomposition of fungal hyphal walls. These lytic extracellular enzymes markedly inhibited mycelial growth of the phytopathogenic fungus Fusarium solani (Lim and Kim, 1995). These enzymes have been found to be responsible for the control activity of some bacteria (Inbar and Chet, 1991). Chernin et al. (1995) reported that gluconase and chitin, lytic enzyme which degrade the fungal walls, also are involved in the biocontrol against certain pathogens.

(d) Antibiotic production

During the past decade many researchers have focused on the role of antifungal metabolites. The production of antimicrobial compounds by some strains of Pseudomonas has been recognized as major factor in the suppression of many root pathogens (Dowling and O’Gara, 1994). Key antibacterial compounds include phenazines (Thomshow and Weller, 1988) and 4-diacylphloroglucinol “DAPG” (Shanahan et al., 1992).

(e) Plant growth promoting hormones

Gupta et al. (2002) isolated the IAA producing fluorescent pseudomonads in the potato rhizosphere. Glick et al. (1999) reported that IAA producing rhizobacteria enhanced the root length which is one of the plant growth promoting activity rhizobacteria. Rhizobacteria also produce Gibberellic acid (Mahmoud et al., 1984), cytokinins (Tien et al., 1979), and ethylene (Glick et al., 1995). Deshwal et al. (2011c) reported that Pseudomonas strains improve plant growth in soybean crop.

(f) Phosphate solubilization

Unsolubilized phosphate is not taken up by plant but some rhizobacteria solubilize phosphate that is readily taken up by plant. Whitelaw et al. (1997) reported that some P- solubilizing organism have been reported as plant growth promoters. Chabot et al. (1996) observed that P-solubilizing rhizobacteria increased plant growth, productivity in maize, lettuce. Pseudomonad is also P-solubilizer (Gupta et al., 2002). Deshwal et al. (2011c) observed that Pseudomonas strains solubilized Phosphorous. Again, Deshwal et al. (2011d) reported Phosphorus solubilizing Pseudomonas aeruginosapart of PMV-14 enhanced productivity in Rice crop.

(g) Vitamins

Certain fluorescent pseudomonads included among plant growth promoting rhizobacteria, have a direct effect on the plant growth by the production of vitamins (Lifshitz et al., 1987). Growth factors such as vitamins are also produced by pseudomonads and also indirectly effect the growth of rhizobia in the rhizosphere (Derylo and Skorupska, 1993), reason behind this is some strains of genus of rhizobia don’t produces biotin, these strain may benefit when grown with biotin producing bacteria.
Factors affecting bioformulation in rhizosphere

According to Baker (1968) competition for particular nutrient is an operational mechanism for bio control. The addition of such nutrient should therefore, must eliminate the bio controls activity. Unless an organism can compete with other organism and effectively scavenge and utilize the available nutrients only then it will constitute a significant proportion of the rhizoplane population (Hattori, 1988). The activity of biocontrol agents is influenced profoundly by extrinsic factor of the environment. Microorganism competes with each other for carbon source, mineral and infection sites of roots. The efficient competitors should have the ability to colonize at faster rate so as to displace pathogen for disease suppression. The competitive exclusion of deleterious rhizospheric organisms is directly linked to an ability to successfully colonize the root surface. This mechanism has been suggested to play a role in biocontrol by fluorescent Pseudomonas species against S. sclerotiorum, M. phaseolina, Fusarium spp. (Elad and Baker, 1985; Gupta et al., 2001).

Benefits

Chemical control of disease is very harmful to human as well as they reduce the soil fertility so there is need to select such a type of alternative which is not harmful and also improves soil fertility and increase microbial niche. So, Pseudomonads is one of the potential bacterial, which controlled the pathogens and also enhanced the plant growth by production of metabolites.

In many crop pathogen systems, the primary mechanism of biocontrol by Pseudomonas is production of antibiotics (Shanahan et al., 1992). Under certain conditions, antibiotics improve the ecological fitness of these rhizobacteria in rhizosphere, which can further influence long-term biological control efficacy (Shanahan et al., 1992). Pseudomonads have capacity to uptake iron from soil and this chelation reduces the chlorosis in plant (Bienfait, 1989). Siderophores that chelate iron and other metals also contributes to disease control by confirming a competitive advantage to biological control agent for the limited supply of essential trace minerals in natural habitats (Höfte et al., 1994). Siderophore producing strain P. fluorescens GL 20 inhibited spore germination and hyphal growth of Fusarium solani because strain uptake iron which is necessary for germination and growth of fungus (Lim et al., 1999). Other biological control metabolites such as HCN, extracellular lytic enzymes like chitinase and β-1, 3-glucanase also controls deleterious pathogens. Glick et al. (1998) observed that seven bacterial strains which were positive for ACC deaminase production promoted canola (Brassica napus L.) seedling root elongation under gnotobiotic conditions.

Paulitz (1991) proposed that bacterial antagonist may catabolizes specific component of seed exudates that function as chemical “signals” in triggering sporangial germination of Pythium ultimum. Such observation have exciting implications for the potential specificity of interaction between bacterial antagonists and target soil borne pathogen that is reminiscent of the specific interactions between epiphytic bacterial populations. It has been observed that biocontrol pseudomonads potentially show more competency of establishment to form the root colonization of host plant in pathogenic soil (Bagnasco et al., 1998). So it gives clear-cut advantage with potential biologically active Pseudomonas. Root colonization is an important and first step in the process of interaction of beneficial bacteria with plants (Kloepper and Beauchamp, 1992). Pseudomonas is well-known biocontrol agent and rhizobia are known as symbiotic N₂ fixer. When co-inoculated, they enhanced the growth and yield of forage legume (Fuente et al., 2002). Deshwal and Vig (2010) mentioned that Co-inoculation of Pseudomonas-MP3 and Rhizobium-GR-23 enhanced plant growth activity of Peanut (Arachis hypogaea L.).

Pseudomonas in Field trials

Pseudomonas fluorescense strain F113 produces antibiotics “DAPG”, which colonizes the rhizosphere of sugar-beet seedling and controls damping-off disease in soil microorganisms prepared with Pythium infection (Fenton et al., 1992). In pot trials of bean with P. fluorescens GL 20, disease incidence was remarkably reduced up to 94% compared with Fusarium solani (Lim et al., 1999). Cattelan et al. (1999) observed that Pseudomonas GN 1201 increased significantly the dry shoot weight, root length and root dry weight in soybean crop as compared to control. Pseudomonads increase plant growth and grain yield in wide range of crops like rapeseed (Hofte et al., 1991), kidney beans (Lim and Kim, 1997) and Wheat (Raaijmakers et al., 1999). Further, experiments carried out by Prikryl and Vancura (1980) showed that root colonized by microorganism like Pseudomonas release up to twice the amount of root exudates released aseptically is unclear, it could be due to the ability of some bacteria to increase the cell permeability of roots as was proposed by Bowen (1980). Expert and Degit (1995) observed that antagonistic strain Ti-5 control growth of S. sclerotiorum when at least 1x10⁶ antagonistic bacteria in the spermosphere are present and successfully colonize the rhizosphere so it shows that biocontrol bacterial population also play an significant role to control pathogens.

Future Research Priorities
At present only a few of them have been identified as biological control agent. By intensifying and diversifying the research, we stand to diversify the base of mechanisms, in turn would increase the chance for formulating biological control agent or mixture of agents that will provide predictable control. It is, therefore, essential to continue the screening process to obtain potential strains.

In order to harness potential benefits of biofertilizer in commercial agriculture, the consistency of their performance must be improved. This requires research in many diverse area as these biological system involves complex interactions among the host, other rhizosphere microflora and fauna and the environment. The survival and competitive ability of the strains to be introduced must be improved. Very little is known about the competitiveness of the microorganism and factors governing it. These biological bacteria some time inhibit the pathogen in vitro but fail in field trails. Question arise that what are the factors that control the biological control in field or other bacteria which inhibit biocontrol bacteria or control their biocontrol activity.

Research to understand the mechanism by which the introduced microorganism benefits the crop is critically important. Although pseudomonads are well known for their specific function biocontrol activity, siderophore, HCN, IAA and phosphate uptake, research has shown that these properties improved crop productivity. So it is better when the biological control bacteria have some other plant growth productivity activity so that it can control deleterious bacteria by competition and enhance plant growth productivity. Lot of research should gain considerable attention in future in this field.

CONCLUSION

The plant pathogen presence and higher use of chemical control reduce plant growth as well as total yield. This causes an enormous loss of productivity. Although chemical treatments show immediate effect but disturb soil environment, contaminate underground water as well as proved toxic to mammals. Moreover, repetitive application of these chemicals develops resistant variety of target pathogens, which respond weakly or may even remain completely unaffected. This again projects a series of problems with metastatic effect over environment, economy as well as human population. On the other hand, biocontrol agents are cheaper, have no harmful effect on human population, they have long term effect in soil and markedly, showed no deleterious effect on environment.

Plant growth promoting rhizobacteria can affect plant growth either directly or indirectly. The direct promotion of plant growth usually entails by providing the plant either a compound viz. synthesized by bacterium and / or, bacteria facilitate the uptake of nutrient from soil by the plant. The indirect promotion of plant occurs when PGPR prevent the deleterious effect of one or more pathogens. It is therefore, desirable to replace these chemical agent with biological control agents which are eco-friendly and non-hazardous.

Pseudomonads can produce a variety of substances that can limit or inhibit the growth of pathogen in spermosphere, rhizosphere and phyllosphere. Although inoculation of seed with large numbers of bacterial cells result in the establishment of population of the introduced strain in rhizosphere (Suslow, 1982), the size of these populations is generally only a fraction of that of total heterotrophic bacteria that can be culture in the rhizosphere (Loper et al., 1984). Thus, the impact of introduced population on precluding population establishes a small competition in the rhizosphere. Moreover, pseudomonads show synergism with different type of resident soil microorganism such as rhizobia and result in plant growth enhancement. Now scientists are concerned with Pseudomonas that has capability to control pathogens along with unique characteristic such as phosphate solubilization and IAA production. Such type of PGPR activity also enhanced the plant growth. Enough phosphate is present in soil but plants are not able to uptake total required phosphate for growth, main reason behind this is most of phosphate present in soil is not in solubilized form but pseudomonads have capability to solubilize phosphate, and then plants can uptake it. Many researchers observed that P-solubilizing rhizosphere bacteria increased plant growth and productivity. Pseudomonads also produced plant growth promoting hormones. These hormones enhanced the root-elongation, root-branching, plant height, leaf area etc.

REFERENCES


