

Induction and modulation of resistance in crop plants against disease by bioagent fungi (arbuscular mycorrhiza) and hormonal elicitors and Plant Growth Promoting Bacteria

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ABSTRACT: Arbuscular mycorrhizal (AM) fungi and bacteria and hormonal elicitors can interact synergistically to stimulate plant growth through a range of mechanisms that include improved nutrient acquisition and inhibition of fungal plant pathogens. These interactions may be of crucial importance within sustainable, low-input agricultural cropping systems that rely on biological processes rather than agrochemicals to maintain soil fertility and plant health. Although there are many studies concerning interactions between AM fungi and bacteria and hormonal elicitors the underlying mechanisms behind these associations are in general not very well understood, and their functional properties still require further experimental confirmation. Future mycorrhizal research In this context, the present article seeks to review and discuss the current knowledge concerning interactions between AM fungi and plant growth promoting rhizobacteria, and hormonal elicitors physical interactions between AM fungi and bacteria, and hormonal enhancement of phosphorus and nitrogen bioavailability through such interactions, and finally the associations between AM fungi and their bacterial endosymbionts. Overall, this review summarizes what is known to date within the present field, and attempts to identify promising lines of future research.

Keywords: Mycorrhizal fungi, interrelations, plant growth promoting rhizobacteria, hormonal elicitors

INTRODUCTION

The first clear indication of improved plant growth and biological control of root pathogens due to seed bacterization with rhizobacteria came from the works of (Burr et al., 1978) and (Kloepper et al., 1980) who reported the plant growth promoting effects of *Pseudomonas* strains which were antagonistic to a wide range of plant pathogens *in vitro*. Rhizobacteria have also been studied as plant growth promoters for increasing agricultural production and as biocontrol agents against plant diseases (Burris, 1998; Chen et al., 1996).

Plant growth promoting rhizobacteria (PGPR) exhibit direct and indirect mechanisms as plant growth promoters and biological control agents. Direct mechanisms by PGPR, include the provision of bio available phosphorus for plant uptake, nitrogen fixation for plant use, sequestration of iron for plant by siderophores, production of plant hormones like auxins, cytokinins and gibberellins and lowering plant ethylene levels using ACC deaminase that accumulate during biotic and abiotic stresses (Mayak et al., 2004). Indirect mechanisms of PGPR include production of antibiotics, viz. 2,4-Diacetyl phloroglucinol (DAPG), phenazine, pyoluteorin and pyrrolnitrin against pathogenic fungi and bacteria, reduction of iron available to phytopathogens in the rhizosphere, synthesis of fungal cell wall and insect-gut membrane lysing enzymes, chitinase enzyme for hydrolysis of chitin layer of the

eggshell of nematode and also competition with detrimental microorganisms for sites on plant roots and induction of systemic resistance against various pathogens and pests in plants (Ramamoorthy et al., 2001). These studies also provided the first evidence that the rhizosphere microbiota could be modified significantly with microorganisms introduced with the planting material. (Kloepper et al., 1989) coined the term plant growth promoting rhizobacteria (PGPR) to include bacteria inhabiting the root and rhizosphere soil which have the ability to increase plant growth. Plant Growth-Promoting Rhizobacteria (PGPR) have been reported to be key elements for plant establishment under nutrient-imbalance conditions (Egamberdiyeva and Höflich, 2004). PGPR may improve plant growth and yield by direct and indirect mechanisms (Noel et al., 1996). Indirect mechanisms of plant growth stimulation include a variety of mechanisms by which the bacteria prevent phytopathogens from inhibiting plant growth and development (Glick and Bashan, 1997). Direct mechanisms may act on the plant itself and affect growth by providing plants with fixed nitrogen, phytohormones, iron and soluble phosphate (Kloepper and Schroth, 1978).

PGPR can also protect plants from the deleterious effects of some environmental stresses including flooding (Grichko and Glick, 2001), drought (Mayak et al., 2004a), salt (Mayak et al., 2004b) and phytopathogens (Harman and Björkman, 1998). *Bacillus subtilis* can induce plant resistance to stress and produces various plant hormones for growth improvement (Han and Lee, 2005). Many workers have showed that inoculation of plants with *B. subtilis* increased plant growth, yield and nutrient uptake, especially under salt stress conditions (Ashraf et al., 2004; Saleh et al., 2005), by influencing phytohormone production (auxin, cytokinin, or gibberellin), and/or by enzymatic lowering of plant ethylene levels (Glick, 2001). Arbuscular Mycorrhizal Fungi (AMF) form symbiotic associations with the roots of most plant species (Al-Karaki and Al-Raddad, 1997). These symbiotic associations can enhance plant growth and nutrient uptake under various environmental stress conditions such as salinity, drought and low fertility (Zuccarini and Okurowska, 2008). Also under conditions of low nutrient availability the hyphae of AMF can absorb nutrient from soil beyond the zone depleted by roots so they increase the effectiveness with which the soil volume is explored (Smith and Read, 1997). The beneficial effect of mycorrhizal fungi on plant growth was attributed to enhanced phosphorus uptake (Al-Karaki et al., 2001). Some authors also point out how AM fungi can increase plant resistance to salt stress by influencing the hormonal balance of the host plant (Danneberg et al., 1992) or by increasing water uptake (Ruiz-Lozano and Azcon, 1995).

It has been suggested that microorganisms isolated from the root or rhizosphere of a specific crop adopted better to that crop and provide effective control of diseases than organisms originally isolated from other plant species. Such plant associated microorganisms serve as better biocontrol agents because they are already closely associated and adapted to the plant or plant part as well as to the particular environmental condition in which they are supposed to function. The screening of such locally adopted strains has yielded improved biocontrol strains in some cases (Cook and Baker, 1983). However, now-a-days microbial biodiversity studies have enhanced the identification of potential bioagents suited to varied environmental conditions.

Finally to use antibiotic production, growth promotion, biocontrol potential and induced responses of PGPR as an effective pathogens management tool, we should evaluate their effects on plant performance and yield under agricultural settings. Although induction of disease resistance using arbuscular mycorrhiza fungi or hormonal inducers separately is well documented, there is little information about the mechanism of defense when used together. Thus, the objective of the present work was carried out to study the possible actions of the interaction

Between AM fungi and hormonal elicitors and PGPR in the induced defense mechanisms in plants against diseases.

1.1 RHIZOBACTERIA AS BIOCONTROL AGENTS

Rhizobacteria are ideal for use as biocontrol agents since they inhabit the rhizosphere that provides the front line defense for roots against attack by pathogens. Pathogens encounter antagonism from rhizobacteria before and during their primary infection of roots. Rhizobacteria are reported to provide protection against diverse plant pathogens.

(Sedra and Malouhy, 1994) studied six antagonists from 420 samples obtained from conducive and suppressive soils, for their inhibitory activity against *F. oxysporum* f.sp. *albedinis*. These antagonists suppressed the growth of *F. oxysporum* f.sp. *albedinis* in vitro by 24-47 per cent and its sporulation by 70-99 per cent. (Gupta et al., 1999) isolated *P. aeruginosa* from potato rhizosphere that displayed a strong antagonistic activity against important fungal pathogens, viz. *Macrophomina phaseolina* and *Fusarium oxysporum*.

(Tripathi and Johri, 2002) studied the biocontrol potential of fluorescent *Pseudomonas* isolated from rhizosphere of pea and wheat in vitro and in vivo against maize sheath blight caused by *Rhizoctonia solani*. They found some isolates to possess multiple disease control potential, while some others exhibited biocontrol potential

against specific pathogens, which indicated that fluorescent pseudomonads are diverse with respect to their biocontrol potential. (Ahmadzadeh et al., 2004) reported that antagonistic rhizobacteria, more specifically fluorescent pseudomonads and certain *Bacillus* species possessed the ability to inhibit fungal and bacterial root diseases of agricultural crops.

Plant growth-promoting rhizobacterial strain belonging to fluorescent pseudomonads was isolated from the rhizosphere of rice and sugarcane. Among 40 strains that were confirmed as fluorescent pseudomonads, 18 exhibited strong antifungal activity against *Fusarium oxysporum* and *Rhizoctonia bataticola*, mainly through production of antifungal metabolites (Ramesh Kumar et al., 2002).

(Tiwari and Thrimurthy, 2007) reported that twenty-one isolates of *Pseudomonas fluorescens* were isolated from the rhizosphere of rice, maize, wheat, chickpea, mung, urd, soybean and sunflower from Raipur and Bastar regions. Among these seven isolates which showed bright fluorescence under UV light were further tested. The isolates showed positive response of siderophore production and plant growth promoting activity on rice cv. Bamleshwari. Among the isolates PFR 1 and PFR 2 were found significantly superior to control in increasing the shoot length and root length. In vitro evaluation of the *P. fluorescens* isolates also confirmed their antagonistic ability against both *Pyricularia grisea* and *Rhizoctonia solani* in dual culture tests.

Pure culture of *P. aeruginosa* was obtained from the soil and studied for siderophore production. The antifungal activity of the strain against three phytopathogenic fungi, viz. *F. moniliformae*, *Alternaria solani* and *Helminthosporium halodes* was assayed by poison food technique. Inhibition of these fungal pathogens appeared to be due to production of antifungal secondary metabolites by *P. aeruginosa* (Kanika Sharma et al., 2007).

Various antagonistic rhizobacteria have been reported to be active in the rhizosphere and shown to play a significant role in suppressing the population and activity of *R. solanacearum*. The mechanisms by which they bring about the beneficial effect vary. (Jagadeesh, 2000) reported that bacterial wilt of tomato caused by *R. solanacearum* was controlled by the rhizobacteria to an extent of 16.66 to 83.33 per cent. Inoculation of three strains (fluorescent pseudomonas strain RJA112 and RBG 114 and *Arthrobacter* RBE 201) and the reference strain (*P. fluorescens* CHAO) resulted in 83.33 per cent disease control.

(Anith et al., 2004) reported that when PGPR (*Pseudomonas putida*, *Bacillus pumilus*) and Actigard (acibenzolar-S-methyl) application were combined, the bacterial wilt incidence caused by *R. solanacearum* was reduced when compared to the untreated control.

(Sikora, 1990) found that *P. fluorescens* exhibited an in vitro repellent effect towards *R. similis* and *Meloidogyne* spp.

(Jonathan et al., 2000) studied the efficacy of plant growth promoting uncharacterized actinomycetes (strain 29 and 45) and the nematode parasitic bacterium *Pasteuria penetrans* (isolate 100) against *M. incognita* race 1 on tomato and banana. Seed treatment with *P. fluorescens* and *P. chlororaphis* significantly reduced the root gall of *M. incognita* race 1 in tomato cv. Rutgers (Jonathan et al., 2000).

Pseudomonas fluorescens, *Bacillus* spp. and arbuscular mycorrhizae were tested against *M. incognita* and *Tylenchulus semipenetrans* in horticultural crops such as citrus, tomato, potato and chilli. The results showed that these organisms could be used as successful biocontrol agents for the management of plant parasitic nematode (Rajendran et al., 2001). (Seenivasan and Lakshmanan, 2001) studied the nematotoxic effects of culture filtrates of *P. fluorescens* strain Pf1 on *Hirschmanniella gracilis* at 25, 50, 75 and 100 per cent concentration in vitro. Application of *P. fluorescens* or *B. subtilis* increased the growth and yield of chickpea and reduced the infection by *M. incognita* by minimizing the number of galls/root system, egg mass production and soil population (Khan et al., 2001). Mortality of *M. incognita* juveniles was observed to be similar both in unheated and heated culture filtrates of *P. fluorescens* and the mortality increased with increase in concentration (Sirohi et al., 2000).

(Devi and Dutta, 2002) reported that bhendi seeds treated with *P. fluorescens* for 12 hours exhibited significant increase in growth and reduced root galling of *M. incognita*. Culture filtrates of *Pseudomonas fluorescens* caused significant reduction in egg hatching of *M. incognita* and resulted in considerable reduction in nematode population densities in soil and subsequent root knot development in tomato (Khan and Akram, 2000).

(Jonathan et al., 2006) reported that there was highest reduction in nematode egg hatch and the greatest mortality of *M. incognita* juveniles in the culture filtrate of *P. fluorescens* strain Pfb22 at 100 per cent concentration. The nematode infestation was reduced both in soil and roots, with the least number of adult females, number of egg masses, number of eggs per egg mass and gall index of *M. incognita* in banana plants treated with the local isolate of *P. fluorescens* Pfb22 under glasshouse conditions. Cultures and cell free culture filtrates (CFC) of 133 bacterial strains at three different dilutions (S-4, S-16, S-64) were tested in vitro against *Meloidogyne javanica* J2 mortality. Sixteen isolates showing 50 per cent or more mortality at lowest dilution were short listed. Four isolates,

identified as *Providencia rettgeri*, *Vibrios* sp. and *Pseudomonas putida* reduced egg hatching of *M. javanica* from 20 to 30 per cent (Ashima Kapoor et al., 2007).

2. BIOCHEMICAL CHARACTERIZATION OF PGPR

Various phenotypic and biochemical methods have been developed and used for characterizing fluorescent pseudomonad isolates. The genus *Pseudomonas* is characterized by gram-negative rod shaped aerobic cells and are associated with plants. The important species include *P. fluorescens*, *P. putida*, *P. aeruginosa* and *P. aureofaciens*. Most of the tests conducted for identification of fluorescent pseudomonads have been based on physiological and nutritional tests (Krieg and Holt, 1984). Among the *Pseudomonads* group, *P. aeruginosa* forms a light cluster and grows above 41°C (Hildebrand et al., 1992). Most of the associated *Pseudomonas* sp. belong to *P. fluorescens* and *P. putida* complex.

There was no clear distinction between *P. fluorescens* and *P. putida* (Sheath et al., 1981). However, these two species are identified based on trehalose utilization and gelatin liquefaction. In this, *P. fluorescens* exhibits positive for both the tests whereas *P. putida* show negative response (Hildebrand et al., 1992). The species of fluorescent pseudomonas are again grouped in different biovars and subgroups based on similarity in biochemical tests. Thus, rapid identification of potentially and economically viable bioagents is possible through various methods of biochemical characterization (Zehnder et al., 2000; Singh et al., 2000).

2.1 BIOCONTROL MECHANISMS OF PGPR

PGPR exhibit multiple numbers of mechanisms to promote plant growth and to serve as potential biocontrol agents. Generally, PGPR traits associated with the biocontrol of plant pathogens include:

- (1) Atmospheric nitrogen fixation and its supply to plants
- (2) Synthesizing various phytohormones including auxins and cytokinins
- (3) Providing mechanisms for the solubilization of minerals such as phosphorus
- (4) Antibiotic synthesis (Haas and Defago, 2005),
- (5) Secretion of iron binding siderophores to obtain soluble iron from the soil and provide it to a plant thereby deprive fungal pathogens in the vicinity, of soluble iron (Neilands and Leong, 1986; Dowling et al., 1996).
- (6) Production of low molecular weight metabolites such as hydrogen cyanide with antifungal activity (Dowling and O'Gara, 1994).
- (7) Production of enzymes including chitinase, b-1-3-glucanase, protease and lipase which can lyse some fungal cells (Chet and Inbar, 1994).
- (8) Production of oxidative stress enzymes such as catalases, superoxide dismutases, peroxidase and polyphenol oxidases for scavenging active oxygen species.
- (9) Out-competing phytopathogens for nutrients and occupying niches on the root surface (O'Gara, 1992).
- (10) Lowering the production of stress ethylene in plants with the enzyme ACC deaminase (Penrose et al., 2001).

1.2. PLANT GROWTH PROMOTION

Rhizobacterial strains were found to increase plant growth after inoculation in seeds and therefore called "Plant growth promoting rhizobacteria" (Kloepper et al., 1980). The mechanisms of growth promotion by these PGPR are complex and appear to comprise both changes in the microbial balance in the rhizosphere and alterations in host plant physiology (Glick et al., 1999). Plant growth promoting rhizobacteria, including fluorescent pseudomonads are capable of surviving and colonizing the rhizosphere of all field crops. They promote plant growth by secreting auxins gibberellins and cytokinins (Vidyasekaran, 1998).

PGPR has a significant impact on plant growth and development in both indirect or direct ways. Indirect promotion of plant growth occurs when bacteria or prevent some of the deleterious effects of a phytopathogenic organism by one or more mechanisms. On the other hand, the direct promotion of plant growth by PGPR generally entails providing the plant with compound that is synthesized by the bacterium or facilitating the uptake of nutrients from the environment (Glick et al., 1999). Plant growth benefits due to the addition of PGPR include increase in germination rates, root growth, yield including grain, leaf area, chlorophyll content, magnesium, nitrogen and protein content, hydraulic activity, tolerance to drought and salt stress, shoot and root weights and delayed leaf senescence (Lucy et al., 2004). Seed treatment with PGPR resulted in increased yield and growth in potato under field conditions (Kloepper et al., 1980). (Van Peer and Schippers, 1988) documented the increased root and shoot fresh weight of tomato, cucumber, lettuce and potato as a result of bacterization with *Pseudomonas* strains.

(Mashooda Begum et al., 2003) studied the effectiveness of plant growth promoting rhizobacterial isolates against some seed borne fungal diseases. Among them *B. pumilus* (SE-34), *B. pasteurii* (T4), *B. subtilis* (IN 937-6) and *B. subtilis* (GB-03) strains stood first in the improvement of crop, both in greenhouse and field condition. Potential strains increased the biomass of plants, total number of leaves, fruits, length, girth, biomass of the fruit. The colonization of these bacterial strains reduced the incidence of seed mycoflora which indirectly enhanced the per cent seed germination and vigour index of seedlings. (Minakshi et al., 2005) isolated a total of 113 rhizobacteria from different rhizotic zones of pigeonpea. Seed treatment using four isolates, viz. RS29, RS39, RS41 and RP3 resulted in 90 per cent seed germination in contrast with 50 per cent obtained in untreated control after 72 h of incubation and the isolates RS34, ER17, RP7 and RS41 increased shoot height and shoot dry biomass as compared to uninoculated control whereas isolates RS45, RS36, RS37, ER23, RP24 influenced root dry biomass significantly.

2.4. HYDROGEN CYANIDE (HCN) PRODUCTION

The cyanide ion is exhaled as HCN and metabolized to a lesser degree in to other compounds. HCN first inhibits the electron transport and the energy supply to the cell is disrupted leading to the death of the organisms. It inhibits proper functioning of enzymes and natural receptors reversible mechanism of inhibition (Corbett, 1974) and it also known to inhibit the action of cytochrome oxidase (Gehring et al., 1993). HCN is produced by many rhizobacteria and is postulated to lay a role in biological control of pathogens (Defago et al., 1990). Production of HCN by certain strains of fluorescent pseudomonads has been involved in the suppression of soil borne pathogens (Voisard et al., 1989). Suppression of black root rot of tobacco (Stutz et al., 1986) and take-all of wheat (Defago et al., 1990) by *P. fluorescens* strain CHAO was attributed to the production of HCN. *Pseudomonas fluorescens* HCN inhibited the mycelial growth of *Pythium* in vitro (Weststeijn, 1990).

The cyanide producing strain CHAO stimulated root hair formation, indicating that the strain induced and altered plant physiological activities (Voisard et al., 1989). Four of the six PGPR strains that induced systemic resistance in cucumber against *Colletotrichum orbiculare* produced HCN (Wei et al., 1991). Fluorescent pseudomonas strain RRS1 isolated from Rajanigandha (tuberose) produced HCN and the strain improved seed germination and root length (Saxena et al., 1996). (Pessi and Haas, 2000) reported that low oxygen concentrations are a prerequisite for the activity of the transcription factor ANR which positively regulates HCN biosynthesis.

HCN from *P. fluorescens* strain CHAO not repressed by fusaric acid played a significant role in disease suppression of *F. oxysporum* f.sp. *radicis-lycopersici* in tomato (Duffy et al., 2003). (Ramettee et al., 2003) reported that hydrogen cyanide is a broadspectrum antimicrobial compound involved in biological control of root disease by many plant associated fluorescent pseudomonads. Further, they noted that the enzyme HCN synthase is encoded by three biosynthetic genes (henA, henB and henC).

2.2. INDOLE-3-ACETIC ACID (IAA) PRODUCTION

IAA is phytohormone which is known to be involved in root initiation, cell division and cell enlargement (Salisbury, 1994). This hormone is very commonly produced by PGPR (Barazani and Friedman, 1999). (Vessey, 2003) has reviewed the production of this hormone and implicated it in the growth promotion by PGPR. However, the effect of IAA on plants depends on the plant sensitivity to IAA and the amount of IAA produced from plant associated bacteria and induction of other phytohormones (Peck and Kende, 1995). Patten and Glick (2002) demonstrated that bacterial IAA from *P. putida* played a major role in the development of host plant root system. Similarly, IAA production in *P. fluorescens* HP 72 correlated with suppressing of creeping bent grass brown patch (Suzuki et al., 2003).

2.3. ANTIBIOSIS

Antibiotics are generally considered to be organic compounds of low molecular weight produced by microbes. Antibiosis plays an active role in the biocontrol of plant disease and it often acts in concert with competition and parasitism. Antibiosis has been postulated to play an important role in disease suppression by rhizobacteria (Gutterson et al., 1986).

Fluorescent pseudomonad strains are known to reduce fungal growth in vitro by the production of one or more antifungal antibiotics that may also have activity in vivo (Whipps, 2001). Several strains of *Pseudomonas* spp. have been shown to produce wide array of antibiotics which include 2,4-diacetyl phloroglucinol, hydrogen cyanide, kanosamine, phenazine-1-carboxylic acid, pyoluteorin, oomycin A, pyrrolnitrin, pyocyanin and viscosinamide as well as several other uncharacterized moieties (Tharne et al., 1999). Phloroglucinols phenazines, pyoluteorin,

pyrrolnitrin and cyclic lipopeptides all of which are diffusible and hydrogen cyanide is volatile in nature (Haas and Defago, 2005).

Root associated fluorescent pseudomonads produce and excrete secondary metabolites which are inhibitory to plant pathogenic organisms including fungi, bacteria and nematodes (Haas and Keel, 2003). Among these metabolites the polyketide compound, DAPG has received particular attention because of its broad spectrum antifungal, antibacterial and antihelminthic activity (Keel and Defago, 1997). Phenazines (PHZ) are N containing heterocyclic pigments synthesized by species of *Pseudomonas*, *Streptomyces*, *Burkholderia* and *Brevibacterium* (Stevens et al., 1994). Pyrrolnitrin (PRN) is a broad spectrum antifungal metabolite produced by many fluorescent and non-fluorescent strains of the genus *Pseudomonas* (Howell and Stipanovic, 1979). A phenyl pyrrol derivative of PRN has been developed as an agricultural fungicide. Pyrrolnitrin persists actively in the soil for at least 30 days.

Pyoluteorin (PLT) is an aromatic polyketide antibiotic consisting of a resorcinol ring derived through polyketide biosynthesis. PLT is produced by several *Pseudomonas* sp. including strains that suppress plant diseases caused by phytopathogenic fungi (Murhofer et al., 1994). PLT mainly inhibits the oomycetous fungi including *Pythium ultimum* against which it is strongly active when applied to seeds. PLT-producing *Pseudomonads* decrease the severity of *Pythium* damping off (Nowak-Thompson et al., 1999). *Pseudomonas fluorescens* strain CHAO and its antibiotic over-producing derivative CHAO/PME 3424, repeatedly reduced *M. incognita* galling in tomato, brinjal, mung and soybean in early growth stage. A strong negative correlation existed between rhizobacteria colonization and nematode invasion (Siddiqui and Shaukat, 2003).

2.5. INDUCED SYSTEMIC RESISTANCE (ISR)

Induced resistance is defined as an enhancement of the plants defensive capacity against a broad spectrum of pathogens and pests that is acquired after appropriate stimulation. The resulting elevated resistance due to an inducing agent upon infection by pathogen is called induced systemic resistance (ISR) or systemic acquired resistance (SAR) (Hammerschmidt and Kuc, 1995). The induction of systemic resistance by rhizobacteria is referred to as ISR, whereas that by other agencies is called SAR (Van Loon et al., 1998). Once resistance is induced, it will afford non-specific protection against pathogenic fungi, bacteria, nematodes and viruses as well as against insect pests.

A large number of defense enzymes that have been associated with ISR include phenylalanine ammonia lyase (PAL), chitinase, β -1,3-glucanase, peroxidase (PO), polyphenol oxidase (PPO), superoxide dismutase (SOD), catalase (CAT), lipoxygenase (LOX), ascorate peroxidase (APX) and proteinase inhibitors (Van Loon, 1997). These enzymes also bring about liberation of molecules that elicit the initial steps in induction of resistance, phytoalexins and phenolic compounds (Van Loon et al., 1998).

Induced systemic resistance by PGPR has been achieved in large number of crops including *Arabidopsis* (Pieterse et al., 1996), cucumber (Wei et al., 1996), tomato (Duijff et al., 1997), potato (Doke et al., 1987), radish (Leeman et al., 1996), carnation (Van Peer et al., 1991), sugarcane (Viswanathan and Samiyappan, 1999), chilli brinjal (Bharathi et al., 2004), rice (Nandakumar et al., 2001) and mango (Vivekananthan et al., 2004) against broad spectrum of pathogens including fungi (Leeman et al., 1995), bacteria (Liu et al., 1995), nematodes (Paul and Kumar, 2003) and viruses (Kandan et al., 2005).

Seed treatment and seedling root dipping induced early and enhanced levels of PO in rice plants (Nayar, 1996). Two peroxidase isoforms were induced in the PGPR-treated rice plants inoculated with the sheath blight pathogen, *R. solani* (Nandakumar et al., 2001). High level expression of PO was reported in *P. fluorescens* Pf1 treated chilli plants challenged with *C. capsici* (Bharathi et al., 2004). Similarly, increased activity of PPO was observed in PGPR treated tomato plants challenged with *F. oxysporum* f.sp. *lycopersici* (Ramamoorthy et al., 2002).

Plants treated with *Pseudomonas* strains initially showed higher level of PAL compared to control (Chen et al., 2000). (Radjacommare et al., 2004) reported that seedling dip with talc based formulation of *P. fluorescens* induced the activity of PAL in finger millet leaves against blast disease. The inoculation of PGPR strains *P. putida* 89B-27 and *Serratia marcescens* 90-166 and the pathogen, *F. oxysporum* f.sp. *cucumerinum* on separate halves of roots of cucumber seedlings exhibited that both PGPR strains induced systemic resistance against the *Fusarium* wilt as expressed by delayed disease symptom development and reduced number of dead plants (Liu et al., 1995). The same PGPR strains also induced systemic resistance in cucumber against bacterial angular leaf spot caused by *Pseudomonas syringae* pv. *lachrymans* (Liu et al., 1995).

Maize plants raised from *P. fluorescens* treated seeds showed higher activity of peroxidase, polyphenol oxidase and PAL, when leaf sheaths were inoculated with the pathogen, *R. solani*. The bacterized seeds with *P. fluorescens* lead to accumulation of higher phenolic compounds and higher activity of PO, PPO and PAL that may

play a role in defense mechanism in plants against pathogen (Sivakumar and Sharma, 2003). (Kloepper et al., 2004) also observed, control of nematode diseases in tomato and bell pepper by treatments with PGPR strains through induction of systemic resistance. (Siddiqui and Shaukat, 2002) observed that the application of PGPR strains to one half of the split root system of tomato caused a significant reduction (42%) in nematode penetration in the other half of the split root system and this was attributed to ISR activity of the strain.

(Hariprasad and Umesh, 2007) reported that PGPR application were made by seed, root and foliar spray treatments separately in combinations in field. Among the PGPR strains *Bacillus subtilis* strain GB3 was the most effective in providing significant suppression of bacterial spot and was well correlated with increased activity of defense related enzymes, viz. peroxidase and PAL. PGPR that were effective in greenhouse were also able to induce resistance in tomato against bacterial spot under field conditions.

3.1. BIOFORMULATIONS OF PGPR STRAINS

An important area of biological control is the development of formulations that would case for viable microbial activity for long period of time. Mass multiplication of PGPR in a suitable medium and development of a powder formulation was first carried out in 1980. A dried powder formulation of PGPR, especially is important for seed treatment and soil application. The survival of PGPR in a dried formulation and the effectiveness of methyl cellulose in a powder formulation for coating sugarbeet seed has been well documented (Suslow, 1980). A talc-based formulation of PGPR has been developed for inoculation of potato seed pieces (Kloepper and Schroth, 1981). Talc based formulation of *P. fluorescens* isolated from the rhizosphere of different crops has been developed (Vidhyasekaran and Muthamilan, 1995)

Root dip with *P. fluorescens* formulated in talc was found to be effective in reducing *M. incognita* and caused 40 per cent reduction in root galls under glasshouse conditions in grape vine (Mani, 1996). Glasshouse and microplot experiment with *P. fluorescens* strain Pf1 at 1, 2 or 4 g/plant in grape vine reduced the severity of infection by *M. incognita* (race 3) and enhanced root colonization by the rhizobacterium. Colonization was observed in the roots produced during second season (Shanthi et al., 1998). Biopriming of plants with some PGPR can also provide systemic resistance against a broad spectrum of plant pathogens. Diseases of fungal bacterial and viral origin and in some instances, even damage caused by insects and nematodes can be reduced after application of PGPR (Compant et al., 2005).

3. RHIZOBACTERIA IN THE MANAGEMENT OF PLANT DISEASES

PGPR are having the ability to protect above ground plant parts against fungal, bacterial and viral diseases by induced systemic resistance (ISR). (Kloepper et al., 1992) reported that among the PGPR, fluorescent pseudomonads are the most exploited bacteria for biological control of soil borne and foliar plant pathogens. In the past three decades numerous strains of fluorescent pseudomonads have been isolated from the soil and plant roots by several workers and their biocontrol activity against soil borne and foliar pathogens have been reported (Ramamoorthy et al., 2002).

Pseudomonas fluorescens isolates are effective bacterial antagonists for the management of soil borne and foliar diseases. Among the various isolates tested, *P. fluorescens* isolate Pf1 effectively inhibited mycelial growth of the pathogen in vitro conditions and decreased the fruit rot incidence under greenhouse conditions (Ramamoorthy and Samiyappan, 2001).

The application of biocontrol PGPR strains has given promising results in cereals, vegetables, fruit and ornamental plant production under glass house and field conditions (Raupach and Kloepper, 1998). In greenhouse and field experiments, PGPR strain *B. pumilus* INR-7 effectively protected pearl millet against downy mildew (Niranjan Raj et al., 2003).

PGPR mediated resistance in mango trees infected with *Colletotrichum gloeosporioides* significantly reduced the anthracnose infection besides enhancing fruit yield under field conditions (Vivekananthan et al., 2004). These studies clearly indicate the PGPR have diverse mechanism to operate to combat the pests and pathogens and work efficiently in both greenhouse and field conditions.

Since 1987 in China, PGPR, called yield increasing bacteria (YIB) have been largely applied in 48 different crops over 3.35 millions of hectares. In that country, productivity gains as high as 23.1% and 22.5% were obtained, respectively, in sweet potatoes and potatoes. Additionally, remarkable 85.5% and 80.3% reduction levels of diseases caused by *Xanthomonas oryzae* pv. *oryzae* and *Glomerella cingulata*, respectively, were recorded (Zhang et al., 1996).

Black rot caused by *Xanthomonas campestris* pv. *campestris* (Xcc) causes severe economic losses in all developmental crucifer stages (Mariano et al., 2001). *Bacillus* spp. isolated from healthy cabbage, kale, and radish

had reduced black rot incidence in kale and cabbage in greenhouse and field experiments (Assis et al., 1996). (Monteiro et al., 2005) showed that four of these *Bacillus* strains produced lipopeptides active against Xcc during its late growth phase. These peptide antibiotics are amphiphilic compounds with surfactant activity (Zuber et al. 1993). Recently, it was demonstrated that lipopeptides can stimulate ISR in plants, probably by interacting with plant cell membranes and inducing temporary alterations in the plasma membrane which could raise plant defenses (Ongena et al., 2009).

Fusarium wilt of banana caused by *Fusarium oxysporum* f. sp. *cubense* is a very destructive disease in Brazil and other parts of the world. The rhizomes and pseudostems of infected plants used for propagation are the principal sources of inoculums and disease dispersion. Therefore, micropropagated health plantlets are used to prevent or delay the introduction of this pathogen in soils. However, these plantlets are more susceptible to this and other soilborne pathogens and should be protected before transplanting. PGPR are an alternative for improving this system. In greenhouse studies, endophytic and epiphytic bacteria applied, isolated or in mixtures, as root and substrate treatments, significantly increased the growth of micropropagated banana plantlets and controlled fusarium wilt (Mariano et al., 2004). According to (Nowak and Shulaev, 2003), the production of high-quality propagules with disease resistance may be achieved among others methods by their "in vitro" and "ex vitro" biopriming (priming with beneficial microorganisms).

Commonly, control is based on the use of single biocontrol agents. This strategy must be changed because, from the ecological point of view, the disease is part of a complex agroecosystem. As reported by (Fravel, 2007), a holistic view of this system can help take correct decisions about management. Therefore, a special approach for improving the PGPR efficiency is the use of mixtures containing different genera or species that presents additive or synergistic effects such as nitrogen-fixing bacteria and mycorrhiza helper bacteria (MHB). Another strategy is to use PGPR, mixed or alternated with fungicides, integrating biological and chemical control.

MHB are those which either assist mycorrhiza formation or promote the functioning of their symbiosis. They exist in arbuscular and ectomycorrhizal systems. MHB present three significant functions: nutrient mobilization from soil minerals, fixation of atmospheric nitrogen, and plant protection against root pathogens (Frey-Klett et al., 2007). According to these authors, PGPR induced increases in mycorrhizal root colonization from 1.1 to 17.5 fold in different interactions. Some of the MHB cited were *Pseudomonas fluorescens*, *P. monteilli*, *Bacillus coagulans*, *B. subtilis*, *Paenibacillus brasiliensis*, *Rhizobium leguminosarum*, and *Bradyrhizobium japonicum*.

Wheat seeds treated with different mixtures of *Paenibacillus macerans* and difenoconazole showed significant reduced incidences of pathogens (Luz, 2003a), and in field all treatments promoted germination and grain yield except for difenoconazole alone that increased only yield. Similar results were obtained when corn seeds were bacterized with the same bioprotector β fludioxonil β metalaxyl M (Luz, 2003b). Also *Bacillus*-based treatments have been successfully combined with traditional chemical seed treatments (Bugg et al., 2009). Therefore, the use of such mixtures may lead to a substantial reduction of pesticide use in several crops. It is also important to focus on the critical stages of commercialization of biocontrol agents. Screening for new agents should consider the biology and ecology of the pathosystem, as well as agricultural practices associated with the crop (Fravel, 2007).

This knowledge will help prevent variation in field performance which is responsible for lack of wider adoption of biocontrol for disease management. The formulation stage aim is to deliver the biocontrol agent in a physiologically active state to provide the needed control. The formulation must be economical and present good shelf-life and a suitable form for shipping, storage, and application. Risk assessment to human health and to the environment are needed before releasing the new product, and early in the screening; even microorganisms with good biocontrol potential but capable of growing at human body temperature should be eliminated (Fravel, 2007). However, rhizobacteria can reduce the activity of pathogenic microorganisms not only through microbial antagonism, but also by activating the plant to better defend itself. This phenomenon, termed 'induced systemic resistance' (ISR) was first described by (Van Peer et al., 1991) in carnation that was systemically protected against *Fusarium oxysporum* f.sp. *dianthi* upon treatment with strain WCS417, and by (Wei et al., 1991) in cucumber, where six out of 94 rhizobacterial strains protected the leaves against anthracnose caused by *Colletotrichum orbiculare*. Protection as a result of microbial antagonism was excluded because the inducing rhizobacteria and the challenging pathogens were inoculated at, and remained confined to, spatially separated parts on the same plants. Hence, the protective effect was plant-mediated. ISR confers on the plant an enhanced defensive capacity (Van Loon and Bakker, 2005).

Upon infection with a challenging pathogen this enhanced defensive capacity is manifested as a reduction in the rate of disease development, resulting in fewer diseased plants or in lesser disease severity. The induced resistance is also evident locally and sometimes does not extend systemically (Van Loon, 2000). When only local, it is difficult to prove, because the inducing bacterium and the challenging pathogen are not separated from each

other and direct antagonism is difficult to rule out. Only when specific eliciting components of the inducer are active in stimulating resistance in the plant but inactive in antagonizing the pathogen in vitro on different types of media, can locally induced resistance be inferred. Induction of resistance by live organisms always requires proof that the organisms cannot contact each other, a condition that can be met when an inducing rhizobacterium remains confined to the roots and the challenging pathogen colonizes only the leaves. Under such situations the inducing bacterium must trigger the roots to locally produce a signal that moves to the leaves to activate the enhanced defensive capacity systemically. The nature of this mobile signal has so far remained elusive.

Since its discovery, rhizobacteria-mediated ISR has been documented in at least 15 plant species (Van Loon and Bakker, 2006). Its induction has been shown to share several characteristics, but its expression can involve different physiological mechanisms. ISR can be induced by various non-pathogenic microorganisms and by some types of stress that activate the same response in the plant. In contrast to R-gene-mediated resistance, it is not specific but active against all types of pathogens, as well as against several nematodes and insects. Once induced, plants may remain protected for a considerable part of their lifetime, indicating that when the state of ISR has been triggered in the plant, it is rather stable (Van Loon et al., 1998).

Upon challenge inoculation, ISR is expressed as a result of the altered physiological state of the plant. Expression may take different forms, depending on the activity of the inducing rhizobacterium and the nature of the interaction between the pathogen and the plant (Chester, 1933). In fact, 'induced resistance' is an operational term to denote a condition in which a plant becomes less diseased compared to a control plant that was not induced. There are many ways in which developmental and environmental factors can influence plant-pathogen interactions. Damping-off due to infection by *Pythium*, *Fusarium* or *Rhizoctonia* is often confined to the seedling stage. Any condition that results in more rapid plant growth will shorten the vulnerable stage and be apparent as enhanced resistance. Rhizobacteria acting through growth promotion could protect plants through this mechanism. A similar type of ISR could occur in potato where accelerated development leads to enhanced adult plant resistance against late blight caused by *Phytophthora infestans* (Visker et al., 2003).

Some reports on ISR have indicated reduced symptom expression in the absence of a reduction in pathogen proliferation. This tolerance of the plant to the pathogen must have a physiological basis. Examples are the reduced damage of *Pythium ultimum*-infected cucumbers and lesser extent of soft rot of potato infected by *Erwinia carotovora* pv. *carotovora* upon prior treatment of the plants with ACC deaminase-containing rhizobacterial strains. By lowering the level of stress ethylene in the plant due to pathogenic attack, ACC deaminase acted synergistically with other mechanisms of biocontrol in reducing symptom development without having an effect on the population density of the pathogen (Wang et al., 2000).

Reduced disease can also be the outcome of alterations in the microbial populations in the rhizosphere as a result of altered host physiology. Numbers of resistance-inducing bacteria may be changed, or their expression of resistance-inducing traits may be altered (Mark et al., 2005). Plants commonly react to root colonization by rhizobacteria by increasing the release of exudates, and quantity and composition of root exudates vary with plant developmental stage (Phillips et al., 2004). Thus, plant growth promotion could alter root exudation. Moreover, rhizobacteria that act as minor pathogens or are perceived by the plant as a potential threat, are likely to change the rate and composition of exudates, and to increase the release of lysates. The population densities and the diversity of the root microflora may affect the number and activity of resistance-inducing rhizobacteria. Quorum sensing (QS) within and between bacterial populations is a major regulatory mechanism in bacteria to adjust their metabolism to crowded conditions or other changes in the biotic and abiotic environment (Whitehead et al., 2001). Interference with bacterial QS by host plants has been documented. Plants can produce and secrete various compounds that mimic QS signals of bacteria and, thereby, alter bacterial activities in the rhizosphere (Bauer and Mathesius, 2004). The ecological diversity and its consequences for metabolic activity of rhizosphere bacteria are only poorly known at present and deserve further investigation. Rhizobacteria can also alter plant secondary metabolism, resulting in changed plant-insect relationships. Root colonization of cucumber by four different PGPR reduced the level of cucurbitacin, which acts as a feeding stimulant to cucumber beetles (Zehnder et al., 1997).

Similar effects on insects that can transmit viruses, might reduce virus diseases through induced resistance against the insect vector rather than against the virus itself. Finally, non-pathogenic rhizobacteria may activate inducible defence mechanisms in the plant in a similar way to pathogenic microorganisms. Such mechanisms can include reinforcement of plant cell walls, production of anti-microbial phytoalexins, synthesis of pathogenesis-related proteins (PRs) (Hammond-Kosack and Jones, 1996), as well as an enhanced capacity to express these defence responses upon challenge inoculation with a pathogen, a mechanism known as 'priming' (Conrath et al., 2006).

Activation of defence reactions suggests that even a beneficial rhizobacterium may be perceived by the plant as a potential threat, and that such perception involves production of resistance-eliciting compounds that act mechanistically similar to elicitors produced by plant pathogenic fungi and bacteria. Both nitrogen-fixing Rhizobia in legume root nodules and vesicular-arbuscular (VA) mycorrhizal fungi in roots have been shown to activate plant host defences when the symbiotic interaction becomes unproductive (Hause and Fester, 2005). Plants possess sensitive mechanisms to perceive both fungi and bacteria through conserved components that are specific to their kingdoms and act as general elicitors. These are commonly referred to as 'pathogen-associated molecular patterns' (PAMPs) (Nürnberger and Lipke, 2005).

During compatible plant-pathogen interactions and effective symbioses, the microorganisms actively suppress defensive activities in the host (Da Cunha et al., 2006). The relationship between root-colonizing, resistance-inducing PGPR and their hosts seems substantially less intimate than with either Rhizobia or mycorrhizal fungi, but the idea that PGPR may at the same time trigger and suppress defence reactions in the host, deserves consideration.

3.2. Mechanisms involved in the AM-mediated biocontrol

Reduction in the deleterious effects of soilborne pathogens after root colonization with AM fungi was described a long time ago (Gerdemann, 1974) and has been observed on various fungi, stramenopiles, nematodes and bacteria (Whipps, 2004). *Glomus mosseae* in symbiosis with clover plants cv. Sonja was even able to totally prevent infection by *Pythium ultimum* (Carlsen et al., 2008). The characteristics of this biological control regarding to its amplitude related to the pathogen/AM fungus/plant taxa association, conditions of culture, level of root colonization, time of AM/pathogen inoculation and harvest, etc. and the mechanisms hypothesized to be involved were described in various reviews (Akhtar and Siddiqui, 2009). The disease symptoms induced were even shown to be systemically reduced in non-mycorrhizal roots of plants grown in split-root systems inoculated with AM fungi (Khaosaad et al., 2007).

Various hypotheses have been put forward in an attempt to explain the AM-mediated biocontrol of soilborne phytopathogens. The fact that pathogen induced symptoms are systemically regulated by AM colonization suggests the establishment of induced systemic resistance (Pozo and Azcón-Aguilar, 2007). New isoforms of superoxide dismutases and peroxidases (Garmendia et al., 2006), PR-1 proteins (pathogenesis-related proteins type 1; Cordier et al., 1998) and higher concentrations of phenolic acids (Zhu and Zao, 2004) (ISR-related compounds) were detected in plants colonized with AM species with biocontrol activities. Accumulation of jasmonic acid involved in the rhizobacteria-mediated ISR (Pozo et al., 2004) in mycorrhizal roots (Isayenkov et al., 2005) could be related to the systemic pathogen biocontrol. Additionally, (Cordier et al., 1998) identified local cell-wall modifications such as callose accumulation around arbuscule-containing cortical cells of tomato roots.

Furthermore, the synthesis of constitutively and additional isoforms of defense related enzymes such as chitinases, chitosanases, β -1,3-glucanases, peroxydases and superoxide dismutase has been locally detected in mycorrhizal roots (Pozo et al., 1999). Nonetheless, the level of production of these enzymes or of flavonoids was shown to be unrelated to the capacity of biocontrol of the AM species (Carlsen et al., 2008). Moreover, transcript profiling and real-time quantitative PCR used to explore the transcriptional changes triggered by AM colonization revealed a complex pattern of local and systemic changes in gene expression in roots of *Medicago truncatula* (Liu et al., 2007) but, transcripts for defense-related proteins were only locally expressed. Furthermore, concentrations of defense related compounds such as rosmarinic and caffeic acids, phenolics and essential oils were not increased by colonization with *G. mosseae* protecting basil plants against *Fusarium oxysporum* f. sp. *Basilica* highlighting the role of other mechanisms in the AM-mediated biocontrol than the stimulation of systemic and localized plant defense mechanisms (Toussaint et al., 2008).

The most frequently documented response to AM colonization is an increase in phosphorus nutrition of the host plant which would consequently be more vigorous and more resistant to pathogen invasion. Nonetheless, the AM mediated biocontrol was shown to be unrelated to the soil P availability and/or the P status in plant tissues and then more dependent on other mechanisms (Toussaint et al., 2008). AM fungi would compete for space and nutrients with soilborne pathogens within the mycorrhizosphere and the host roots. (Larsen and Bødker, 2001), using signature fatty acids profiles, demonstrated the decrease in biomass and energy reserves of both *G. mosseae* and *Aphanomyces euteiches* co-occupying pea roots.

(Cordier et al., 1996) also showed that *Phytophthora nicotianae* and *G. mosseae* never occupied simultaneously the same tomato root tissues. A reduction in the extent of mycorrhizal colonization by different plant pathogens has been reported (Krishna and Bagyaraj, 1983) indicating the possible occurrence of competitive interactions. Because of this competition, the AM fungus is often inoculated before the pathogen in order to favor

biocontrol efficiency. However, *F. solani* f. sp. *Phaseoli* (Fsp) genomic DNA quantified using quantitative real time PCR was significantly reduced not only in the mycorrhizosphere and the mycosphere but also in the bulk soil of a compartmentalized soil-root system inoculated with *G. intraradices* (Filion et al., 2003).

The AM genomic DNA was not significantly modified by the pathogen in the soil. Reduction in Fsp growth as well as root rot symptoms as a result of colonization with *G. intraradices* would not be the consequence of competition for resources and habitat between the two fungi but mostly caused by the biotic and/or abiotic characteristics of the established mycorrhizosphere. The *G. intraradices* extraradical network has been shown to directly reduce the growth of the nematodes *Radopholus similis* and *Pratylenchus coffeae* and of conidial formation of the fungus *F. o. f.sp. chrysanthemi* (Foc) in root and other microorganism-free in vitro conditions (Elsen et al., 2003). However, these negative impacts were not significant for all nematode developmental stages and were unrelated to the AM fungus mycelial or spore densities (Elsen et al., 2003). Furthermore, the Foc spore germination and hyphal growth were significantly increased in presence of the AM fungus suggesting that the direct inhibition of pathogen development by AM structures would be weakly involved in biocontrol (St- Arnaud et al., 1995).

Studies on the impact of exudates from extraradical AM network or mycorrhizal roots both grown in vitro on pathogen can lead to results in contradiction. Crude extracts from *G. intraradices* extraradical network unambiguously reduced Foc conidia germination (Filion et al., 1999). Analogous inhibitive effects were observed with exudates liberated by strawberry roots colonized by *G. etunicatum* and *G. monosporum* on the pathogen *P. fragariae* sporulation (Norman and Hooker, 2000). Meanwhile, depending on the harvest time, exudates from in vitro grown tomato roots colonized with *G. intraradices* were repulsive or more attractive than exudates from non-AM inoculated roots to *P. nicotianae* zoospores (Lioussanne et al., 2008). Moreover, microconidia germination of *F. o. f. sp. lycopersici* (Fol) was more than doubled in the presence of root exudates from tomato plants grown in soil and colonized with *G. mosseae* compared with exudates from non-mycorrhizal plants (Scheffknecht et al., 2006).

The only study of the direct impact of exudates from mycorrhizal plants in the AM mediated biocontrol directly measured in soil conditions by quantification of the capacity of root infection by the pathogen was performed by (Lioussanne et al., 2009d). Application of root exudates from tomato plants colonized with *G. intraradices* or *G. mosseae* on tomato roots had no impact on *P. nicotianae* intraradical growth while direct inoculation of these AM fungi significantly reduced this data suggesting that exudates from mycorrhizal plants would not directly or indirectly (through stimulation of other beneficial microorganisms) inhibit the capacity of pathogen intraradical proliferation. Furthermore, no compound antagonistic to pathogen development directly exuded by AM fungi has yet been identified.

3.3. The mycorrhizosphere: a zone unfavorable to pathogen development

The mycorrhizosphere has been hypothesized to constitute an environment conducive to microorganisms antagonistic to soilborne pathogen proliferation. Indeed, co-culture of the non-mycorrhizal species *Dianthus caryophyllus* with the mycorrhizal species *Tagetes patula* in presence of *G. intraradices* clearly reduced the disease caused by *F. o. dianthi* in *D. caryophyllus* in a manner clearly unrelated to plant nutrition which suggests a reduction in the pathogen development within the mycorrhizosphere (St-Arnaud et al., 1997). Moreover, a reduction in the number of infection loci of tomato roots pre-colonized with *G. mosseae* and inoculated with *P. nicotianae* zoospores infers that the pathogen may be affected prior to root penetration in the mycorrhizosphere (Vigo et al., 2000).

The mycorrhizosphere influenced by the rhizobacteria- AM-root tripartite association presents specific characteristics, in which each actor influences the others growth and health. Notably through the liberation of glycoproteins such as glomalin, AM fungi favor the formation of aggregates which provide stable microsites favorable to root and microbe establishment (Rillig and Mummey, 2006). The AM extraradical network also constitutes specific microsites which favor the growth of some bacteria. Among Plant growth promoting rhizobacteria (Bowen and Rovira, 1999), P-solubilizing and N-fixing-bacteria have been shown to synergistically interact with AM fungi, increasing P and N availability to the plant and promoting its growth and probably favoring its capacity to counteract pathogen impact on plant growth (Lioussanne et al., 2009b).

PGPR can also display biocontrol properties and impact pathogen proliferation through direct liberation of toxic compounds, competition for space and nutrients, reduction of Fe and Mn availability, modification of the plant hormone balance and stimulation of plant defense mechanisms (Bowen and Rovira, 1999). A synergistic or additive control of pathogen impact on plant growth by dual inoculation of AM fungi with rhizobacteria showing biocontrol properties would depend on the bacterial/fungal species combination used, the soil nutritional status and probably other environmental conditions (Barea et al., 2005).

Maximum reduction in galling and nematode multiplication causing root-rot in chick pea was observed with combined inoculation of *G. intraradices* with the biocontrol agents *Pseudomonas straita* and *Rhizobium* sp. (Akhtar and Siddiqui, 2008) and dual inoculation of *G. mosseae* with *Pseudomonas fluorescens* (Siddiqui and Mahmood, 1998). (Järderlund et al., 2008) showed that interactions between the two PGPR *P. fluorescens* SBW25 and *Paenibacillus brasilensis* PB177 with *G. mosseae* and *G. intraradices* investigated on winter wheat infested with *Microdochium nivale* were fungal and bacterial species specific. Several studies have demonstrated that microbial antagonists to pathogens, either fungi or PGPR, do not exert any negative effect against AM fungi (Barea et al., 2005). Mycorrhization Helper Bacteria (MHB), defined by (Garbaye, 1994) as bacteria which consistently promote mycorrhizal development, would even increase AM impact on pathogens. Rhizobacteria and conditions of stimulation of mycorrhizal symbiosis have been listed by (Frey-Klett and colleagues, 2007).

4. CONCLUSION

Mycorrhiza forming species strongly modify the structure and dimension of rhizospheric microorganisms, either by direct interactions, or indirectly by influencing the release of the root exudates in rhizosphere. The mycorrhizae exercise, generally, a strong selective pressure on rhizospheric habitats, stimulating the development of mutualistic or comensal microbiota. The mycorrhizae influence all the relations established between different categories of organisms in rhizospheric microhabitats under late successional stages, and in young rhizospheric microhabitats the mycorrhizal success depends on the microbial community already established. The plant benefits from all of mutualistic relations established between mycorrhizal species and the soil organisms, while the fungal partner often competes with different soil organisms for the plant carbohydrates. The elucidation of the intimate mechanisms that underline the structure of microbial community and the processes that influence the mycorrhizal intensity and rate are premises in the elaboration of the efficient ecological reconstruction strategies or for the sustainable agriculture development. There are needed some extensive researches concerning signal phase prior to tripartite mutualistic relations development and the involved factors, in order to use and optimize them in the purpose of integrated pest management strategies development.

From these results we concluded that interaction between hormones and AM fungi signals and PGPR mediated the expression of the majority of different PR-proteins leading to increasing defense mechanism against diseases infection. Thus, reduction in disease symptoms and enhancing in growth and metabolic activities in crop plants treated with AM fungi and/or hormonal elicitors and PGPR might be related to its roles in the activation of biochemical and structural defense systems that helps ward off the spread of pathogen and consequently increase crop production. Finally, the new mechanism of the combination strategy between bioagent and hormonal signals (either synergistically or antagonistically) played important role for altering expression of defense genes leading to different PR-proteins and also working together to increased resistance in crop plants against diseases.

REFERENCES

- Akhtar MS, Siddiqui ZA. 2008. Biocontrol of a rootrot disease complex of chickpea by *Glomus intraradices*, *Rhizobium* sp. and *Pseudomonas straita*. *Crop Protect* 27, 410-417.
- AKHTAR MS, Siddiqui MA. 2009. Arbuscular mycorrhizal fungi as potential bioprotectants against plant pathogens. In: *Mycorrhizae: sustainable agriculture and forestry* (Siddiqui Z.A., Akhtar S., Futai K., eds). Ed Springer, The Netherlands. pp. 61-98.
- Al-Karaki, GN and A Al-Raddad. 1997. Effect of arbuscular mycorrhizal fungi and drought stress on growth and nutrient uptake of two wheat genotypes differing in drought resistance. *Mycorrhiza*, 7:87-88.
- Al-Karaki GN and R Hammad. 2001. Mycorrhizal influence on fruit yield and mineral content of tomato grown under salt stress. *J. Plant Nutrition*, 24(8):1311- 1323.
- Ahmadzadeh M, Tehrani AS and Jahromi KT. 2004. Study on production of some antimicrobial metabolites by fluorescent pseudomonads. *Iranian J. Agric. Sci.*, 35(3): 731-739.
- Anith KN, Momol MT, Kloepper JW, Marois JJ, Olson SM and Jones JB. 2004 Efficacy of plant growth promoting rhizobacteria, acibenzolar-S-methyl and soil amendment for integrated management of bacterial wilt on tomato. *Pl. Dis.*, 88: 669-673.
- Ashima Kapoor, Anil Kumar, Walia KK and Walia RK. 2007. Isolation and in vitro screening of rhizospheric/rhizoplane bacteria for toxicity to root-knot nematode, *Meloidogyne javanica*. Paper presented in National Symp. Nematology in 21st Century : Emerging Paradigms. Assam Agric. Univ., Jorhat, Assam.
- Ashraf M, S Hasnain, O Berge and T Mahmood. 2004. Inoculation wheat seedlings with exopolysaccharide-producing bacteria restricts sodium uptake and stimulates plant growth under salt stress. *Biol. Fertil. Soils*, 40:157-162.
- Barazani O and Friedman J. 1999. Is IAA the major root growth factor secreted from plant growth-mediating bacteria ? *J. Chem. Ecol.*, 25: 2397-2406.
- Barea JM, Pozo MJ, Azcon R, Azcon-Aguilar C. 2005. Microbial co-operation in the rhizosphere. *J Exp Bot* 56, 1761-1778.
- Bauer WD, Mathesius U. 2004. Plant responses to bacterial quorum sensing signals. *Current Opinion in Plant Biology*, 7, 429-433.
- Bharathi R, Vivekananthan R, Harish S, Ramanathan A and Samiyappan R. 2004. Rhizobacteria-based bio-formulations for the management of fruit rot infection in chillies. *Crop Protect.*, 23: 835-843.
- Bowen GD, Rovira AD. 1999. The rhizosphere and its management to improve plant growth. *Adv Agronom* 66, 1-102.

- Bugg K, Hairston W, Riggs J .2009. Succeeding in a traditional Ag-chemical company despite the “snake oil”/“foo-foo dust” concepts of biological-based products. In: Weller D, Thomashow L, Loper J, Paulitz T, Mazzola M, Mavrodi D, Landa BB, Thompson J (eds) 8th International PGPR Workshop. Portland, USA, p 17.
- Burr TJ, Schroth MN and Suslow TV. 1978. Increased potato yields by treatment of seed pieces with specific strains of *Pseudomonas fluorescens* and *P. putida*. *Phytopathol.*, 68: 1377-1383.
- Burris RH. 1998. 100 years of discoveries in biological N₂ fixation. In: Nitrogen fixation : Hundred Years After, Ed.s Bothe, H. Brujin, F.J. and Newton, W.E., New York, pp.21-30.
- Caelsen SCK, Understrup A, Fomsgaard IS, Mortensen AG, Ravnskov S.2008. Flavonoids in roots of white clover: interaction of arbuscular mycorrhizal fungi and a pathogenic fungus. *Plant Soil* 302, 33-43.
- Chen C, Belanger RR, Benhamou N and Paulitz TC. 2000 Defense enzymes induced in cucumber roots by treatment with plant growth promoting rhizobacteria (PGPR). *Physiol. Mol. Pl. Pathol.*, 56: 13-23.
- Chen Y, Mei R, Lu S, Liu L and Kloepper JW. 1996.The use of yield increasing bacteria (YIB) as plant growth promoting rhizobacteria in Chinese agriculture. In: Management of Soil Borne Diseases, Eds. Utkhede, R.S. and Gupta, V.K., Kalyani Publishers, New Delhi, pp.165-184.
- Chester KS. 1933. The problem of acquired physiological immunity in plants (continued). *The Quarterly Review of Biology*, 8, 275–324.
- Compant S, Duffy B, Nowak J, Clement C and Barka EA. 2005. Use of plant growthpromoting bacteria for biocontrol of plant disease : Principles, mechanisms of action and future prospects. *Applied and Environmental Microbiology*, 71: 4951- 4959.
- Conrath U, Beckers GJM, Flors V, Garcí'a-Agusti'n P, Jakab G, Mauch F, Newman M.-A, Pieterse CMJ, Poinssot B, Pozo MJ, Pugin A, Schaffrath U, Ton J, Wendehenne D, Zimmerli L, Mauch-Mani B. 2006. Priming: Getting ready for battle. *Molecular Plant-Microbe Interactions*, 19, 1062–1071.
- Cook RJ and Baker KF. 1983. *The Nature and Practice of Biological Control of Plant Pathogens*. American Phytopathol. Soc., St. Paul, Minnesota, p.539.
- Cordier C, Gianinazzi S, Gianinazzi-Pearson V. 1996. Colonisation patterns of root tissues by *Phytophthora nicotianae* var. *parasitica* related to reduced disease in mycorrhizal tomato. *Plant Soil* 185, 223-232.
- Cordier C, Pozo MJ, Barea JM, Gianinazzi S, Gianinazzi -Pearson V. 1998. Cell defense responses associated with localized and systemic resistance to *Phytophthora parasitica* induced in tomato by an arbuscular mycorrhizal fungus. *Mol Plant-Microb Interact* 11, 1017- 1028.
- Da Cunha L, McFall AJ, Mackey D .2006. Innate immunity in plants: A continuum of layered defenses. *Microbes and Infection*, 8, 1372–1381.
- Danneberg G, C Latus, W Zimmer, B Hundeshagen HJ, Schneider-Poetsch and H Bothe. 1992. Influence of vesicular-arbuscular mycorrhiza on phytohormone balances in maize (*Zea mays* L.). *J.Plant Physiol.*, 141:33-39.
- Defago G and Haas D. 1990. Pseudomonads as antagonists of soil borne plant pathogens: modes of action and genetic analysis. *Soil Biochem.*, 6: 249-291.
- Devi LS and Dutta U. 2009. Effect of *Pseudomonas fluorescens* on root knot nematode (*Meloidogyne incognita*) of okra plant. *Indian J. Nematol.*, 32(2): 185-233.
- Doke N, Ramirez AV and Tomiyama K. 1987. Systemic induction of resistance in potato plants against *Phytophthora infans* by local treatment with hyphal wall components of the fungi. *J. Phytopathol.*, 119: 232-239.
- Dowling DN and O'Gara F. 1994. Metabolites of *Pseudomonas* involved in the biocontrol of plant disease. *Trends Biotechnol.*, 12: 133-141.
- Dowling DN, Sexton R, Fenton A, Delany I, Fedi S, McHugh B, Callanan M, Moenne Loccoz Y and O'Gara F. 1996. Iron regulation in plant-associated *Pseudomonas fluorescens* M114 : implications for biological control. In: *Molecular Biology and Pseudomonads*. Eds. Nakazawa, T., Furukawa K, Haas D, Silver S. American Society for Microbiology Press, Washington, DC, pp.502-511.
- Duffy B, Schouten A and Raajimakers J. 2003. Pathogen self-defense : mechanisms to counteract microbial antagonism. *Annu. Rev. Phytopathol.*, 45: 501-538.
- Duijff BJ, Gianinazzi-Pearson V And Lemanceall P. 1997. Involvement of the outer membrane lipopolysaccharides in the endophytic colonization of tomato roots by biocontrol *Pseudomonas fluorescens* strain WCS 41 7r. *New Phytologist*, 135: 325-334.
- Egamberdiyeva D and G Höflich. 2004. Effect of plant growth-promoting bacteria on growth and nutrient uptake of cotton and pea in a semi-arid region of Uzbekistan. *J. Arid Environments*, 56:293-301.
- Elsen A, Declerck S, Waele DD. 2003. Use of root organ cultures to investigate the interaction between *Glomus intraradices* and *Pratylenchus coffeae*. *Appl Environ Microbiol* 69, 4308-4311.
- Filion M, ST-Arnaud M, Fortin JA. 1999. Direct interaction between the arbuscular mycorrhizal fungus *Glomus intraradices* and different rhizosphere microorganisms. *New Phytol* 141, 525-533.
- Filion M, ST-Arnaud M, Jabaji-Hare SH., 2003. Quantification of *Fusarium solani* f. sp. *phaseoli* in mycorrhizal bean plants and surrounding mycorrhizosphere soil using real-time polymerase chain reaction and direct isolations on selective media. *Phytopathology* 93, 229- 235.
- Fravel D .2007. Commercialization of biocontrol agents for use against plant pathogens. In: IX Reuniao Brasileira sobre Controle Biológico de Doenças de Plantas, Campinas, S. Paulo, Brasil, CD-ROM, pp 1–2
- Frey-Klett P, Garbaye J, Tarkka M .2007. The mycorrhiza helper bacteria revisited. *New Phytol* 176:22–36
- Garbaye J.1994. Mycorrhization helper bacteria: a new dimension in mycorrhizal symbiosis. *Act Bot Gall* 141, 517-521.
- Garmendia I, Aguirreolea J, Goicoechea N. 2006. Defence-related enzymes in pepper roots during interactions with arbuscular mycorrhizal fungi and/or *Verticillium dahliae*. *Biocontrol* 51, 293-310.
- Gehring PJ, Mohan RJ and Watamare PG. 1993. Solvents, fumigants and related compounds. In: *Handbook of Pesticide Toxicology*, Vol. 2, Eds., Hayes, W.J. and Laws, E.R., Academic Press, inc., San Diego, California, pp.646-649.
- Glick BR and Y Bashan. 1997. Genetic manipulation of plant growth-promoting bacteria to enhance biocontrol of fungal phytopathogens. *Biotechnol. Adv.* 15:353-378.
- Glick BR, Patten CL, Holguin G and Penrose DM. 1999. *Biochemical and Genetic Mechanisms used by Plant Growth Promoting Bacteria*. Imperial College Press, London, Frankenberger WT, pp.125-140.
- Glick BR, C Liu, S Ghosh and EB Dumbrof. 2003. Early development of canola seedlings in the presence of the plant growth-promoting rhizobacterium *Pseudomonas putida* GR12-2. *Soil Biol. Biochem.* 29 (8), 1233–1239.
- Grichko VP and BR Glick. 2001. Amelioration of flooding stress by ACC deaminase-containing plant growth-promoting bacteria. *Plant Physiol.Biochem.* 39, 11–17.
- Gerdemann JW. 1974. *Vesicula-arbuscular mycorrhiza*. Academic Press, NY.

- Gupta CP, Sharma A, Dubey RC and Maheshwari DK. 1999. *Pseudomonas aeruginosa* (GRG) as a strong antagonist of *Macrophomina phaseolina* and *Fusarium oxysporum*. *Ctobios*, 99: 185-189.
- Gutterston N, Layton TJ and Warren GJ. 1986. Molecular cloning of genetic determinants for inhibition of fungal growth by a fluorescent *Pseudomonad*. *J. Bacteriol.*, 165: 696-703.
- Hammerschmidt R and Kuc J. 1995. *Induced Resistance to Disease in Plants*. Kluwer Academic Publishers, Dordrecht, The Netherlands, p.182.
- Han HS and KD Lee. 2005. Physiological responses of soybean-inoculation of *Bradyrhizobium japonicum* with PGPR in saline soil conditions. *Res. J.Agric. and Biol. Sci.*, 1(3): 216-221.
- Harman GE and T Bjo`rkman. 1998. Potential and existing uses of *Trichoderma* and *Gliocladium* for plant disease control and plant growth enhancement. In: Harman, G.E., Kubicek, C.P. (Eds.), *Trichoderma and Gliocladium*, vol. 2. Taylor & Francis, London, United Kingdom, pp. 229–265.
- Hammond-Kosack KE and Jones JDG.1996. Resistance gene-dependent plant defense responses. *The Plant Cell*, 8, 1773–1791.
- Haas D and Defago G. 2005. Biological control of soil borne pathogens by fluorescent pseudomonads. *Nature Reviews Microbiol.*, AOP, Published online 10 March 2005: 1-13.
- Haas D and Keel C. 2003. Regulation of antibiotic production in root colonizing *Pseudomonas* spp. and relevance for biological control of plant disease. *Annu.Rev. Phytopathol.*, 41: 117-153.
- Hariprasad P and Umesha S. 2007. Induction of systemic resistance in field growth tomato by PGPR against *Xanthomonas vesicatoria* incitant of bacterial spot. *J. Mycol. Pl. Path.*, 37: 460-463.
- Hause B, Fester T. 2005. Molecular and cell biology of arbuscular mycorrhizal symbiosis. *Planta*, 221, 184–196.
- Howell CR and Stipanovic RD. 1979. Control of *Rhizoctonia solani* on cotton seedlings with *Pseudomonas fluorescens* and with an antibiotic produced by the bacterium. *Phytopathol.*, 69: 480-482.
- Jagadeesh KS. 2000. Selection of rhizobacteria antagonistic to *Ralstonia solanacearum* E.F. Smith causing bacterial wilt in tomato and their biocontrol mechanisms. Ph.D. Thesis, Univ. Agric. Sci., Dharwad.
- Jonathan EI, Basker KR, Abdel-Alim FF, Vrain TC and Dickson DW. 2000. Biological control of *Meloidogyne incognita* on tomato and banana with rhizobacteria, Actinomycetes and *Pasteuria penetrans*. *Nematologica*, 30: 231-240.
- Jonathan EI, Sandeep AI, Cannayane and Umamaheswari R. 2006. Bioefficacy of *Pseudomonas fluorescens* on *Meloidogyne incognita* in banana. *Nematologia Mediterranea*, 34: 19-25.
- Jaderlund L, Arthurson V, Granhall U, Jansson JK. 2008. Specific interactions between arbuscular mycorrhizal fungi and plant growth-promoting bacteria: as revealed by different combinations. *FEMS Microbiol Lett* 287, 174-180.
- Hildebrand DC, Schroth MN and Sands DC. 1992. *Pseudomonas*. In: *Laboratory guide for Identification of Plant Pathogenic Bacteria* (Ed. N. W. Schaad) 2nd edition. International Book Distributing Co., Lucknow, pp.60-80.
- Kandan A, Ramaiah M, Vasanthi VJ, Radjacammare R, Nandakumar R, Ramanathan A And Samiyappan R. 2005. Use of *Pseudomonas fluorescens*-based formulation for management of tomato spotted wilt virus (TSMV) and enhanced yield in tomato. *Biocontrol Sci. Tech.*, 15: 553-569.
- Kanika Sharma, Anuj Saxena, Gunmala Dak, Rekha Sharma and Arti Agarwal. 2007. Isolation and assay of antifungal activity of siderophore producing strains of *Pseudomonas aeruginosa*. *J. Mycol. Pl. Pathol.*, 37: 251-253.
- Khan MR and Akram M. 2000. Effect of certain antagonistic fungi and rhizobacteria on wilt disease complex of tomato caused by *Meloidogyne incognita* and *Fusarium oxysporum* f.sp. *lycopersici*. *Nematologia Mediterranea*, 28: 139-141.
- Khan MR, Khan SM and Khan N. 2001. Effects of soil application of certain fungal and bacterial bioagents against *Meloidogyne incognita* infecting chickpea (abstract). *Proceedings of National Congress on Centenary of Nematology in India: Appraisal and Future Plans held at Division of Nematology, Indian Agricultural Research Institute, New Delhi, India, 5-7 December, 2001* : 148.
- Khaosaad T, Garcia-Garrido JM, Steinkellner S, Vierheilig H. 2007. Take-all disease is systemically reduced in roots of mycorrhizal barley plants. *Soil Biol Biochem* 39, 727-734.
- Keel C and Defago G. 1997. Interactions between beneficial soil bacteria and root pathogens: mechanisms and ecological impact. In: *Multitrophic Interactions in Terrestrial System*, Eds. Gange, A.C., Brown, V.K., Oxford Blackwell Science, pp.27-47.
- Kloepper JW and Beauchamp CJ. 1992. A review of issues related to measuring colonization of plant roots by bacteria. *Canadian J. Microbiol.*, 38: 1219-1232.
- Kloepper JW, Leong J, Teintze M and Schroth MN. 1981. Enhanced plant growth by siderophores produced by plant growth-promoting rhizobacteria. *Nature*, 286: 885- 886.
- Kloepper JW, Leong J, Teintze M and Schroth MN. 1980. *Pseudomonas* siderophores: A mechanism explaining disease suppressive soils. *Current Microbiology*, 4: 317- 320.
- Kloepper JW and MN Schroth. 1978. Plant growth-promoting rhizobacteria on radishes. IV. International Conference on Plant Pathogenic Bacteria. Angers France, 2:879-882.
- Krishna KR, Bagyaraj DJ. 1983. Interaction between *Glomus fasciculatum* and *Sclerotium rolfsii* in peanut. *Can J Bot* 61, 2349-2351.
- Larsen J, Bodker L. 2001. Interactions between pea root-inhabiting fungi examined using signature fatty acids. *New Phytol* 149, 487-493.
- Leeman M, Den Ouden FM, Van Pelt JA, Dirx FPM, Steijl H, Bakker PAHM and Schippers B. 1996. Iron availability affects induction of systemic resistance to *Fusarium* wilt of raddish by *Pseudomonas fluorescens*. *Phytopathology*, 86: 149- 155.
- Liu L, Kloepper JW and Tuzun S.1995. Induction of systemic resistance in cucumber against *Fusarium* wilt by plant growth promoting rhizobacteria. *Phytopathol.*, 85:695-698.
- Lioussanne L, Jolicoeur M, St-Arnaud M. 2008. Mycorrhizal colonization with *Glomus intraradices* and development stage of transformed tomato roots significantly modify the chemotactic response of zoospores of the pathogen *Phytophthora nicotianae*. *Soil Biol Biochem* 40, 2217-2224.
- Lioussanne L, Beauregard M.-S, Hamel C, Jolicoeur M, St-Arnaud M. 2009b. Interactions between arbuscular mycorrhiza and soil microorganisms. In: *Advances in mycorrhizal biotechnology: a Canadian perspective* (Khasa D., Piché Y., Coughlan A. eds). NRC Press, Ottawa.
- Liu JY, Maldonado-Mendoza I, Lopez-Meyer M, Cheung F, Town CD, Harrison MJ. 2007. Arbuscular mycorrhizal symbiosis is accompanied by local and systemic alterations in gene expression and an increase in disease resistance in the shoots. *Plant J* 50, 529-544.
- Lucy M, Reed E and Glick BR. 2004. Application of free living plant growth promoting rhizobacteria. *Antonie Van Leeuwenhoek*, 86: 1-25.
- Luz WC .2003a. Avaliac,a`o dos tratamentos biolo`gico e qui`mico na reduc,a`o de pato`genos em semente de trigo. *Fitopatol Bras* 28:093–095

- Luz WC .2003b. Combinaco dos tratamentos biolgico e qumico de semente de milho. *Fitopatol Bras* 28:37–40
- Mani MP. 1996. Effect of *Pasteuria penetrans* and *Pseudomonas fluorescens* against *Meloidogyne incognita* in grape vine. M.Sc.(Agri.) Thesis, Tamil Nadu Agric. Univ., Coimbatore, India, p.46.
- Mariano RLR, Silveira EB, Assis SMP, Gomes AMA, Oliveira IS, Nascimento ARP . 2001. Diagnose e manejo de fitobacterioses de importancia no Nordeste Brasileiro. In: Michereff SJ, Barros R (eds) *Proteco de Plantas na Agricultura Sustentavel*. UFRPE, Recife, Brasil, pp 141–169
- Mariano RLR, Medeiros FHV, Albuquerque VV, Assis SMP, Mello MRF .2004. Growth-promotion and biocontrol of diseases in fruits and ornamentals in the states of Pernambuco and Rio Grande do Norte, Northeastern Brazil. In: Kobayashi K, Gasoni L, Terashima H (eds) *Biological control of soilborne plant diseases*. JICA, Buenos Aires, Argentina, pp 70–80
- Mark GL, Dow JM, Kiely PD, Higgins H, Haynes J, Baysse C, Abbas A, Foley T, Franks A, Morrissey J, O’Gara, F. 2005. Transcriptome profiling of bacterial responses to root exudates identifies genes involved in microbe-plant interactions. *Proceedings of the National Academy of Sciences of the United States of America*, 102, 17454–17459.
- Mashooda Begum, Ravisankar Rai V and Lokesh S. 2003. Effect of plant growth promoting rhizobacteria on seed borne fungal pathogens in okra. *Indian Phytopath.*, 56(2): 156-158.
- Mayak S, T Tirosh and BR Glick. 2004. Plant growth-promoting bacteria confer resistance in tomato plants to salt stress. *Plant Physiology and Biochemistry*, 42:565-572.
- Mayak S, T Tirosh and BR Glick. 2004a. Plant growth-promoting bacteria that confer resistance to water stress in tomatoes and pepper. *Plant Sci*. 166, 525–530.
- Mayak S, T Tirosh and BR Glick. 2004b. Plant growth-promoting bacteria confer resistance in tomato plants to salt stress. *Plant Physiol. Biochem*. 42, 565– 572.
- Minakshi Saxena AK and Matta NK. 2005. Selection of culturable PGPR from diverse pool of bacteria inhabiting pigeonpea rhizosphere. *Indian J. Microbiol.*, 45: 21-26.
- Monteiro L, Mariano RLR, Souto-Maior AM .2005. Antagonism of *Bacillus* spp. against *Xanthomonas campestris* pv. *campestris*. *Braz Arch Biol Technol* 48:23–29
- Murhofer M, Hae C, Meuwly P, Metraux JP and Defago G. 1994. Induction of systemic resistance of tobacco to Tobacco necrosis virus by the root-colonizing *Pseudomonas fluorescens* strain CHAO : Influence of the *gacA* gene and of pyoverdine production. *Phytopathol.*, 84: 139-146.
- Nandakumar R, Babu S, Viswanathan R, Raguchander T. and Samiyappan R. 2001. Induction of systemic resistance in rice against sheath blight disease by plant growth promoting rhizobacteria. *Soil Biol. Biochem.*, 33: 603-612.
- Nayar K. 1996. Development and evaluation of a biopesticide formulation for control of foliar diseases of rice. Ph.D. Thesis, Tamil Nadu, Agric. Univ., Coimbatore, p.223.
- Neilands JB and Leong SA. 1986. Siderophores in relation to plant growth and disease. *Ann. Rev. Pl. Physiol.*, 37: 187-208.
- Niranjana Raj S, Chaluvaraju G, Amruthesh KN, Shetty HS, Reddy MS and Kloepper JW. 2003. Induction growth promotion and resistance against downy mildew of pearl millet (*Pennisetum glaucum*) by rhizobacteria. *Plant Disease*, 87: 380-384.
- Noel TC, C Sheng, CK Yost, RP Pharis and MF Hynes. 1996. *Rhizobium leguminosarum* as a plant growth promoting rhizobacterium: direct growth promotion of canola and lettuce. *Can. J. Microbiol.*, 42: 279-283.
- Norman JR, Hooker JE. 2000. Sporulation of *Phytophthora fragariae* shows greater stimulation by exudates of non-mycorrhizal than by mycorrhizal strawberry roots. *Mycol Res* 104, 1069-1073.
- Nowak J, Shulaev V .2003. Priming for transplant stress resistance in in vitro propagation. *In Vitro Cell Dev Biol Plant* 39:122–130
- Nowak-Thompson B, Chaney N, Wing JS, Gould SJ and Loper JE. 1999. Characterization of the pyoluteorin biosynthetic gene cluster of *Pseudomonas fluorescens* Pf-5. *J. Bacteriol.*, 181: 2166-2174.
- Nu’rmerger T, Lipka V .2005. Non-host resistance in plants: New insights into an old phenomenon. *Molecular Plant Pathology*, 6, 335–345.
- Ongena M, Daay F, Jacques P, Thonart P, Benhamou N, Paulitz TC, Cornelis P, Koedam NM and Belanger RR. 1999. Protection of cucumber against *Pythium* root rot by fluorescent pseudomonads: predominant role of induced resistance over siderophores and antibiosis. *Pl. Pathol.*, 48: 66-76.
- Ongena M, Henry G, Adam A, Jourdan E, Thonart P .2009. Plant defense reactions stimulated following perception of *Bacillus* lipopeptides. In: Weller D, Thomashow L, Loper J, Paulitz T, Mazzola M, Mavrodi D, Landa BB, Thompson J (eds) *8th International PGPR Workshop*. Portland, USA, p 43
- Paul D and Kumar A. 2003. How plant growth promoting rhizobacteria (PGPR) help the plant in promotion and disease suppression. *Spice India*, 16: 34.
- Peck SC and Kende H. 1995. Sequential induction of the ethylene biosynthetic enzymes by indole-3-acetic acid in etiolated peas. *Plant Mol. Biol.*, 28: 293-301.
- Penrose DM, Mofatt BA and Glick BR. 2001. Determination of 1-aminocyclopropane-1- carboxylic acid (ACC) to assess the effects of ACC deaminase-containing bacteria on roots of canola seedlings. *Canadian J. Microbiol.*, 47: 77-80.
- Pessi G and Haas D. 2000. Transcriptional control of the hydrogen cyanide biosynthetic genes *hcn ABC* by the anaerobic regulator ANR and the quorum-sensing regulators LasR and RhlR in *Pseudomonas aeruginosa*. *J. Bacteriol.*, 182: 6940-6949.
- Phillips DA, Fox TC, King MD, Bhuvaneshwari TV, Teuber L R. 2004. Microbial products trigger amino acid exudation from plant roots. *Plant Physiology*, 136, 2887–2894.
- Pieterse CMI, Van Wees SCM, Hoeffland E., Van Pelt JA and Van Loon LC. 1996. Systemic resistance in *Arabidopsis* induced by biocontrol bacteria is independent of salicylic acid accumulation and pathogenesis-related gene expression. *Plant Cell*, 8: 1225-1237.
- Pozo MJ, Aacon-Aguilar C, DUMAS-GAUDOT E, BAREA JM. 1999. Beta-1,3-glucanase activities in tomato roots inoculated with arbuscular mycorrhizal fungi and/or *Phytophthora parasitica* and their possible involvement in bioprotection. *Plant Sci* 141, 149-157.
- Pozo MJ, Aacon-Aguilar C. 2007. Unravelling mycorrhiza-induced resistance. *Curr Opin Plant Biol* 10, 393-398.
- Pozo MJ, Van Loon LC, Pieterse CMJ. 2004. Jasmonates - Signals in plant-microbe interactions. *J Plant Growth Regul* 23, 211-222.
- Radjaccomare R, Kandan A, Nandakumar R and Samiyappan R. 2004. Association of the hydrolytic enzyme chitinase against *Rhizoctonia solani* in rhizobacteria-treated rice plants. *J. Phytopathol.*, 152: 365-370.
- Rajendran G, Ramakrishnan S and Subramanian S. 2001. Biomanagement of nematodes in horticultural crops. *South Indian Horticulture*, 49: 227-230.

- Ramamoorthy V and Samiyappan R. 2001. Induction of defense-related genes in *Pseudomonas fluorescens* treated chilli plants in response to infection by *Colletotrichum capsici*. *J. Mycol. Plant Pathol.*, 31: 146-155.
- Ramamoorthy V, Raguchander T and Samiyappan R. 2002. Enhancing resistance of tomato and hot pepper to *Pythium* diseases by seed treatment with fluorescent pseudomonads. *European J. Pl. Pathol.*, 108: 429-441.
- Ramesh Kumar N, Thirumalai Arasu V and Gunasekaran P. 2002. Genotyping of antifungal compounds producing plant growth promoting rhizobacteria, *Pseudomonas fluorescens*. *Current Sci.*, 82: 1463-1466.
- Ramatte A, Frapolli M., Defago G and Moenne-Loccoz Y. 2003. Phylogeny of HCN synthase-encoding *hcnbc* genes in biocontrol fluorescent pseudomonads and its relationship with host plant species and HCN synthesis ability. *Molecular Biol.. Pl. Microbe Interaction*, 16: 525-535.
- Raupach GS and Kloepper JW. 1998. Mixtures of plant growth promoting rhizobacteria enhance biological control of multiple cucumber pathogens. *Phytopathol.*, 88: 1158-1164.
- Rillig MC, Mummey DL. 2006. Mycorrhizas and soil structure. *New Phytol* 171, 41-53.
- Ruiz-Lozano JM and R Azcon. 1995. Hyphal contribution to water uptake in mycorrhizal plants as affected by the fungal species and water status. *Physiologia Plantarum*, 95: 472-478.
- Saleh SA, H Heuberger and WH Schnitzler. 2005. Alleviation of salinity effect on artichoke productivity by *Bacillus subtilis* FZB24, supplemental Ca and micronutrients. *J. App. Bot. Food Qual.*, 79: 24- 32.
- Saxena A, Sharma A, Goel R and Johri BN. 1996. Functional characterization of a growth promoting fluorescent pseudomonads from Rajnigandha rhizosphere, 37th Annual Conference of the Association of Microbiologists of India, December 4-6, IIT, Chennai, p.135.
- Scheffknecht S, Mammerler R, Steinkellner S, Vierheilg H. 2006. Root exudates of mycorrhizal tomato plants exhibit a different effect on microconidia germination of *Fusarium oxysporum* f. sp. *Lycopersici* than root exudates from non-mycorrhizal tomato plants. *Mycorrhiza* 16, 365-370.
- Sedra MH and Malouhy MA. 1994. Isolation of microorganisms antagonistic to *Fusarium oxysporum* f.sp. *albedinis* from suppressive soils in palm grooves in marrqkech. *Al Awamia*, 86: 3-19.
- Seenivasan N and Lakshmanan PL. 2001. Effect of culture filtrates of *Pseudomonas fluorescens* on rice root nematode, *Hirschmanniella gracilis*. *Pestol.*, 25: 11-12.
- Shanthi A, Rajeswari S and Sivakumar. 1998. Soil application of *Pseudomonas fluorescens* Migula for the control of root-knot nematode, *Meloidogyne incognita* on grape vine, *Vitis vinifera* Linn. In: Proceedings of the Third International Symposium of Afro-Asian Society of Nematologists (TISAASN) on Nematology : Challenges and Opportunities in 21st Century, held at Sugarcane Breeding Institute (ICAR), Coimbatore, India, 16-19, Arpil, pp.203-206.
- Sheath PHA, Stevens M and Sackin MJ. 1981. Numerical taxonomy of *Pseudomonas* based on published records of substrate utilization. *Antonie Van Leeuwenhoek*, 47: 423-448.
- Siddiqui ZA, Mahmood I. 1998. Effect of a plant growth promoting bacterium, an AM fungus and soil types on the morphometrics and reproduction of *Meloidogyne javanica* on tomato. *Appl Soil Ecol* 8, 77-84.
- Siddiqui IA and Shaukat SS. 2002. Rhizobacteria-mediated induction of systemic resistance in tomato against *Meloidogyne javanica*. *J. Phytopath.*, 150: 469-472.
- Siddiqui IA and Shaukat SS. 2003. Role of iron in rhizobacteria-mediated suppression of root-infecting fungi and root-knot nematode in tomato, *Nematologia Mediterranea*, 31: 11-14.
- Sikora RA. 1990. Bedeutung Von Rhizosphere-Microorganismen fur die biologische Bekämpfung Von Nematoden, die pflanzengesundheit und das management des Antagonistischen potentials des bodens. *Phytomed. Mitt.*, 20: 15.
- Singh KK, Pelvi SK and Singh H. 1980. Medicinal properties of *Coleus forskohlii*. *Bulletin of Medico-fthano Botanical Res.*, 1: 4.
- Sirohi, A., Chawla, G. and Dhawan, S.C., 2000, *Bacillus* and *Pseudomonas* culture filtrates offer promise of nematode management (Abstract). Paper presented at National Symposium on Integrated Nematode Management for Sustainable Agriculture in the changing Agro-ecological and Economic scenario in the New Millennium held at Orissa Univ. Agric. Tech., Bhubaneswar, India, 23-24 Nov. 2000, p.72.
- Sivakumar G and Sharma RC. 2003. Induced biochemical changes due to seed bacterization by *Pseudomonas fluorescens* in maize plants. *Indian Phytopath.*, 56: 134-137.
- Smith SE and DJ Read. 1997. *Mycorrhizal symbiosis*. London: Academic Press.
- St-Arandu M, Hamel C, Vimard B, Caron M, Fortin JA. 1995. Altered growth of *Fusarium oxysporum* f. sp. *chrysanthemii* in an in vitro dual culture system with the vesicular arbuscular mycorrhizal fungus *Glomus intraradices* growing on *Daucus carota* transformed roots. *Mycorrhiza* 5, 431-438.
- St-Arnaud M, Hamel C, Vimard B, Caron M, Fortin JA. 1997. Inhibition of *Fusarium oxysporum* f.sp. *dianthi* in the non-VAM species *Dianthus caryophyllus* by co-culture with *Tagetes patula* companion plants colonized by *Glomus intraradices*. *Can J Bot* 75, 998-1005.
- Stevens AM, Dolan KM and Greenberg EP. 1994. Synergistic binding of the *Vibrio fischeri* LuxR transcriptional activator domain and RNA polymerase to the lux promoter region. *Proc. Natl. Acad. Sci.*, 91: 12619-12623.
- Stutz EW, Defago G and Kern H. 1986. Naturally occurring fluorescent pseudomonads involved in suppression of black root rot of tobacco. *Phytopathol.*, 76: 181-185.
- Suslow TV. 1980. Growth and yield enhancement of sugarbeet by pelleting with specific *Pseudomonas* spp. *Phytopathol. News*, 12: 40.
- Suzuki S, He Y and Oyaizu H. 2003. Indole-3-acetic acid production in *Pseudomonas fluorescens* HP72 and its association with suppression of creeping bentgrass brown patch. *Current Microbiol.*, 47: 138-143.
- Thrane C, Olsson S, Neilsen TH and Sorensen J. 1999. Vital fluorescent stains for detection of stress in *Pythium ultimum* and *Rhizoctonia solani* challenged with viscosinamide from *Pseudomonas fluorescens* DR54. *FEMS Microbiology Ecol.*, 30: 11-23.
- Tiwari PK and Thirumrthy VS. 2007. Isolation and characterization of the *Pseudomonas fluorescens* from rhizosphere of different crops. *J. Mycol. Pl. Pathol.*, 37: 231-234.
- Toussaint JP, Kraml M, Nell M, Smith SE, Smith FA, Steinkellner S, Schmiderer C, Vierheilg H, Novak J. 2008. Effect of *Glomus mosseae* on concentrations of rosmarinic and caffeic acids and essential oil compounds in basil inoculated with *Fusarium oxysporum* f.sp. *basilici*. *Plant Pathol* 57, 1109-1116.
- Tripathi M and Johri BN. 2002. In vitro antagonistic potential of fluorescent pseudomonads and control of sheath blight of maize caused by *Rhizoctonia solani*. *Indian J. Microbiol.*, 42: 207-214.
- Van Loon LC. 1997. Induced resistance in plants and the role of pathogenesis-related proteins. *European J. Pl. Pathol.*, 103: 753-765.

- Van Loon LC. 2000. Systemic induced resistance. In A. J. Slusarenko, R. S. S. Fraser & L. C. Van Loon (Eds.), Mechanisms of resistance to plant diseases (pp. 521–574). Dordrecht, The Netherlands: Kluwer Academic Publishers.
- Van Loon LC, Bakker PAHM and Pieterse CMJ. 1998. Systemic resistance induced by rhizosphere bacteria. *Annu. Rev. Phytopathol.*, 36: 453-483.
- Van Loon LC, Bakker PAHM. 2005. Induced systemic resistance as a mechanism of disease suppression by rhizobacteria. In Z. A. Siddiqui (Ed.), PGPR: Biocontrol and biofertilization (pp. 39–66). Dordrecht, The Netherlands: Springer Science.
- Van Loon LC, Bakker PAHM. 2006. Root-associated bacteria inducing systemic resistance. In S. S. Gnanamanickam (Ed.), Plant-associated bacteria (pp. 269–316). Dordrecht, The Netherlands: Springer.
- Van Loon LC, Bakker PAHM, Pieterse CMJ. 1998. Systemic resistance induced by rhizosphere bacteria. *Annual Review of Phytopathology*, 36, 453–483.
- Van Peer R and Schippers B. 1988. Plant growth response in bacterization with selected *Pseudomonas* spp. strains and rhizosphere microbial development in hydroponic cultures. *Canadian J. Microbiol.*, 35: 456-463.
- Van Peer R, Neimann GJ and Schippers B. 1991. Induced resistance and phytoalexin accumulation in biological control of Fusarium wilt of carnation by *Pseudomonas* sp. Strain WCS 417. *Phytopathol.*, 81: 728-734.
- Vessey KJ. 2003. Plant growth promoting rhizobacteria as biofertilizers. *Plant Soil*, 255: 571-586.
- Vidyasekaran P and Muthamilan M. 1995. Development of formulations of *Pseudomonas fluorescens* for control of chickpea wilt. *Pl. Dis.*, 79: 782-786.
- Vidhyasekaran P. 1998. Biological suppression of major diseases of field crops using bacterial antagonists. In: Biological Suppression of Plant Disease, Phytoparasitic Nematodes and Weeds (Eds.) Singh, S.P. and Hussaini, S.S., National seminar on Biological suppression of plant disease, phytoparasitic nematodes and weeds – present scenario and future thrust. Project Directorate of Biological Control, Bangalore, India, pp.81-95.
- Vigo C, Norman JR, Hooker JE. 2000. Biocontrol of the pathogen *Phytophthora parasitica* by arbuscular mycorrhizal fungi is a consequence of effects on infection loci. *Plant Pathol* 49, 509-514.
- Visker MHPW, Keizer LCP, Budding DJ, Van Loon LC, Colon LT, Struik PC. 2003. Leaf position prevails over plant age in reflecting resistance to late blight in potato. *Phytopathology*, 93, 666–674.
- Vivekananthan R, Ravi M, Ramanathan A and Samiyappan R. 2004. Lytic enzymes induced by *Pseudomonas fluorescens* and other biocontrol organisms mediate defence against the anthracnose pathogen in mango. *World J. Microbiol. Biotechnol.*, 20: 235-244.
- Voisard C, Keel C, Haas D and Defago G. 1989. Cyanide production by *Pseudomonas fluorescens* helps suppress black root rot of tobacco under gnotobiotic conditions. *EMBO J.*, 8: 351-358.
- Viswanathan R and Samiyappan R. 1999. Induction of systemic resistance by plant growth promoting rhizobacteria against red rot disease caused by *Colletotrichum falcatum* Went. in sugarcane. *Proc. Sugar Technol. Assoc. India*, 61: 24-39.
- Wang C, Knill E, Glick BR, De fago G. 2000. Effect of transferring 1-aminocyclopropane-1-carboxylic acid (ACC) deaminase genes into *Pseudomonas fluorescens* strain CHA0 and its *gacA* derivative CHA96 on their growth-promoting and disease-suppressive capacities. *Canadian Journal of Microbiology*, 46, 898–907.
- Wei G, Kloepper JW and Tuzun S. 1991. Induction of systemic resistance of cucumber to *Colletotrichum orbiculare* by selected strains of plant growth promoting rhizobacteria. *Phytopathol.*, 81: 1508-1512.
- Wei G, Kloepper JW and Tuzun S. 1996. Induced systemic resistance to cucumber diseases and increased plant growth by plant growth promoting rhizobacteria under field conditions. *Phytopathol.*, 86: 221-224.
- Weststeijn WA. 1990. Fluorescent pseudomonads isolate E11-2 as biological agent for *Pythium* root rot in tulips. *Netherlands J. Pl. Pathol.*, 96: 262-272.
- Whipps JM. 2001. Microbial interactions and biocontrol in the rhizosphere. *J. Exponential Botany*, 52: 487-511.
- Whipps JM. 2004. Prospects and limitations for mycorrhizas in biocontrol of root pathogens. *Can J Bot* 82, 1198-1227.
- Whitehead NA, Barnard AM, Slater H, Simpson NJ, Salmond GP. 2001. Quorum-sensing in Gram-negative bacteria. *FEMS Microbiology Reviews*, 25, 365–404.
- Zehnder GW, Yao C, Wei G and Kloepper JW. 2000. Influence of methyl bromide fumigation on microbe-induced resistance in cucumber. *Biocontrol Sci. Technol.*, 10: 687-693.
- Zehnder G, Kloepper J, Yao CB, Wei G. 1997. Induction of systemic resistance in cucumber against cucumber beetles (Coleoptera: Chrysomelidae) by plant growth-promoting rhizobacteria. *Journal of Economic Entomology*, 90, 391–396.
- Zhang F, Dashti N, Hynes RK, Smith DL. 1996. Plant growth-promoting rhizobacteria and soybean [*Glycine max* (L.) Merr.]. Nodulation and fixation at suboptimal root zone temperatures. *Ann Bot* 7:453–459.
- Zhu HH, Zao Q. 2004. Localized and systemic increase of phenols in tomato roots induced by *Glomus versiforme* inhibits *Ralstonia solanacearum*. *J Phytopathol* 152, 537- 542.
- Zuber P, Nakano MM, Marahiel MA. 1993. Peptide antibiotics. In: Sonenshein AL, Hoch JA, Losick R (eds) *Bacillus subtilis* and other Gram-positive Bacteria. ASM, Washington, USA, pp 897–916.
- Zuccarini P and P Okurowska. 2008. Effects of mycorrhizal colonization and fertilization on growth and photosynthesis of sweet basil under salt stress. *Journal of Plant Nutrition*, 31(3): 497-513.