Fluorescent *Pseudomonas:* A Natural Resource from Soil to Enhance Crop Growth and Health

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María Lis Yanes and Natalia Bajsa

Abstract

Fluorescent *Pseudomonas* had always played an important role in the development of biopesticides and biofertilizers since the concern for more sustainable agricultural production systems exists. They produce a distinctive soluble yellowish-green siderophore called pyoverdin, show an excellent root-colonizing ability, and exert a wide battery of mechanisms to promote plant growth, either directly by facilitating nutrient acquisition or synthesizing phytohormones or indirectly by biological control of plant pathogens. Fluorescent *Pseudomonas* have been applied successfully to control plant pathogens on different pathosystems due to their ability of producing secondary metabolites such as antibiotics, induction of systemic resistance in the host plant, and/or competition for niches and nutrients. They are very suitable for developing market inoculants, as they are abundant in soil and roots, can use a variety of carbon sources, have a high growth rate, can be introduced into the rhizosphere by seed bacterization, and are amenable to genetic manipulation. However, compared to the volume of research that has been performed with these bacteria, few strains have been successfully developed into commercial products for plant biocontrol and biostimulation. Some drawbacks for their field application need to be overcome, as variations observed in field performance, and the constraints found during registration of market products, due to some opportunistic human pathogenic *Pseudomonas* that have been reported. The development of suitable formulations for bacterial delivery, genetic modification of promising strains, and coinoculation with other plant growth-promoting microorganisms are discussed as potential ways of strengthening the use of *Pseudomonas* spp. in agricultural systems.

M.L. Yanes • N. Bajsa (✉)
Laboratorio de Ecología Microbiana, Instituto de Investigaciones Biológicas Clemente Estable, Av. Italia 3318, Montevideo, Uruguay
e-mail: nbajsa@iibce.edu.uy

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15.1 Introduction

Fluorescent *Pseudomonas* had always played an important role in the development of biopesticides and biofertilizers since the concern for more sustainable agricultural production systems exists. A huge background regarding the application of these microorganisms for plant fertility purposes can be traced to the first patent obtained in the 1910s (Coates 1910). Since that, fluorescent *Pseudomonas* have gained more and more relevance, and nowadays they can be compared to rhizobia in terms of scientific knowledge and applied research.

Fluorescent *Pseudomonas* are a heterogeneous collection of non-enteric strains, Gram-negative chemoheterotrophs, and generally aerobic, non-fermentative, and mobile bacteria which have a polar flagellum (Dwivedi and Johri 2003). The genus is comprised of the species *P. aeruginosa*, *P. aureofaciens*, *P. chlororaphis*, *P. fluorescens*, *P. putida*, and more recently *P. protegens* and also plant-pathogenic species such as *P. cichorii* and *P. syringae*. Fluorescent *Pseudomonas* differ from other *Pseudomonas* spp. for their distinctive soluble yellowish-green pigment, which corresponds to the siderophore called pyoverdin, which emits light when exposed to ultraviolet radiation (Meyer and Abdallah 1978). What makes pseudomonads so interesting? It is because of the fact that they have excellent root-colonizing ability and a wide battery of mechanisms to promote plant growth (i) directly by facilitating nutrient acquisition or synthesizing phytohormones and (ii) indirectly by biological control of plant pathogens. Besides, they are very suitable for being applied as inoculants; for instance, they can use a variety of carbon sources, are amenable to genetic manipulation, are abundant in soil and roots, have a high growth rate, and can be introduced into the rhizosphere by seed bacterization (Whipps 2001).

Fluorescent *Pseudomonas* have been applied successfully to control plant pathogens on different pathosystems, and as the interest of reducing chemical pesticide inputs on different crops increases, the number of host plant increases too. Biocontrol mechanisms are diverse and can be simultaneously exerted by a single strain or a combination of strains. The main mechanisms involve the synthesis of secondary metabolites like antibiotics, the induction of systemic resistance in the host plant, and the competition for niches and nutrients.

Direct promotion of plant growth mediated by fluorescent *Pseudomonas* is another approach to reduce the use of chemical fertilizers. Fluorescent *Pseudomonas* exert this effect by improving plant nutritional status and/or by synthesizing plant hormone-like compounds.

15.2 Mechanisms that Promote Plant Health by Fluorescent *Pseudomonas* spp.

15.2.1 Antibiotics: Diversity of Antimicrobial Metabolites Produced by *Pseudomonas* spp.

Early studies on suppression of plant pathogens by antagonistic bacteria were focused on their ability to produce siderophores which efficiently compete for iron

acquisition, so that pathogens remain devoid of this nutrient (Kloepper et al. 1980). However, in the last 30 years, it was demonstrated that other secondary metabolites such as antibiotics, enzymes and volatile compounds have an important role in controlling pathogen development (Weller 2007; De La Fuente et al. 2004).

Most fluorescent *Pseudomonas* strains are capable of synthesizing one or more antibiotics (Weller 2007). The structure, biosynthetic pathways and regulation of the main antibiotics produced by *Pseudomonas* spp. have been fully characterized. Among them are the phenazine derivatives, 2,4-diacetylphloroglucinol, pyrrolnitrin, pyoluteorin and cyclic lipopeptides.

Phenazines are heterocyclic nitrogen-containing secondary metabolites, produced not only by *Pseudomonas* but also by various microbial genera like *Brevibacterium*, *Burkholderia* and *Streptomyces*, among others (Mavrodi et al. 2006). It has been described over 100 phenazine derivatives of microbial origin, and a single microorganism can produce up to ten of these ones (Dwivedi and Johri 2003; Mavrodi et al. 2013). Phenazine compounds have a broad spectrum of activity against bacteria, fungi, and parasites. Their mode of action relies on its redox properties and the capacity to promote the production of toxic reactive oxygen species (ROS) in the target organism (Mavrodi et al. 2012). However, their biocontrol activity has also been attributed to the induction of systemic resistance (ISR) in the host plant and the reduction in toxin production by the pathogen. For instance, phenazine 1-carboxylic acid (PCA)-producing *Pseudomonas fluorescens* LBUM223 controls the development of common scab in potato caused by *Streptomyces scabies*. But the antagonistic effect was demonstrated not to be mediated by antibiosis but, instead, by a reduction in the expression of thaxtomin A by *S. scabies* which is necessary to produce the necrotic lesions or scabs. In field trials, an increase of 46 % in tuber weight was achieved by inoculation with LBUM223, which did not reduce pathogen soil populations, but altered the expression of a key pathogenesis gene, leading to reduced virulence (Arseneault et al. 2013, 2015).

Five enzymes are required to transform the chorismate in pyocyanin (PYO) or phenazine-1,6-dicarboxylic acid (PDC), which are encoded by a gene cluster that also contains genes involved in regulation, transport, resistance and transformation of PYO/PDC to strain-specific phenazine derivatives (Mavrodi et al. 2013). Several studies have demonstrated that phenazines are involved in the control of soilborne fungal pathogens. It was shown that some indigenous PCA-producing *Pseudomonas* strains could locally reach high concentrations of the antibiotic around the roots, estimated over 100 mM, enough to inhibit *Rhizoctonia solani* AG-8, an important pathogen of wheat (Mavrodi et al. 2012). The phenazines produced by *Pseudomonas* spp. are also able to inhibit pathogens such as *F. oxysporum*, *Pythium* spp., *Gibberella avenacea*, *Alternaria* spp. and *Drechslera graminea* (Mavrodi et al. 2006).

The antimicrobial metabolite 2,4-diacetylphloroglucinol (DAPG) is another example of a widely characterized antibiotic produced by fluorescent *Pseudomonas* spp. This polyketide compound is responsible for suppressiveness of plant diseases such as tobacco black root rot (Keel et al. 1996) and wheat take-all (Raaijmakers and Weller 1998). DAPG spectrum of action is quite broad and covers bacteria, fungi, oomycetes and nematodes. Recent studies in *Saccharomyces cerevisiae*

provided evidence of the mode of action of DAPG. This compound acts as a proton ionophore interrupting the proton gradient in the mitochondrial membrane, which explains their broad spectrum of action in various eukaryotes (Gleeson et al. 2010; Troppens et al. 2013). The biosynthetic locus involved in the antibiotic production includes *phlA*, *phlC*, *phlB* and *phlD*, responsible for the production of monoacetylphloroglucinol (MAPG) and its conversion to 2,4-DAPG, together with genes encoding efflux and repressor proteins (Bangera and Thomashow 1996; Bangera and Thomashow 1999).

Pyrrolnitrin and pyoluteorin are also extensively characterized antibiotics. The first is a chlorinated phenylpyrrole which is synthesized from L-tryptophan and whose mode of action involves the inhibition of the respiratory chain in fungus. Given its strong antifungal activity, it has been used to develop topical antimycotics for human use or even fungicides for agricultural application (Gross and Loper 2009). The biosynthetic gene cluster involved in pyrrolnitrin synthesis consists of four highly conserved genes, *prnABCD*, that encode enzymes responsible for the conversion of L-tryptophan in pyrrolnitrin in *P. fluorescens*. On the other hand, pyoluteorin is an aromatic polyketide antibiotic that consists in a resorcinol ring attached to a dichlorinated pyrrole moiety. A hybrid nonribosomal peptide synthetase/polyketide synthase (NRPS/PKS) is responsible for pyoluteorin synthesis: the resorcinol ring is synthesized by PKS, while the dichloropyrrol is synthesized from L-proline by NRPS. The biosynthetic operon contains 17 genes, including transport and regulatory genes (Nowak-Thompson et al. 1999). Pyoluteorin is especially toxic to oomycetes and some fungi and bacteria (Howell and Stipanovic 1980), but its mode of action has not yet been clarified.

Fluorescent *Pseudomonas* spp. can also synthesize volatile antimicrobial compounds whose actions are exerted at greater distances than diffusible antibiotics. Within volatile compounds, hydrogen cyanide (HCN) has been the most studied and it is commonly produced by pseudomonads inhabiting soils (Gross and Loper 2009). HCN is a strong inhibitor of metalloenzymes such as cytochrome oxidase; therefore, it is toxic for most organisms (Blumer and Haas 2000). Its production in the rhizosphere environment has shown to contribute to the control of pathogens. The compound is synthesized from glycine by an HCN synthase complex, which is encoded by three structural genes *hcnABC*. The operon is highly conserved among cyanogenic *Pseudomonas* (Loper and Gross 2007), but the genomic context is not conserved, differing among species. The HCN biosynthetic locus had also been reported in *Chromobacterium violaceum* and several species of *Burkholderia*. An example of biological control of a plant pathogen mediated by HCN production is by the use of *P. fluorescens* CHA0 whose ability to produce HCN contributes to the biocontrol of black root rot caused by *Thielaviopsis basicola*, in tobacco (Voisard et al. 1989). There are reports of *Pseudomonas* that inhibit the growth of pathogenic fungi by the production of volatile compounds other than HCN. In many cases these compounds were not identified (Fernando et al. 2005; Trivedi et al. 2008; Weisskopf 2013). But, in other cases, the presence of undecene, undecadiene, (benzyloxy)benzonitrile (Kai et al. 2007), nonanal, benzothiazole and 2-ethyl-1-hexanol (Athukorala et al. 2010) was found.

Another group of secondary metabolites widely distributed among the fluorescent *Pseudomonas* spp. is cyclic lipopeptide (CLP) biosurfactants. These compounds are also produced by *Bacillus* spp. They are amphipathic compounds with an enormous structural diversity that reflects a broad variety of natural roles, some of which may be unique to the producing microorganism (Raaijmakers et al. 2010). CLP are basically composed of a lipid chain attached to a short oligopeptide (8–25 amino acids) that can be linear or cyclic (Raaijmakers et al. 2006). The peptide moiety is synthesized by NRPS which are able to incorporate non-proteinogenic amino acids such as D-amino acids, β-amino acids and hydroxy- or N-methylated residues (Mootz et al. 2002). These non-proteinogenic amino acids protect the synthesized peptide against the action of ubiquitin-dependent proteases (Hashizume and Nishimura 2008). An initial condensation domain is responsible for the attachment of the lipid moiety to the growing peptide chain, which can be of different length even for the same CLP. These compounds usually show strong lytic and growth-inhibitory activities against a variety of microorganisms including viruses, mycoplasma, bacteria, fungi and oomycetes, giving to the producing microorganisms an important advantage in the competition for niches (Raaijmakers et al. 2010). The proposed mode of action is the disruption of cell membranes by pore formation which triggers an imbalance in transmembrane ion fluxes and cell death (van de Mortel et al. 2009). CLP also have a role in bacterial motility and solubilization and diffusion of substrates. Mutants unable to produce CLP usually show little or no motility, which can be recovered by exogenously addition of CLP (de Bruijn et al. 2007). This function is important for microbial dispersal and colonization of ecological niches. Various strains of *Pseudomonas* spp. have proven to be more effective in colonization of roots than their mutant strains (Braun et al. 2001; Nielsen et al. 2005; Tran et al. 2007). CLP might also have an important role in adhesion to surfaces and biofilm formation that depends on the structure of the CPL. Some mutant strains of *Pseudomonas* spp., unable to produce CLP, generate unstable biofilm as compared with the wild strain (Roongsawang et al. 2003; Kuiper et al. 2004). In other cases, the absence of CLP caused a significant reduction in biofilm formation (de Bruijn et al. 2007; de Bruijn et al. 2008). Finally, CLP may have a role in the induction of systemic resistance (ISR) in plants. For example, inoculation of tomato plants with a massetolide (CLP)-producing strain or the pure compound increased the plant leaves' resistance against *Phytophthora infestans* infection (Tran et al. 2007). It is unknown if specific receptors are needed in host plants, but it is thought that CLP can generate distortions or transient channels in plant membranes, initiating a cascade of responses that leads to an increased expression of the plant defense system (Jourdan et al. 2009).

Successful examples of biological control mediated by antibiotic-producing fluorescent *Pseudomonas* were described for forage legumes damping-off seedling diseases (De La Fuente et al. 2004; Quagliotto et al. 2009; Yanes et al. 2012). For instance, *P. protegens* UP61 strain, which can produce HCN, 2,4-diacetylphloroglucinol, pyrrolnitrin and pyoluteorin, can densely colonize the roots of lotus (*Lotus corniculatus*) and alfalfa (*Medicago sativa* L.) and is capable of controlling damping-off caused by *Pythium* spp. on both forage legumes (De La

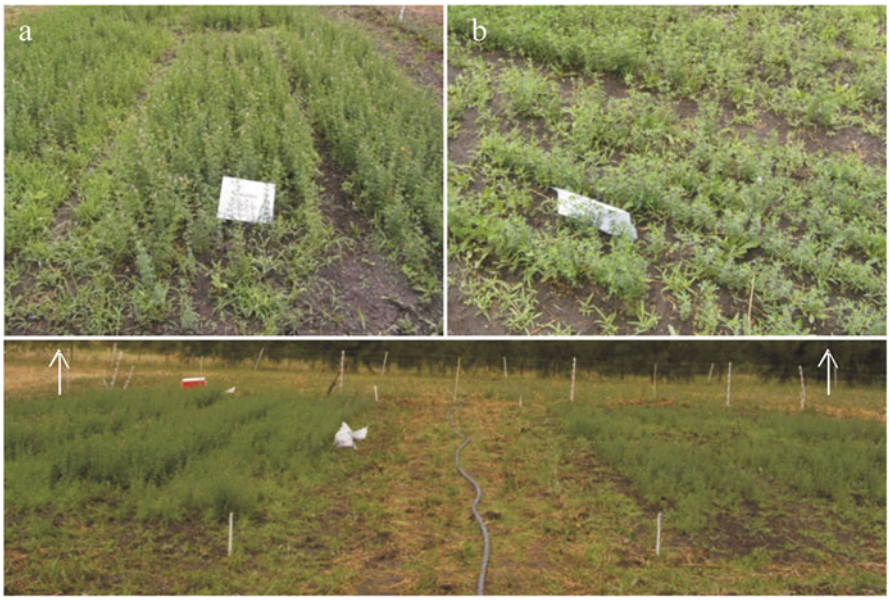

Fig. 15.1 Field assay conducted to determine the biocontrol activity and plant growth-promoting effect of *P. fluorescens* C119 strain. Alfalfa seeds were coinoculated with the pseudomonad strain and *Sinorhizobium meliloti* using a peat-based formulation. Coinoculated plants (**a**) showed a significant increase in alfalfa yield in comparison to the control plants (**b**) which were inoculated only with *S. meliloti*. The picture in the *bottom* shows a partial view of the field assay

Fuente et al., 2004; Quagliotto et al. 2009). Several strains isolated from alfalfa rhizosphere also demonstrated a significant ability to control seedling diseases caused by *Pythium debaryanum* and promoted alfalfa growth (Yanes et al. 2012). Among these strains *P. fluorescens* C119, which produces a CLP with antimicrobial activity, notably promoted the growth of alfalfa (Fig. 15.1).

15.2.2 Biological Control Mediated by Niche and Nutrient Competition

Successful root colonization depends on the ability to compete for nutrients in the root. Competition between beneficial and pathogenic microorganisms by niches in the root surface can result in a decrease in the severity of crop diseases (Kamilova et al. 2005). It is important that beneficial microorganisms colonize efficiently the site of the root that the pathogen uses to get into the plant. Pliego and coworkers (2008) isolated the strains *Pseudomonas pseudoalcaligenes* AVO110 and *P. alcaligenes* AVO73, which were selected by their excellent root-colonizing ability in avocado and antagonism against *Rosellinia necatrix.* Both strains efficiently colonized

different sites of the root, but only AVO110, which was located on the same site as the pathogen, demonstrated a significant protection effect against avocado white root rot.

Competition for iron as biological control mechanism has been widely documented. In the soil, iron availability in the soluble form Fe^{3+} is generally limited. To get access to this nutrient, many microorganisms produce siderophores which are chelant molecules with high affinity for iron. Competition for iron was first reported by Kloepper and colleagues in 1980. Siderophores are structurally diverse and are classified according to their functional group as catechols (enterobactin, vibriobactin, yersiniabactin and pyochelin), hydroxamates (alcaligin and deferoxamine B), and carboxylates (staphyloferrin A and achromobactin) (Miethke and Marahiel 2007). Some *Pseudomonas* are able to internalize the ferric ion using heterologous siderophores synthesized by cohabiting rhizosphere microorganisms, producing a nutrient deficiency that affects the development of competing microorganisms (Loper and Henkels 1999; Saharan and Nehra 2011). An example of an iron competitor strain is the pseudobactin producer *P. putida* WCS358 that is capable to control diseases caused by *Fusarium* spp. in carnation and radish (Weisbeek and Gerrits 1999). The effectiveness of this mechanism was demonstrated by an increase in potato yield when seeds were treated with WCS358 wild-type strain, whereas a siderophore-negative mutant had no effect on tuber yield in field assays (Duijff et al. 1994).

15.2.3 Induction of Defense Responses in Plants

Some nonpathogenic rhizobacteria interact with host plants stimulating an alertness state that would protect plants against subsequent pathogen attack, a phenomenon known as induction of systemic resistance (ISR). This state of preparation for a possible attack enables a fast triggering of defense responses which spread to every organ of the plant, in the presence of a pathogen. While several molecules synthesized by inducing defense bacteria are known to stimulate ISR, there is little information about the molecular mechanisms involved in plant cell recognition of such elicitors. Bacteria-produced molecules involved in the ISR phenotype are lipopolysaccharide (Leeman et al. 1995), siderophores (Maurhofer et al. 1994), flagella (Meziane et al. 2005), and some antibiotics such as those produced by fluorescent *Pseudomonas* spp.: 2,4-diacetylphloroglucinol (Weller et al. 2012), pyocyanin (Audenaert et al. 2002), biosurfactants (Ongena and Jacques 2008), and volatile organic compounds (Ryu et al. 2004). Given this wide variety of elicitors and the enormous diversity of microorganisms that inhabit the rhizosphere, we would expect that plants stand in a continuous state of alert. However, field studies have determined that an effective ISR requires a population density of at least 10^7 CFU of the inducing bacteria per gram of root. For *Pseudomonas* spp., a dose-response study has shown that there is a threshold of 10^5 CFU per gram of root to trigger ISR responses (Raaijmakers et al. 1995). As it is very unlikely to find such a population

density of a specific bacterial genotype on a rhizospheric soil, thus it is very unlikely to find a natural phenomenon of ISR in a soil, except in a suppressive soil (Bakker et al. 2013).

15.3 Mechanisms Involved in Plant Growth Improvement by Rhizospheric *Pseudomonas* spp.

15.3.1 Nutrient Contribution to the Host Plant

Some bacteria including fluorescent *Pseudomonas* spp. can promote plant growth by two major mechanisms: assisting plant nutrition by solubilization of soil nutrients and/or contributing to the pool of the host plant's growth hormones.

Microorganisms that produce metabolites or enzymes that increase the bioavailability of essential plant nutrients such as nitrogen, phosphorus and iron are called biofertilizers (Glick et al. 2007; Berg 2009). The supply of nitrogen by atmospheric N_2-fixing bacteria is the most widely studied example of biofertilization. These bacteria can be endosymbionts, as rhizobia, or free-living diazotrophs that colonize plant rhizosphere, as *Azospirillum*. They synthesize an enzyme complex called nitrogenase which catalyzes, by an energetically costly process, the reduction of N_2 to ammonia; then, the ammonia is taken up by the plant for the biosynthesis of N compounds. Both plants and microorganisms are benefited, because the plants in turn provide carbonated compounds (photosynthates) to the symbiont (Berg 2009). Until a few years ago, it was thought that the genus *Pseudomonas* did not perform atmospheric nitrogen fixation. However, after sequencing the entire genome of *Pseudomonas stutzeri* A1501, it was found that the strain has all the genes necessary to fix N_2, organized in a genomic island possibly obtained by horizontal gene transfer (Yan et al. 2008). This genomic region of *P. stutzeri* A1501 was used to transform *P. protegens* Pf-5 strain, an excellent biocontrol agent of plant diseases. The transformed strain was able to promote the growth of *Arabidopsis thaliana* and other plants of agronomic interest such as alfalfa, fescue and corn under conditions of nitrogen deficiency, opening a new perspective for the production of genetically modified inoculants (Setten et al. 2013).

Phosphorus is an important nutrient and the second limiting element of plant growth, after nitrogen (Gyaneshwar et al. 2002). Phosphate-solubilizing bacteria are another example of biofertilizers, which are taking great relevance due to the high agronomic demand for this nutrient. Most agricultural soils have large reserves of phosphorus due to the repeated application of chemical fertilizers. However, phosphates react with numerous ionic components of the soil, being rapidly immobilized in organic forms by metabolic reactions or in soil mineral particles by adsorption and precipitation. These forms of phosphate are not bioavailable to plants, and only a small fraction of these phosphate forms are available for root absorption (Gyaneshwar et al. 2002). Between 30 and 70 % of the phosphorus in the soil is in organic forms (Shang et al. 1996). Soil microorganisms play an important role in the phosphorus cycle as they produce molecules that assist phosphorus

assimilation by plants (Richardson and Simpson 2011). Two processes are involved in the mobilization of phosphates by soil microorganisms: mineralization of organic phosphorus and solubilization of phosphorus from inorganic sources. Various enzymes such as nonspecific phosphatases, phytases, phosphonatases and C-P lyases release the phosphorus from the organic fraction, while the inorganic phosphorous is mainly released by organic acids such as gluconic acid (Berg 2009; Werra et al. 2009). In summary, phosphate-solubilizing bacteria (PSB) can increase the phosphorus content in plant tissues (Awasthi et al. 2011). Among the best-known PSB are species belonging to the genera *Pseudomonas*, *Bacillus*, *Paenibacillus*, *Escherichia*, *Acinetobacter*, *Enterobacter* and *Burkholderia* (Collavino et al. 2010).

15.3.2 Plant Growth Promotion Mediated by Microbial Phytohormone Production

Plant hormones govern plant growth by spatial and temporal control of division, growth and differentiation of cells. Phytohormones also play an essential role in responses to biotic and abiotic stresses (Peleg and Blumwald 2011). Microorganisms that produce phytohormone-like compounds contribute to the pool of hormones produced by the host plant, intervening in the physiology and promoting plant survival (Dodd et al. 2010; Morel and Castro-Sowinski 2013). Phytohormones act at low concentrations and can be classified in three types: auxins, cytokinins and gibberellins. Indole-3-acetic acid (IAA) is the most commonly found auxin among rhizobacteria. There are at least five biosynthetic pathways for the synthesis of IAA, which mostly use tryptophan (Trp) as precursor. The presence of IAA in the rhizosphere stimulates the formation of lateral roots and root hairs, which greatly increases the absorption surface of roots (Duca et al. 2014). Plants that produce root exudates rich in tryptophan are more prone to be affected by IAA-producing bacteria than those that do not excrete this amino acid precursor. For example, a growth-promoting effect was observed in radish plants, which excrete large amounts of Trp, when inoculated with the IAA-producing strain *P. fluorescens* WCS365, while no effect was observed in plants that excrete tenfold less Trp like tomato, cucumber and pepper (Kamilova et al. 2006).

Cytokinins are involved in cell division and have effects on roots, leaves, flowers, fruits and seeds. The apex of the roots and germinating seeds contains high concentration of cytokinins (Pliego et al. 2011). Members of the genera *Pseudomonas*, *Agrobacterium*, *Erwinia*, *Paenibacillus*, *Azotobacter*, *Azospirillum*, *Bacillus* and *Rhizobium* are cytokinin-producing bacteria, which are also capable of promoting plant growth (García de Salamone et al. 2001).

Gibberellins are involved in various physiological processes of higher plants, in especial root elongation (Pliego et al. 2011). There are more than 130 known gibberellins, which are diterpenoids synthesized by fungi and bacteria (Dodd et al. 2010). The first report of a bacterial gibberellin biosynthetic pathway was for a strain of *B. japonicum* (Morrone et al. 2009). Kang and coworkers (2014)

demonstrated that the gibberellin-producing strain *P. putida* H-2-3 provides the hormone to gibberellin biosynthesis-deficient mutant Waito-C rice plants and also enhances the growth of soybean under saline or drought stresses.

Some bacteria, including *Pseudomonas* spp., are able to decrease the formation of ethylene by plants through the production of the enzyme 1-aminocyclopropane-1-carboxylic acid (ACC) deaminase. The enzyme hydrolyzes ACC, a precursor of ethylene synthesis, with the formation of α-ketobutyrate and ammonium. Plants produce ethylene in response to environmental stresses (Penrose et al. 2001). In these situations plants produce two pulses of ethylene: a small pulse a few hours after the sustained stress is imposed, which is likely to activate the defense genes, and a second pulse of production, a more intense one a few days later. This second pulse of ethylene production generates negative effects on the plant such as senescence, abscission and chlorosis (Glick et al. 2007). It has been proposed a model that explains the promoting effect of ACC deaminase-producing bacteria (Glick et al. 2007) as follows: IAA-producing bacteria can stimulate the synthesis of ACC synthase in plants, which converts S-adenosyl methionine (SAM) in ACC. The ACC is partly exuded by the roots where it is hydrolyzed by ACC deaminase-producing bacteria. To maintain the balance between the concentrations of ACC in and out of the roots, the plant secretes more ACC, decreasing the amount of available ACC necessary to produce ethylene and thus reducing the inhibition effect imposed by ethylene in a stressful situation. The repression of IAA synthesis mediated by ethylene also decreases which results in an increased production of this hormone, increasing the promoting effect. Genes for synthesis of ACC deaminase have been characterized in several strains of *Pseudomonas* spp. (Klee et al. 1991; Cheng et al. 2007), and many of them had proved to be involved in the phenotype of plant growth promotion under stress conditions. For instance, Zahir and colleagues (2011) reported a significant improvement of root elongation, nodulation, and yield of lentil when plants were coinoculated with ACC deaminase-producing *Pseudomonas jessenii* and *Rhizobium leguminosarum* in pot assays and field trial. They showed the implication of ACC deaminase-producing bacteria in the recovery of stressed lentil plants by a bioassay that involved the addition of 6 mM ACC to etiolated lentil seedlings. This triggered an ethylene-specific triple response that consisted in a decrease in root length and shoot length and an increase in shoot diameter. Such effect was significantly reduced when seedlings were coinoculated with both rhizobacteria, which were even more effective than the addition of $CoCl_2$, an inhibitor of ethylene production.

15.4 Fluorescent *Pseudomonas* into the Market

Fluorescent *Pseudomonas* have been widely explored as biocontrol agents and biofertilizers due to the several characteristics mentioned above. However, compared to the volume of research that has been performed with these bacteria, few strains have been successfully developed into commercial products. In Table 15.1, a list of

Table 15.1 *Pseudomonas* spp.-based commercial products for biocontrol or biofertilization, formulated alone or in combination with other PGP microorganisms

Species and strain	Product (name/ quantity of products)	Activity or pathogen controlled	Plant culture	Company/country	References
P. fluorescens EG1053	(2)	*Pythium ultimum*, *Rhizoctonia solani*	Cotton	USA	Cook et al. (1996)
P. fluorescens 1629RS	Frostban A y D (2)	Ice nucleation by *Pseudomonas* spp.		Frost Technology Corporation (USA)	Cook et al. (1996)
P. fluorescens NCIB 12089	(1)	Bacterial blight	Edible fungi	USA	Cook et al. (1996)
P. fluorescens A506	BlightBan A506; Frostban A y B (3)	*Erwinia amylovora*, *Pseudomonas* spp. causing blight and freeze damage	Fruits, tomato, potato	Plant Health Technologies Nufarm Inc. Frost Technology Corporation (EEUU)	Cook et al. (1996), Mark et al. (2006), and Zeller (2006) www.nufarm.com
P. aureofaciens Tx-1	BioJect Spot-Less	*Pythium aphanidermatum*, *Sclerotinia homeocarpa*, *Colletotrichum graminicola*, *Microdochium nivale*	Grass	Eco Soil Systems Inc. Turf Science Laboratories (EEUU)	Mark et al. (2006) www.ecosoil.com
P. chlororaphis 63-28	AtEze	*Pythium* spp., *R. solani*, *Fusarium oxysporum*	Garden and greenhouse plants	Eco Soil Systems Inc. Turf Science Laboratories (USA)	Mark et al. (2006) www.ecosoil.com
P. fluorescens CL0145A	Zequanox	*Dreissena polymorpha*, *D. bugensis* (aquatic animals)		Marrone Bio Innovations (USA)	www.marronebioinnovations.com

(continued)

Table 15.1 (continued)

Species and strain	Product (name/ quantity of products)	Activity or pathogen controlled	Plant culture	Company/country	References
P. fluorescens	Rizofos Liq Maíz Rizofos Liq Trigo (2)	Phosphorous solubilization and mineralization; production of phytohormones, siderophores, and antibiotics	Maize, wheat	Rizobacter (Argentina)	Naiman et al. (2009) www.rizobacter.com.ar
P. fluorescens	Fosforiz	Phosphorous solubilization	Beet	FUNDASES (Colombia)	www.fundases.org
P. fluorescens mogo5	Bioprotection Fosforin	*Pythium* sp., *Phytophthora* sp., phosphorous solubilization and mineralization		Dr. Obregón Laboratories (Costa Rica)	www.doctor-obregon.com
P. fluorescens RA56	AgroBac (4)	Several, e.g., *Streptomyces scabies*	Potato, legumes, cereals	Phytobacter (Germany)	Behn (2008)
Pseudomonas sp. DSMZ13134	Proradix (6)	*R. solani*, *Phytophthora* sp., *Erwinia* sp., *Fusarium* sp., *Microdochium nivale*, *P. ultimum*	Potato, tomato, cucumber, paprika, zucchini, grass	Sourcon-Padena (Germany)	Buddrus-Schiemann et al. (2010) http://www.sourcon-padena.com
P. chlororaphis MA 342	Cedomon, Cerall, Cedress	Seed-borne fungi and leave diseases	Cereals (barley, oat, wheat, rye)	BioAgri (Austria, Finland, Norway, Sweden)	Johnsson et al. (1998) and Mark et al. (2006) www.bioagri.se
P. fluorescens 2P24		*Ralstonia solanacearum*, *F. oxysporum*, *R. solani*		China	Gao et al. (2012)

P. fluorescens	Ecomonas	*R. solani*	Rice	India	Vijay Krishna Kumar et al. (2009)
P. fluorescens	Florezen P	*R. solani*	Rice	India	Vijay Krishna Kumar et al. (2009)
P. fluorescens	Bio Protector	Damping-off, wilt, blight	Rice	Manidharma Biotech (India)	www.manidharmabiotech.com
P. fluorescens				Zen Cropcare (India)	www.zencropcare.com
P. fluorescens	Deepa Bio Plus – Pseudo	Wilt, blight		Deepa Farm Inputs (India)	www.indiamart.com/deepa-farminputs-pvtltd
P. fluorescens	Basmonas	Damping-off, wilt, blight	Several	Basarass Biocon (India)	www.indiamart.com/basarass-bioconpvtltd
P. fluorescens		Bactericide, fungicide		Bharat Biocon (India)	www.indiamart.com/bharat-biocon
P. fluorescens	TNAU – Pf1	Soil fungi	Several	Tamil Nadu Agricultural University (India)	www.indiamart.com/tnau
P. fluorescens		Pathogen fungi		Rajathi Group (India)	www.indiamart.com/rajathi-group
P. fluorescens	Yash Pseudomonas			Yash Krishi (India)	www.indiamart.com/yashkrishi-takniki-ewam
P. fluorescens	Conquer/Victus	*Pseudomonas tolaasii*	Edible fungi	Mauri Foods/Sylvan Spawn (Australia)	Nakkeeran et al. (2006) and Fernandéz and Juncosa (2002)
P. fluorescens	Biomonas	*Schlerotinia*, *Rhizoctonia*, *Pythium*, *Alternaria*, *Ascochyta*, *Cercospora*, *Macrophomina*, *Myrothecium*, *Ramularia*, *Xanthomonas*, *Erwinia*, *Fusarium*, *Verticillium*, *downy mildews*, *powdery mildews*	Cotton, cereals, pulses, vegetables, oilseeds, fruit, and floriculture	Biotech International (India)	www.biotech-int.com Bettiol et al. (2012)

(continued)

Table 15.1 (continued)

Species and strain	Product (name/ quantity of products)	Activity or pathogen controlled	Plant culture	Company/country	References
P. syringae ESC-10 and ESC-11	Bio-save 10 and 11	Postharvest diseases (*Penicillium*, *Botrytis*, *Mucor*, *Helminthosporium*, *Rhizopus*)	Apple, pear, cherry, potato, sweet potato	Jet Harvest Solutions (USA)	Bettiol et al. (2012)
P. fluorescens (yeast and *two Bacillus* spp.*)*	BioGro	Nitrogen fixation, phosphorous solubilization, organic matter mineralization	Rice	Hanoi University of Science (Vietnam)	Adesemoye and Egamberdieva (2013)
P. fluorescens, *Trichoderma viride*	ANOKA	Control of damping-off, seed and root, nematode wilt		Bio Sciences (India)	Woo et al. (2014)
P. fluorescens, *Trichoderma viride*, *Trichoderma harzianum*, *Bacillus subtilis*	Bio Protector	Root-/soilborne and air borne diseases		Bacto Agro Culture Care	Woo et al. (2014)
Pseudomonas, *Trichoderma harzianum*, *Glomus intraradices*	Micover Gold & Plus	Soilborne pathogens		Agrifutur (European Union)	Woo et al. (2014)
Pseudomonas	Micosat Fito	Induced resistance to bacteria, soilborne fungi, pathogens, insects, and nematodes		CCS Aosta (Italy)	Woo et al. (2014)
T. harzianum					
Glomus spp., *Pichia*					

Pseudomonas-based products for plant biocontrol and biostimulation has been reviewed.

A successful example is the biocontrol agent *Pseudomonas chlororaphis* strain MA342, which has been formulated in several commercial products. Initially, MA342 was tested in 105 field experiments in different zones of Sweden for 5 years to control seed-borne diseases of barley, oats, wheat and rye. After consistent results, comparable to chemical fungicide application, an inoculant formulation was developed for the protection of cereals and then for peas. However, the performance of these microorganisms may not always be consistent, as showed by their high effectiveness against *Alternaria* spp. on cabbage and carrots and against *Colletotrichum lindemuthianum* on beans, but a poor performance in seed-borne *Ascochyta* spp. on peas (Amein et al. 2011; Johnsson et al. 1998; Tinivella et al. 2009). Thus, all plant-pathogen interactions can not be overcome by the inoculation with a single *Pseudomonas* strain.

15.5 Benefits and Challenges of Using Fluorescent *Pseudomonas* for Plant Growth Promotion

The application of beneficial microorganisms to improve plant health of economically important crops has many advantages over chemically synthesized pesticides and fertilizers as they are safer with the environment, have a specific targeted activity, do not favor the development of resistant pathogens due to the diversity of mechanisms involved in their biocontrol phenotype, are effective in small doses, decay faster, and can be applied either in conventional or integrated management systems (Berg 2009). Fluorescent *Pseudomonas* are a good example of the sustainable use of microbes, but the commercial adoption of agricultural products based on these bacteria is not so widespread, due to some drawbacks that their application still present and that need to be overcome.

15.5.1 Consistency in Field Performance

As also happens with other plant growth-promoting rhizobacteria (PGPR) (except from rhizobia and some free-living nitrogen fixers), the lack of consistency in field performance of *Pseudomonas*-based bioproducts, in different crops and conditions, is a limitation for its commercial application (Barreto et al. 2010). In some cases, there is a gap between results obtained in controlled laboratory conditions and those achieved in the field. Different host-plant genotypes produce root exudates with different chemical compositions. These variations may play a role in the bacterial responses, which are needed to support inoculum presence and activity (Khalid et al. 2004; Dalmastri et al. 1999). Usually, the process of plant breeding for the selection of cultivars does not take into account the association of plant with beneficial microorganisms. Considering this aspect, it would be a valuable tool to coordinate the selection of host plant varieties and biofertilizers; this strategy could

enhance the ability of the microorganism to interact with the plant and to express promotion traits, e.g., production and responsiveness to hormones (Remans et al. 2008).

The failure of microorganism-based protectant formulations under field conditions has been explained by their low stability, with limited persistence and resistance, but not by a lack of effectiveness (Dorn et al. 2007). This phenomenon can be sometimes explained by the complex regulation of biocontrol-related traits as colonization and production of active metabolites, which are highly dependent on environmental conditions. In some cases, the need for the inoculation with a high population of the biocontrol agent, in order to achieve effective control of the pathogen, is also a limiting factor.

Usually, successful field inoculation experiments have been difficult to establish due to the low disease incidence of plant pathogens during the assays. The strain *P. chlororaphis* R47 was highly active in the protection of potato against *Phytophthora infestans* in vitro and in greenhouse, but its activity could not be verified in the field due to unfavorable infection conditions (Guyer et al. 2015). In another study, from 20 field trials installed in two different regions of Uruguay, and during a 4-year evaluation of effectiveness of three *P. fluorescens* strains to protect alfalfa from emergence diseases, only 11 trials were conducive for damping-off (Quagliotto et al. 2009).

The challenges to overcome this limitation for a wider adoption of *Pseudomonas* spp. as biofertilizers include the characterization of the ecology and colonization behavior of these bacteria in the rhizosphere at different situations, as well as their mechanisms of plant promotion and regulation. The development of more stable formulations will enable the strains to be established at adequate cell densities in order to behave as plant promoters under field conditions. A comprehensive consideration of the biology and ecology of the crop system, and the agricultural practices, will help to reduce the variation of results in field performance of these inoculant bioformulations (Barreto et al. 2010; Guyer et al. 2015).

15.5.2 Is Genetic Modification an Option?

Some researchers have proposed the development of genetically modified (GM) microorganisms aimed to combine different plant growth-promoting abilities in a single microbe. For example, the introduction of a plasmid carrying the genomic information for IAA production into *P. fluorescens* BSP53a, a strain that has the ability to block the development of some phytopathogenic fungi due to siderophore synthesis, was seen as an interesting inoculant strain, which combines plant stimulation and protection effects. The recombinant strain produced an increase in root weight and changes in root morphology, as compared to the wild-type strain in black currant, but not in cherry plants (Dubeikovsky et al. 1993).

Several studies have incorporated antimicrobial traits in *Pseudomonas* spp strains. When their impact on resident microbial communities was analyzed, the effect caused by inoculation was transient, suggesting a rapid inactivation of the antibiotic

in the soil (Bajsa et al. 2013). However, a long-term effect in wheat rhizosphere was caused by *P. putida* strains that constitutively express PCA or DAPG, which changed the resident bacterial communities in a 4-year experiment (Viebahn et al. 2006).

This approach, the use of GM microorganisms, is a matter of serious controversy in terms of the spread of a GM bacterium in nature, especially if the microbe has better colonization, competence, or persistent abilities in the natural environment. The constraints for the registration of natural microbes for their application in the environment are numerous, but even more are with GM strains that would involve a more detailed risk assessment.

15.5.3 Let's Cooperate: Coinoculation

Another strategy to enhance effectiveness and reduce variation in performance is the use of bioproducts with a *Pseudomonas* strain in combination with another microorganism, presenting additive or synergistic effects, such as nitrogen-fixing bacteria, mycorrhizal fungi, or other plant biocontrol agents. The rhizosphere is a complex ecosystem, as are the interactions involved in plant growth stimulation or disease, so the use of bioformulations containing several PGPR can improve their effectiveness, as compared with single bacterial inoculation (Barreto et al. 2010; Morel and Castro-Sowinski 2013).

Nitrogen-fixing bacteria are widespread PGPR used to improve crop yield, and the nitrogen fertilization using chemicals could be completely prevented if the right strain and adequate management practices are employed. The combination of diazotrophs with other PGPR strain has been widely reported. The coinoculation practice, using *Pseudomonas* spp. and *Rhizobium* spp., was demonstrated to enhance nodulation, nitrogen fixation, plant biomass and grain yield in various leguminous species including alfalfa, soybean, green gram, and chickpea. For example, the nodule occupancy by a *Rhizobium* strain in pigeon pea increased from 50 % to 85 % in the presence of *Pseudomonas putida*, improving plant growth and nitrogen fixation (Tilak et al. 2006; Remans et al. 2007). In addition, *in vitro* and in-field experiments carried on in lotus and alfalfa plants showed that the coinoculation with rhizobia and fluorescent *Pseudomonas* decreases damping-off incidence and fixes nitrogen, compared with single inoculation (Quagliotto et al. 2009; De La Fuente et al. 2002).

The success of this mixed formulation depends also on abiotic factors as nutrient availability, as showed by the following examples. ACC deaminase-producing *P. putida* UW4 is able to enhance nodulation by *Rhizobium* in *Phaseolus vulgaris* mainly under P-deficiency conditions. In this stress situation, the production of ethylene is increased and bacterial ACC deaminase activity can reduce this stress response and revert nodulation inhibition (Remans et al. 2007). ACC deaminase-producing *Pseudomonas* sp. also promotes groundnut (*Arachis hypogea*) nodulation by *Bradyrhizobium* under saline stress condition (Saravanakumar and Samiyappan 2007).

Some bacteria, including strains of *Pseudomonas* spp., can act as mycorrhiza helper bacteria (MHB), assisting the establishment or promoting the functioning of

arbuscular or ectomycorrhizal symbiosis. They can mobilize nutrients from soil, fix nitrogen and protect plant from pathogens, which can lead to enhance fungal germination and root colonization (Frey-Klett et al. 2007; Velivelli et al. 2015). A few examples include the increased infection by ectomycorrhizal fungi as *Laccaria* spp. and *Pisolithus alba* in Douglas fir, *Eucalyptus* and *Acacia* trees, higher colonization by endomycorrhizal fungi *Glomus* spp. in diverse plant species (maize, potato, tomato, wheat, barley, clover and *Acacia* sp.), and protection of papaya from *Fusarium oxysporum* infection by coinoculation with *Pseudomonas* spp. and *Glomus* spp. (Frey-Klett et al. 2007; Hernández-Montiel et al. 2013). The molecular mechanisms involved in these tripartite interactions are poorly understood. For the establishment of the bacteria/mycorrhizal fungus/plant network, the release of active biomolecules and physical contact among the partners seem important, where quorum sensing, biofilm formation and secretion systems seem to be involved (Bonfante and Anca 2009). For instance, ACC deaminase and ethylene production by *P. putida* is involved in the promotion of endomycorrization by *Gigaspora rosea* in cucumber (Gamalero et al. 2008). Carbon sources may also be involved in the selection of bacterial communities associated with the mycorrhizosphere. The secretion of trehalose, the main carbohydrate used as carbon sink by the fungus, acts in facilitating the colonization of MH *Pseudomonas* and formation of biofilms on hyphae (Duponnois and Kisa 2006; Uroz et al. 2007).

Several *Pseudomonas* strains showed positive effects on the growth of the ectomycorrhizal fungus *Laccaria bicolor* and the effectiveness of the symbiosis with *Populus deltoides* roots. Results suggest that these effects are due to the induction of the presymbiotic status of the fungus (Labbé et al. 2014). In other types of interactions, the production of antibiotics by the MHB is stimulated by the mycorrhiza, but this mechanism has not been described yet in interactions with *Pseudomonas* spp. (Riedlinger et al. 2006).

Plant protection can also be enhanced by the combination of *Pseudomonas* strains with other protective microorganisms. The coinoculation of faba bean (*Vicia faba*) with *Pseudomonas fluorescens* and *Rhizobium leguminosarum* reduced the symptoms produced by yellow mosaic virus, probably by the induction of the systemic resistance of the plant. Among other examples, the coinoculation of cucumber with *Pseudomonas putida* and *Serratia marcescens* and the coinoculation of cucumber and *Arabidopsis thaliana* with *Trichoderma harzianum* and *Pseudomonas* sp. showed additive and positive effects on plants infected with *Fusarium* wilt (Mohr et al. 2008; Liu et al. 1995; Elbadry et al. 2006; Alizadeh et al. 2013).

15.5.4 Suitable Formulations for *Pseudomonas* Delivery

For the successful application of a bioproduct, the formulation (a mixture of ingredients prepared for the direct application on the field or seeds) must deliver the microbial agent in a physiologically active state and with the potential to express all the microbial capabilities (Barreto et al. 2010). Moreover, the product has to be long

lasting for a certain period of time to ensure the proper microbial state for effective field application. As *Pseudomonas* spp. are bacteria that do not produce resistant structures as spores, refrigeration for preservation of the inoculants may be necessary. Temperatures of 4–10 °C enable the conservation of some formulations up to 2–6 months, at room temperature about 3 weeks, but freeze conditions are needed for longer periods. Certain products are marketed as powder-soluble formulas containing lyophilized bacteria and compounds (e.g., milk or canola oil) that improve cell survival and/or effectiveness (Bettiol et al. 2012).

On the other hand, the bacterization of seeds (pre-inoculation of seeds) is also an adequate option of product marketing due to the long-lasting useful life of inoculated seeds that can be stored for long periods (1–2 years) without losing the promoting activity, as it has been shown for *P. chlororaphis*-based products (Johnsson et al. 1998; Bettiol et al. 2012).

Sterile peat is a carrier that has been extensively used for rhizobial formulations, because it allows bacterial survival during storage and commercialization of the final product. It was demonstrated that this carrier is also suitable for fluorescent *Pseudomonas*, maintaining constant populations of $10x10^9$ UFC per gram of peat at 4 °C during 6 months (Bagnasco et al. 1998; Date 2001).

15.5.5 The Tougher Step: Registration

During the process of registration of market products containing microorganisms, and especially when they are based on *Pseudomonas* spp., a large number of constraints are found (Mathre et al. 1999). A risk assessment to human health and to the environment is needed before releasing a product (Fravel 2005). Bacteria of the genus *Pseudomonas* include a few species and strains that can be opportunistic human pathogens, so the massive use of formulations containing this microbe is always under debate. Bacteria unable to grow at human body temperature could circumvent this concern, but forward studies are needed to clearly differentiate truly health threat bacteria and safe ones (Bodilis et al. 2004).

As more information regarding safety of microbial formulation is generated, the big challenge may be the simplification of registration procedures, as it occurs with chemical products whose registration is much more standardized despite its potential but known toxicity.

The ecological impact of some *Pseudomonas* strains is expected to be low, as their populations rapidly decrease after been inoculated, in 1–2 weeks or months in field or greenhouse conditions. This could be a favorable trait for environmental safety but not for its performance in agricultural systems, except if the action that the microbes exert in the rhizosphere is quick enough to induce the positive effect on plants, e.g., during the pathogen control of seedling diseases (Guyer et al. 2015; Quagliotto et al. 2009). Further studies are required to better characterize the ecological behavior of bacteria before their release to the soil.

15.6 Final Remarks

Plant-associated *Pseudomonas* are multifaceted bacteria, with a high metabolic diversity and interesting plant growth traits (Fig. 15.2). Even if there is no doubt regarding the benefits of *Pseudomonas* spp. inoculation on plant growth promotion, there is also a public concern due to their potential as opportunistic pathogenic agents. Thus, further knowledge and experience about their use in agroecosystems, in single or mixed formulation, will give insight to the potential use of *Pseudomonas* spp. in agricultural systems.

The adoption of sustainable agronomical practices for preservation of the native, active and adapted *Pseudomonas* populations in every location would reduce the need for reintroducing them as inoculants (Agaras et al. 2012).

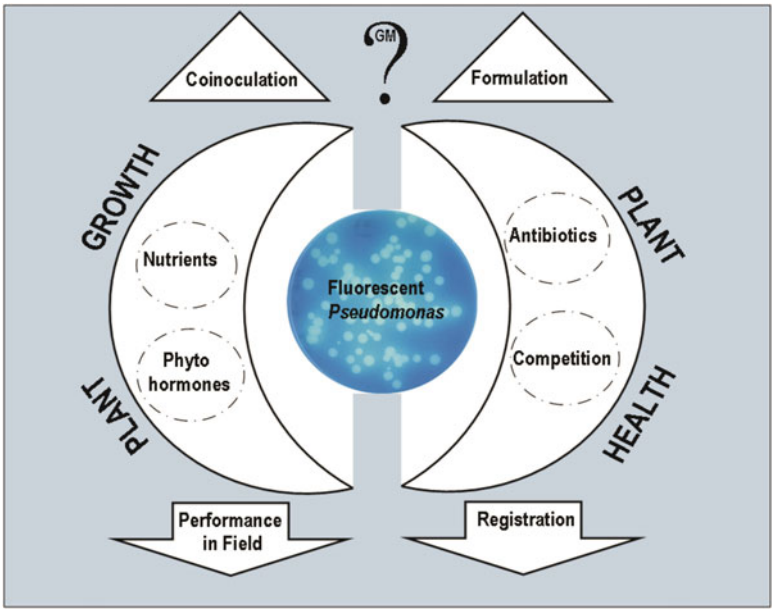

Fig. 15.2 Fluorescent *Pseudomonas*: plant health and plant growth-promoting activities. Limitations in field performance and in registration process have delayed the wider commercial adoption of *Pseudomonas*-based inoculants. The improvement of commercial formulations and/or the coinoculation with other PGPR may overcome those drawbacks. Genetic modification (GM) is a more questionable alternative

References

Adesemoye AO, Egamberdieva D (2013) Beneficial effects of plant growth-promoting rhizobacteria on improved crop production: prospects for developing economies. In: Maheshwari D, Saraf M, Aeron A (eds) Bacteria in agrobiology: crop productivity. Springer, Berlin, pp 45–63

Agaras B, Wall LG, Valverde C (2012) Specific enumeration and analysis of the community structure of culturable pseudomonads in agricultural soils under no-till management in Argentina. Appl Soil Ecol 61:305–319

Alizadeh O, Azarpanah A, Ariana L (2013) Induction and modulation of resistance in crop plants against disease by bioagent fungi (arbuscular mycorrhiza) and hormonal elicitors and plant growth promoting bacteria. Int J Fat Allied Sci 2(22):982–998

Amein T, Amein T, Wright SA, Wikström M, Koch E, Schmitt A, Stephan D, Jahn M, Tinivella F, Gullino ML, Forsberg G, Werner S, van der Wolf J, Groot SPC (2011) Evaluation of non-chemical seed treatment methods for control of *Alternaria brassicicola* on cabbage seeds. J Plant Dis Prot 118(6):214–221

Arseneault T, Goyer C, Filion M (2013) Phenazine production by *Pseudomonas* sp. LBUM223 contributes to the biological control of potato common scab. Phytopathology 103:995–1000

Arseneault T, Goyer C, Filion M (2015) *Pseudomonas fluorescens* LBUM223 increases potato yield and reduces common scab symptoms in the field. Phytopathology 105:1311–1317

Athukorala SNP, Fernando WGD, Rashid KY (2010) The role of volatile and non-volatile antibiotics produced by *Pseudomonas chlororaphis* strain PA23 in its root colonization and control of *Sclerotinia sclerotiorum*. Biocontrol Sci Tech 20(8):875–890

Audenaert K, Pattery T, Cornelis P, Höfte M (2002) Induction of systemic resistance to *Botrytis cinerea* in tomato by *Pseudomonas aeruginosa* 7NSK2: role of salicylic acid, pyochelin, and pyocyanin. Mol Plant-Microbe Interact 15(11):1147–1156

Awasthi R, Tewari R, Nayyar H (2011) Synergy between plants and P-solubilizing microbes in soils: effects on growth and physiology of crops. Int Res J Microbiol 2(12):484–503

Bagnasco P, De La Fuente L, Gualtieri G, Noya F, Arias A (1998) Fluorescent *Pseudomonas* spp. as biocontrol agents against forage legume root pathogenic fungi. Soil Biol Biochem 30(10–11):1317–1322

Bajsa N, Morel MA, Braña V, Castro-Sowinski S (2013) The effect of agricultural practices on resident soil microbial communities: focus on biocontrol and biofertilization. In: de Bruijn FJ (ed) Molecular microbial ecology of the rhizosphere, vol 2. Wiley Blackwell, Hoboken, pp 687–700

Bakker PAHM, Doornbos RF, Zamioudis C, Berendsen RL, Pieterse CMJ (2013) Induced systemic resistance and the rhizosphere microbiome. Plant Pathol J 29(2):136–143

Bangera MG, Thomashow LS (1996) Characterization of a genomic locus required for the synthesis of the antibiotic 2,4-diacetylphloroglucinol by the biological control agent *Pseudomonas fluorescens* Q2-87. Mol Plant-Microbe Interact 9(3):83–90

Bangera MG, Thomashow LS (1999) Identification and characterization of a gene cluster for synthesis of the polyketide antibiotic 2,4- diacetylphloroglucinol from *Pseudomonas fluorescens* Q2-87. J Bacteriol 181:3155–3163

Barreto V, Seldin L, Araujo FFD (2010) Plant growth and health promoting bacteria. Microbiol Monog 18:21–44

Behn O (2008) Influence of *Pseudomonas fluorescens* and arbuscular mycorrhiza on the growth, yield, quality and resistance of wheat infected with *Gaeumannomyces graminis*. J Plant Dis Prot 115(1):4–8

Berg G (2009) Plant-microbe interactions promoting plant growth and health: perspectives for controlled use of microorganisms in agriculture. Appl Microbiol Biotechnol 84(1):11–18

Bettiol W, Morandi MAB, Pinto ZV, de Paula Jr TJ, Corrêa EB, Moura AB, Lucon CMM, Costa JCB, Bezerra JL (2012) Produtos comerciais à base de agentes de biocontrole de doenças de plantas, Jaguariúna, SP: Embrapa Meio Ambiente

Blumer C, Haas D (2000) Mechanism, regulation, and ecological role of bacterial cyanide biosynthesis. Arch Microbiol 173(3):170–177

Bodilis J, Calbrix R, Guérillon J, Mérieau A, Pawlak B, Orange N, Barray S (2004) Phylogenetic relationships between environmental and clinical isolates of *Pseudomonas fluorescens* and related species deduced from 16S rRNA gene and OprF protein sequences. Syst Appl Microbiol 27(1):93–108

Bonfante P, Anca I-A (2009) Plants, mycorrhizal fungi, and bacteria: a network of interactions. Annu Rev Microbiol 63:363–383

Braun PG, Hildebrand PD, Ells TC, Kobayashi DY (2001) Evidence and characterization of a gene cluster required for the production of viscosin, a lipopeptide biosurfactant, by a strain of *Pseudomonas fluorescens*. Can J Microbiol 47(4):294–301

Buddrus-Schiemann K et al (2010) Root colonization by *Pseudomonas* sp. DSMZ 13134 and impact on the indigenous rhizosphere bacterial community of Barley. Microb Ecol 60(2):381–393

Cheng Z, Park E, Glick BR (2007) 1-Aminocyclopropane-1-carboxylate deaminase from *Pseudomonas putida* UW4 facilitates the growth of canola in the presence of salt. Can J Microbiol 53(7):912–918

Coates LR (1910) Fertilizer and method of producing same.U.S. Patent No. 947,796. U.S. Patent and Trademark Office, Washington, DC

Collavino MM, Sansberro PA, Mroginski L a, Aguilar OM (2010) Comparison of in vitro solubilization activity of diverse phosphate-solubilizing bacteria native to acid soil and their ability to promote *Phaseolus vulgaris* growth. Biol Fertil Soils 46(7):727–738

Cook RJ, Bruckart WL, Coulson JR, Goettel MS, Humber RA, Lumsden RD, Maddox JV, Mcmanus ML, Moore L, Meyer SF, Quimby Jr PC, Stack JP, Vaughn JL (1996) Safety of microorganisms intended for pest and plant disease control: a framework for scientific evaluation. Biol Control 7(3):333–351

Dalmastri C, Chiarini L, Cantale C, Bevivino A, Tabacchioni S (1999) Soil type and maize cultivar affect the genetic diversity of maize root-associated *Burkholderia cepacia* populations. Microb Ecol 38:273–284

Date R (2001) Advances in inoculant technology: a brief review. Aust J Exp Agric 41:321–325

De Bruijn I, de Kock MJD, Yang M, de Waard P, van Beek TA, Raaijmakers JM (2007) Genome-based discovery, structure prediction and functional analysis of cyclic lipopeptide antibiotics in *Pseudomonas* species. Mol Microbiol 63(2):417–428

De Bruijn I, de Kock MJD, de Waard P, van Beek TA, Raaijmakers JM (2008) Massetolide a biosynthesis in *Pseudomonas fluorescens*. J Bacteriol 190(8):2777–2789

De La Fuente L, Quagliotto L, Bajsa N, Fabiano E, Altier N (2002) Inoculation with *Pseudomonas fluorescens* biocontrol strains does not affect the symbiosis between rhizobia and forage legumes. Soil Biology and Biochemistry 34:545–548

De La Fuente L, Thomashow L, Weller D, Bajsa N, Quagliotto L, Chernin L, Arias A (2004) *Pseudomonas fluorescens* UP61 isolated from birdsfoot trefoil rhizosphere produces multiple antibiotics and exerts a broad spectrum of biocontrol activity. Eur J Plant Pathol 110(7):671–681

de Werra P, Péchy-Tarr M, Keel C, Maurhofer M (2009) Role of gluconic acid production in the regulation of biocontrol traits of *Pseudomonas fluorescens* CHA0. Appl Environ Microbiol 75(12):4162–4174

Dodd IC, Zinovkina NY, Safronova VI, Belimov AA (2010) Rhizobacterial mediation of plant hormone status. Ann Appl Biol 157(3):361–379

Dorn B, Musa T, Krebs H, Fried PM, Forrer HR (2007) Control of late blight in organic potato production: evaluation of copper-free preparations under field, growth chamber and laboratory conditions. Eur J Plant Pathol 119(2):217–240

Dubeikovsky AN, Mordukhova EA, Kochetkov VV, Boronin AM (1993) Growth promotion of blackcurrant softwood cuttings by recombinant strain *Pseudomonas fluorescens* BSP53a synthesizing an increased amount of indole-3-acetic acid. Soil Biol Biochem 25(9):1277–1281

Duca D, Lorv J, Patten CL, Rose D, Glick BR (2014) Indole-3-acetic acid in plant-microbe interactions. Antonie Van Leeuwenhoek 106(1):85–125

Duijff BJ, Bakker PA, Schippers B (1994) Suppression of fusarium wilt of carnation by *Pseudomonas putida* WCS358 at different levels of disease incidence and iron availability. Biocontrol Sci Tech 4(3):279–288

Duponnois R, Kisa M (2006) The possible role of trehalose in the mycorrhiza helper bacterium effect. Can J Bot 84:1005–1008

Dwivedi D, Johri BN (2003) Antifungals from fluorescent pseudomonads: biosynthesis and regulation. Curr Sci 85(12):1693–1703

Elbadry M, Taha RM, Eldougdoug KA, Gamal-Eldin H (2006) Induction of systemic resistance in faba bean (*Vicia faba* L.) to bean yellow mosaic potyvirus (BYMV) via seed bacterization with plant growth promoting rhizobacteria. J Plant Dis Prot 113(6):247–251

Fernandéz C, Juncosa R (2002) Biopesticidas: ¿la agricultura del futuro? Phytoma 141:14–19

Fernando WGD, Ramarathnam R, Krishnamoorthy AS, Savchuk SC (2005) Identification and use of potential bacterial organic antifungal volatiles in biocontrol. Soil Biol Biochem 37(5):955–964

Fravel DR (2005) Commercialization and implementation of biocontrol. Annu Rev Phytopathol 43:337–359

Frey-Klett P, Garbaye J, Tarkka M (2007) The mycorrhiza helper bacteria revisited. New Phytol 176(1):22–36

Gamalero E, Berta G, Massa N, Glick BR, Lingua G (2008) Synergistic interactions between the ACC deaminase-producing bacterium *Pseudomonas putida* UW4 and the AM fungus *Gigaspora rosea* positively affect cucumber plant growth. FEMS Microbiol Ecol 64(3):459–467

Gao G, Yin D, Chen S, Xia F, Yang J, Li Q, Wang W (2012) Effect of biocontrol agent *Pseudomonas fluorescens* 2P24 on soil fungal community in cucumber rhizosphere using T-RFLP and DGGE. PLoS ONE 7(2):e31806

García de Salamone IE, Hynes RK, Nelson LM (2001) Cytokinin production by plant growth promoting rhizobacteria and selected mutants. Can J Microbiol 47(5):404–411

Gleeson O, O'Gara F, Morrissey JP (2010) The *Pseudomonas fluorescens* secondary metabolite 2,4 diacetylphloroglucinol impairs mitochondrial function in *Saccharomyces cerevisiae*. Antonie Van Leeuwenhoek 97(3):261–273

Glick BR, Cheng Z, Czarny J, Duan J (2007) Promotion of plant growth by ACC deaminase-producing soil bacteria. Eur J Plant Pathol 119(3):329–339

Gross H, Loper JE (2009) Genomics of secondary metabolite production by *Pseudomonas* spp. Nat Prod Rep 26(11):1408–1446

Guyer A, DeVrieze M, Bönisch D, Gloor R, Bodenhausen N, Bailly A, Weisskopf L, Musa T (2015) The anti-*Phytophthora* effect of selected potato-associated *Pseudomonas* strains: from the laboratory to the field. Front Microbiol 6:1–14

Gyaneshwar P, Kumar GN, Parekh LJ, Poole PS (2002) Role of soil microorganisms in improving P nutrition of plants. In: Food security in nutrient-stressed environments: exploiting plants' genetic capabilities. Springer, Dordrecht, pp 133–143

Hashizume H, Nishimura Y (2008) Cyclic lipopeptide antibiotics. Stud Nat Prod Chem 35(C):693–751

Hernández-Montiel LG, Rueda-Puente EO, Cordoba-Matson MV, Holguín-Peña JR, Zulueta-Rodríguez R (2013) Mutualistic interaction of rhizobacteria with arbuscular mycorrhizal fungi and its antagonistic effect on *Fusarium oxysporum* in *Carica papaya* seedlings. Crop Prot 47:61–66

Howell CR, Stipanovic RD (1980) Suppression of *Pythium ultimum* induced damping off of cotton seedlings by *Pseudomonas fluorescens* and its antibiotic pyoluteorin. Phytopathology 70(8):712–715

Johnsson L, Hökeberg M, Gerhardson B (1998) Performance of the *Pseudomonas chlororaphis* biocontrol agent MA 342 against cereal seed-borne diseases in field experiments. Eur J Plant Pathol 104(7):701–711

Jourdan E, Henry G, Duby F, Dommes J, Barthélemy JP, Thonart P, Ongena M (2009) Insights into the defense-related events occurring in plant cells following perception of surfactin-type lipopeptide from *Bacillus subtilis*. Mol Plant-Microbe Interact 22(4):456–468

Kai M, Effmert U, Berg G, Piechulla B (2007) Volatiles of bacterial antagonists inhibit mycelial growth of the plant pathogen *Rhizoctonia solani*. Arch Microbiol 187(5):351–360

Kamilova F, Validov S, Azarova T, Mulders I, Lugtenberg B (2005) Enrichment for enhanced competitive plant root tip colonizers selects for a new class of biocontrol bacteria. Environ Microbiol 7(11):1809–1817

Kamilova F, Kravchenko LV, Shaposhnikov AI, Azarova T, Makarova N, Lugtenberg B (2006) Organic acids, sugars, and L-tryptophane in exudates of vegetables growing on stonewool and their effects on activities of rhizosphere bacteria. Mol Plant-Microbe Interact 19(3):250–256

Kang SM, Radhakrishnan R, Khan AL, Kim MJ, Park JM, Kim BR, Shin D-H, Lee IJ (2014) Gibberellin secreting rhizobacterium, *Pseudomonas putida* H-2- 3 modulates the hormonal and stress physiology of soybean to improve the plant growth under saline and drought conditions. Plant Physiol Biochem 84:115–124

Keel C, Weller DM, Natsch A, Défago G, Cook RJ, Thomashow LS (1996) Conservation of the 2, 4-diacetylphloroglucinol biosynthesis locus among fluorescent *Pseudomonas* strains from diverse geographic locations. Appl Environ Microbiol 62(2):552–563

Khalid A, Arshad M, Zahir ZA (2004) Screening plant growth-promoting rhizobacteria for improving growth and yield of wheat. J Appl Microbiol 96(3):473–480

Klee HJ, Hayford MB, Kretzmer KA, Barry GF, Kishore GM (1991) Control of ethylene synthesis by expression of a bacterial enzyme in transgenic tomato plants. Plant Cell 3(11):1187–1193

Kloepper JW, Leong J, Teintze M, Schrotht MN (1980) *Pseudomonas* siderophores: a mechanism explaining disease-suppressive soils. Curr Microbiol 4:317–320

Kuiper I, Lagendijk EL, Pickford R, Derrick JP, Lamers GEM, Thomas-Oates JE, Lugtenberg BJJ, Bloemberg GV (2004) Characterization of two *Pseudomonas putida* lipopeptide biosurfactants, putisolvin I and II, which inhibit biofilm formation and break down existing biofilms. Mol Microbiol 51(1):97–113

Labbé JL, Weston DJ, Dunkirk N, Pelletier DA, Tuskan GA (2014) Newly identified helper bacteria stimulate ectomycorrhizal formation in *Populus*. Front Plant Sci 5:579

Leeman M, Van Pelt JA, Den Ouden FM, Heinsbroek M, Bakker PAHM, Schippers B (1995) Induction of systemic resistance against Fusarium wilt of radish by lipopolysaccharides of *Pseudomonas fluorescens*. Phytopathology 85(9):1021–1027

Liu L, Kloepper W, Tuzun S (1995) Induction of systemic resistance in cucumber against Fusarium wilt by plant growth-promoting rhizobacteria. Phytopathology 85:695–698

Loper JE, Gross H (2007) Genomic analysis of antifungal metabolite production by *Pseudomonas fluorescens* Pf-5. Eur J Plant Pathol 119(3):265–278

Loper JE, Henkels MD (1999) Utilization of heterologous siderophores enhances levels of iron available to *Pseudomonas putida* in the rhizosphere. Appl Environ Microbiol 65(12):5357–5363

Mark GL, Morrissey JP, Higgins P, O'Gara F (2006) Molecular-based strategies to exploit *Pseudomonas* biocontrol strains for environmental biotechnology applications. FEMS Microbiol Ecol 56(2):167–177

Mathre D, Cook R, Callan N (1999) From discovery to use: traversing the world of commercializing biocontrol agents for plant disease control. Plant Dis 83(11):972–983

Maurhofer M, Hase C, Meuwly J, Métraux P, Défago G (1994) Induction of systemic resistance of tobacco to tobacco necrosis virus by the root-colonizing *Pseudomonas fluorescens* strain CHA 0: influence of the gacA gene and of pyoverdine production. Phytopathology 84(2):139–146

Mavrodi DV, Blankenfeldt W, Thomashow LS (2006) Phenazine compounds in fluorescent *Pseudomonas* spp. biosynthesis and regulation. Annu Rev Phytopathol 44:417–445

Mavrodi DV, Mavrodi OV, Parejko JA, Bonsall RF, Kwak YS, Paulitz TC et al (2012) Accumulation of the antibiotic phenazine-1- carboxylic acid in the rhizosphere of dryland cereals. Appl Environ Microbiol 78:804–812

Mavrodi DV, Parejko JA, Mavrodi OV, Kwak Y-S, Weller DM, Blankenfeldt W, Thomashow LS (2013) Recent insights into the diversity, frequency and ecological roles of phenazines in fluorescent *Pseudomonas* spp. Environ Microbiol 15(3):675–686

Meyer JA, Abdallah MA (1978) The fluorescent pigment of *Pseudomonas fluorescens*: biosynthesis, purification and physicochemical properties. J Gen Microbiol 107(2):319–328

Meziane H, van der Sluis I, van Loon LC, Hofte M, Bakker PAHM (2005) Determinants of *Pseudomonas putida* WCS358 involved in inducing. Mol Plant Pathol 6(2):177–185

Miethke M, Marahiel MA (2007) Siderophore-based iron acquisition and pathogen control. Microbiol Mol Biol Rev 71(3):413–451

Mohr TJ, Liu H, Yan S, Morris CE, Castillo JA, Jelenska J, Vinatzer BA (2008) Naturally occurring nonpathogenic isolates of the plant pathogen *Pseudomonas syringae* lack a type III secretion system and effector gene orthologues. J Bacteriol 190(8):2858–2870

Mootz HD, Schwarzer D, Marahiel MA (2002) Ways of assembling complex natural products on modular nonribosomal peptide synthetases. Chembiochem 3(6):490–504

Morel MA, Castro-Sowinski S (2013) The complex molecular signaling network in microbe–plant interaction. In: Arora NK (ed) Plant microbe symbiosis: fundamentals and advances. Springer, New Delhi, pp 169–199

Morrone D, Chambers J, Lowry L, Kim G, Anterola A, Bender K, Peters RJ (2009) Gibberellin biosynthesis in bacteria: separate ent-copalyl diphosphate and ent-kaurene synthases in *Bradyrhizobium japonicum*. FEBS Lett 583(2):475–480

Naiman AD, Latrónico A, García de Salamone IE (2009) Inoculation of wheat with *Azospirillum brasilense* and *Pseudomonas fluorescens*: impact on the production and culturable rhizosphere microflora. Eur J Soil Biol 45(1):44–51

Nakkeeran S, Fernando WGD, Siddiqui ZA (2006) Plant growth promoting rhizobacteria formulations and its scope in commercialization for the management of pests and diseases. In: PGPR: biocontrol and biofertilization. Springer, Dordrecht, pp 257–296

Nielsen TH, Nybroe O, Koch B, Hansen M (2005) Genes involved in cyclic lipopeptide production are important for seed and straw colonization by *Pseudomonas* sp. strain DSS73. Am Soc Microbiol 71(7):4112–4116

Nowak-Thompson B, Chaney N, Wing JS, Gould SJ, Lo- per JE (1999) Characterization of the pyoluteorin biosynthetic gene cluster of *Pseudomonas fluorescens* Pf-5. J Bacteriol 181:2166–2174

Ongena M, Jacques P (2008) *Bacillus* lipopeptides: versatile weapons for plant disease biocontrol. Trends Microbiol 16(3):115–125

Peleg Z, Blumwald E (2011) Hormone balance and abiotic stress tolerance in crop plants. Curr Opin Plant Biol 14(3):290–295

Penrose DM, Moffatt BA, 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. Can J Microbiol 47(1):77–80

Pliego C, de Weert S, Lamers G, de Vicente A, Bloemberg G, Cazorla FM, Ramos C (2008) Two similar enhanced root-colonizing *Pseudomonas* strains differ largely in their colonization strategies of avocado roots and *Rosellinia necatrix* hyphae. Environ Microbiol 10(12):3295–3304

Pliego C, Kamilova F, Lugtenberg B (2011) In: Maheshwari DK (ed) Bacteria in agrobiology: crop ecosystems. Springer, Berlin/Heidelberg, pp 295–343

Quagliotto L, Azziz G, Bajsa N, Vaz P, Pérez C, Ducamp F, Cadenazzi M, Altier N, Arias A (2009) Three native *Pseudomonas fluorescens* strains tested under growth chamber and field conditions as biocontrol agents against damping-off in alfalfa. Biol Control 51(1):42–50

Raaijmakers JM, Weller DM (1998) Natural plant protection by 2, 4-Diacetylphloroglucinol – producing *Pseudomonas* spp. in take-all decline soils. Mol Plant Microbe Interact 11(2):144–152

Raaijmakers JM, Leeman M, van Oorschot MMP, van der Sluis I, Schippers B, Bakker P (1995) Dose-response relationships in biological control of *Fusarium* wilt of radish by *Pseudomonas* spp. Biol Control 85(10):1075–1081

Raaijmakers JM, de Bruijn I, de Kock MJD (2006) Cyclic lipopeptide production by plant-associated *Pseudomonas* spp.: diversity, activity, biosynthesis, and regulation. Mol Plant-Microbe Interact 19(7):699–710

Raaijmakers JM, De Bruijn I, Nybroe O, Ongena M (2010) Natural functions of lipopeptides from *Bacillus* and *Pseudomonas*: more than surfactants and antibiotics. FEMS Microbiol Rev 34(6):1037–1062

Remans R, Croonenborghs A, Gutierrez RT, Michiels J, Vanderleyden J (2007) Effects of plant growth-promoting rhizobacteria on nodulation of *Phaseolus vulgaris* L. are dependent on plant P nutrition. Eur J Plant Pathol 119:341–351

Remans R, Beebe S, Blair M, Manrique G, Tovar E, Rao I, Croonenborghs A, Torres-Gutierrez R, El-Howeity M, Michiels J, Vanderleyden J (2008) Physiological and genetic analysis of root responsiveness to auxin-producing plant growth-promoting bacteria in common bean (*Phaseolus vulgaris* L.). Plant Soil 302(1–2):149–161

Richardson AE, Simpson RJ (2011) Soil microorganisms mediating phosphorus availability update on microbial phosphorus. Plant Physiol 156(3):989–996

Riedlinger J, Schrey SD, Tarkka MT, Hampp R, Kapur M, Fiedler HP (2006) Auxofuran, a novel metabolite that stimulates the growth of fly agaric, is produced by the mycorrhiza helper bacterium *Streptomyces* strain AcH 505. Appl Environ Microbiol 72(5):3550–3557

Roongsawang N, Hase K, Haruki M, Imanaka T, Morikawa M, Kanaya S (2003) Cloning and characterization of the gene cluster encoding arthrofactin synthetase from *Pseudomonas* sp. MIS38. Chem Biol 10:869–880

Ryu C, Farag MA, Hu C, Reddy MS, Kloepper JW, Pare PW (2004) Bacterial volatiles induce systemic resistance in *Arabidopsis*. Plant Physiol 134:1017–1026

Saharan BS, Nehra V (2011) Plant growth promoting rhizobacteria: a critical review. Life Sci Med Res 21:1–30

Saravanakumar D, Samiyappan R (2007) ACC deaminase from *Pseudomonas fluorescens* mediated saline resistance in groundnut (*Arachis hypogea*) plants. J Appl Microbiol 102(5):1283–1292

Setten L, Soto G, Mozzicafreddo M, Fox AR, Lisi C, Cuccioloni M, Angeletti M, Pagano E, Díaz-Paleo A, Ayub ND (2013) Engineering *Pseudomonas protegens* Pf-5 for nitrogen fixation and its application to improve plant growth under nitrogen-deficient conditions. PLoS ONE 8(5):e63666

Shang C, Caldwell DE, Huang PM (1996) Bioavailability of organic and inorganic phosphates adsorbed on short-range ordered aluminum precipitate. Microb Ecol 31(1):29–39

Tilak KVBR, Ranganayaki N, Manoharachari C (2006) Synergistic effects of plant-growth promoting rhizobacteria and *Rhizobium* on nodulation and nitrogen fixation by pigeon pea (*Cajanus cajan*). Eur J Soil Sci 57(1):67–71

Tinivella F, Hirata LM, Celan MA, Wright SAI, Amein T, Schmitt A, Koch E, van der Wolf JM, Groot SPC, Stephan D, Garibaldi A, Gullino ML (2009) Control of seed-borne pathogens on legumes by microbial and other alternative seed treatments. Eur J Plant Pathol 123(2):139–151

Tran H, Ficke A, Asiimwe T, Raaijmakers JM (2007) Role of the cyclic lipopeptide massetolide A in biological control of *Phytophthora infestans* and in colonization of tomato plants by *Pseudomonas fluorescens*. New Phytol 175:731–742

Trivedi P, Pandey A, Palni LMS (2008) In vitro evaluation of antagonistic properties of *Pseudomonas corrugata*. Microbiol Res 163(3):329–336

Troppens DM, Dmitriev RI, Papkovsky DB, O'Gara F, Morrissey JP (2013) Genome-wide investigation of cellular targets and mode of action of the antifungal bacterial metabolite 2,4-diacetylphloroglucinol in *Saccharomyces cerevisiae*. FEMS Yeast Res 13(3):322–334

Uroz S, Calvaruso C, Turpault MP, Pierrat JC, Mustin C, Frey-Klett P (2007) Effect of the mycorrhizosphere on the genotypic and metabolic diversity of the bacterial communities involved in mineral weathering in a forest soil. Appl Environ Microbiol 73(9):3019–3027

van de Mortel JE, Tran H, Govers F, Raaijmakers JM (2009) Cellular responses of the late blight pathogen *Phytophthora infestans* to cyclic lipopeptide surfactants and their dependence on G proteins. Appl Environ Microbiol 75(15):4950–4957

Velivelli SS, Sessitsch A, Prestwich B (2015) The role of microbial inoculants in integrated crop management systems. Potato Res 57(3–4):291–309

Viebahn M, Wernars K, Smit E, van Loon LC, DeSantis TZ, Andersen GL, Bakker PAHM (2006) Microbial diversity in wheat rhizosphere as affected by genetically modified *Pseudomonas putida* WCS358r. IOBC WPRS Bull 29(2):167–172

Vijay Krishna Kumar K, Krishnam Raju S, Reddy MS, Kloepper JW, Lawrence KS, Groth DE, Miller ME, Sudini H, Binghai Du (2009) Evaluation of commercially available PGPR for control of rice sheath blight caused by *Rhizoctonia solani*. J Pure Appl Microbiol 3(2):485–488

Voisard C, Keel C, Haas D, Dèfago G (1989) Cyanide production by *Pseudomonas fluorescens* helps suppress black root rot of tobacco under gnotobiotic conditions. EMBO J 8(2):351–358

Weisbeek PJ, Gerrits H (1999) Iron and biocontrol. In: Stacey G, Keen NT (eds) Plant-microbe interactions. APS Press, St. Paul, pp 4217–4250

Weisskopf L (2013) The potential of bacterial volatiles for crop protection against phytopathogenic fungi. In: Méndez-Vilas A (ed) Microbial pathogens and strategies for combating them: science, technology and education. Formatex Research Center, Badajoz, pp 1352–1363

Weller DM (2007) *Pseudomonas* biocontrol agents of soilborne pathogens: looking back over 30 years. Phytopathology 97(2):250–256

Weller DM, Mavrodi DV, Van Pelt JA, Pieterse CMJ, Van Loon LC, Bakker PAHM (2012) Induced systemic resistance in *Arabidopsis thaliana* against *Pseudomonas syringae* pv. tomato by 2,4-diacetylphloroglucinol-producing *Pseudomonas fluorescens*. Biol Control 102(4):403–412

Whipps JM (2001) Microbial interactions and biocontrol in the rhizosphere. J Exp Bot 52(Spec Issue):487–511

Woo SL, Ruocco M, Vinale F, Nigro M, Marra R, Lombardi N, Pascale A,Lanzuise S, Manganiello G, Lorito M (2014) *Trichoderma*-based products and their widespread use in agriculture. Open Mycol J 8(M4):71–126

Yan Y, Yang J, Dou Y, Chen M, Ping S, Peng J, Lu W, Zhang W, Yao Z, Li H, Liu W, He S, Geng L, Zhang X, Yang F, Yu H, Zhan Y, Li D, Lin Z, Wang Y, Elmerich C, Lin M, Jin Q (2008) Nitrogen fixation island and rhizosphere competence traits in the genome of root-associated *Pseudomonas stutzeri* A1501. Proc Natl Acad Sci U S A 105(21):7564–7569

Yanes ML, De La Fuente L, Altier N, Arias A (2012) Characterization of native fluorescent *Pseudomonas* isolates associated with alfalfa roots in Uruguayan agroecosystems. Biol Control 63:287–295

Zahir ZA, Zafar-ul-Hye M, Sajjad S, Naveed M (2011) Comparative effectiveness of *Pseudomonas* and *Serratia* sp. containing ACC-deaminase for coinoculation with *Rhizobium leguminosarum* to improve growth, nodulation, and yield of lentil. Biol Fertil Soils 47(4):457–465

Zeller W (2006) Status of biocontrol methods against fire blight. Phytopathol Pol 39:71–78