Chapter 6 Role of Phosphate-Solubilizing Actinomycetes in Plant Growth Promotion: Current Perspective

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Abstract Phosphorus (P), an essential plant nutrient, is a nonrenewable resource whose availability depends exclusively on mined rock phosphates. Deficiency of P in soil results in reduction in food production since all plants require an adequate supply of P for its growth and development. Even though synthetic phosphatic fertilizer has played some major roles in enhancing crop production, its excessive use has also dark sides to it where it has been found to damage the environment, destruct soil fertility, and, via food chain, seriously affect the human health. Considering the nuisance of overuse of P, there is an urgent demand by the agriculture practitioners to find nonhazardous strategy that can overcome/reduce the use of agrochemicals in agricultural practices and, hence, may preserve the very integrity of soil ecosystems. In this context, actinobacteria, a group of Grampositive bacteria, ubiquitous in soils, are likely to play some important roles in supplying soluble P to plants by solubilizing/mineralizing complex P resources of soils. Additionally, the extracellular metabolites produced by actinomycetes may inhibit phytopathogens and, sometimes such metabolic compounds may also act as plant growth regulators. These qualities, among others, make actinobacteria an ideal candidate for developing as microbial inoculants for ultimate use in agriculture production system. The potential roles of actinomycetes as phosphate solubilizers in enhancing crop production are discussed.

Keywords Actinomycetes • P solubilizers • Plant growth • Biocontrol

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6.1 Introduction

Phosphorus, identified as an essential element for all life forms (Smil 2000), is the second most important plant macronutrient after N (Donahue et al. 1990). Phosphorus accounts for the biomass buildup, the energy transfer, signal transduction, macromolecular biosynthesis, photosynthesis, and respiration chain reactions (Vance et al. 2003; Fernández et al. 2007). Sadly, this highly vital element is one of the least accessible (only 0.1 % of the total soil P reserve) nutrients to the plants (Zou et al. 1992; Takahashi and Anwar 2007). Owing to the suboptimal levels of P, it is often considered a limiting factor to primary production across a diverse range of ecosystem (Elser et al. 2007) and may cause a 5-15 % loss in plant yields (Hinsinger 2001). The low availability of P in soil is primarily due to its highly reactive nature as it readily forms insoluble complexes with the soil ions. Thus, to fulfill the growing P demands for food production and to maintain a balanced fertility in soils and, hence, to achieve a low-input, sustainable eco-friendly agriculture targets, a better management of the soil P reserve is urgently needed. Many strategies have been introduced by the scientists across different regions where the role of phosphate-solubilizing microorganisms involving bacteria (Zaidi et al. 2009; Ahmad et al. 2013), fungi (Khan et al. 2010), and actinomycetes (Gangwar et al. 2012) in increasing plant yields and soil quality via P supply has been recognized. Among these, actinomycetes as P solubilizers (de Vasconcellos et al. 2010; Hamdali et al. 2012) have been less explored despite showing a better genetic and greater biodiversity potential (Pathom-Aree et al. 2006; Thangapandian et al. 2007). Also, they represent heterogeneous and abundant microbial populations and hugely affect cycling of nutrient in soil ecosystems (Elliot and Lynch 1995; Figueiredo et al. 2010). Recently, the role of actinomycetes in sustainable agriculture (Johansson et al. 2004; Strap 2011) via P supply to plants has been identified. Some other actinomycetes, for example, endophytic actinomycete (Araújo et al. 2000; Kunoh 2002; Lee et al. 2008; Qin et al. 2008), which fix atmospheric N into NH_3 and export the fixed N to the host plants, have also shown PS activity (Gangwar et al. 2012). In a study, almost 44 % of the endophytic actinomycetes isolated from rice had PS activity, among which S. lavendulae R22 solubilized the maximum (26.5 mg/100 ml) amount of P, while Micromonospora R19 isolate could solubilize the minimum amount of P. Realizing the plant-growth-promoting potentials of actinomycetes which involve one or simultaneous mechanisms, here, the recent advances in P solubilization by actinomycetes and its impact on crop production are highlighted.

6.2 Phosphorus Status and P Dynamics in Rhizosphere: Current Perspective

Globally, rock phosphate (RP) is an abundantly available and inexpensive form of P, and therefore, about 80 % of mined RP is used for agricultural fertilizer (Tirado and Allsopp 2012). Worldwide, there is a huge RP deposit in countries such as Morocco and China, while other regions particularly Asian countries depend heavily on import of phosphates for intensive agriculture production systems. From a consumption point of view, China is the largest consumer of P fertilizers accounting for about 34 % of the total world consumption, whereas India ranks second with 19 % of global consumption (FAOSTAT 2012). Similarly, the uptake of P by plants varies greatly (Fig. 6.1). Regrettably, mining of RP and its use as fertilizers pose a serious human health risk (Zhang and Shan 2008; Pan et al. 2010; Tirado and Allsopp 2012). Considering the declining P resources, cost of P-fertilizer production, and direct or indirect human health problems due to their excessive application, scientists around the world have directed their attention toward exploring the natural and inexpensive P resources which could serve as an eco-friendly and economical alternative for chemical fertilizers in sustainable agriculture. In this context, the phosphate-solubilizing actinomycetes have provided some solutions to the expensive P problems.

Further, the chemical and biological processes in the rhizosphere play an important role in soil nutrient availability and crop productivity (Zhang et al. 2010) as presented in Fig. 6.2. Plants acquire P in the form of orthophosphate anions (mainly $H_2PO_4^{-}$ and $H_2PO_4^{-2}$) which they obtain from different soil P reserves including inorganic P (Pi) and organic P (Po). Broadly, on the basis of plant accessibility, soil P has been grouped as follows: (a) solution P, present in the soil solution and immediately available for plant uptake; (b) active P, adsorbed on active sites of soil yet readily available and in equilibrium with solution P; and (c) fixed P, strongly adsorbed and least soluble with slow conversions to active P over a vast period, even years (Syers et al. 2008; Shen et al. 2011). Generally, concentration of plant-available P in soil is below the critical level required for plant growth and yields (Raghothama 1999). Thus, the synthetic phosphatic fertilizer or organic manure is applied to overcome the P deficiency to plants. Of the total P applied to soils, only 15–25 % of it is available for uptake by plants and the remainder is fixed (Shen et al. 2011). The P dynamics and availability in soil is, therefore, controlled by several factors: (1) the ability of plants to form extensive root systems, (2) impact of microbial colonization onto the development of plants, (3) soil microflora affecting biogeochemical cycling of elements, and (4) physicochemical properties of soils supporting plants and microbial life in soils.

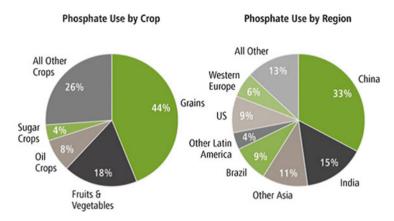


Fig. 6.1 Crop-wise use of phosphorus and its distribution in different countries (Source: CRU, Fertecon, IFA (Potash Corp. 2013)

6.3 Actinomycetes: An Ideal Candidate for Sustainable Crop Production

Actinomycetes are a ubiquitous, saprophytic, and heterogeneous group of microorganisms belonging to an extensive and diverse group of Gram-positive, aerobic, mycelial bacteria that have DNA with high %G+C content (51–73 %) with the exception of freshwater actinobacteria which has low GC content (Takisawa et al. 1993; Ghai et al. 2012). Actinomycetes inhabit both the rhizoplane and rhizosphere (Solans and Vobis 2003; Frioni 2006) and are known to play some important ecological roles in soil nutrient cycling (Elliot and Lynch 1995), probiotics (Lievin et al. 2000), bioremediation (Amoroso et al. 2013), nitrogen fixation, and deterioration and decomposition of plant and animal residues and also provide many bioactive compounds such as vitamins, enzymes, and antibiotics (de Boer et al. 2005; Busti et al. 2006; Prapagdee et al. 2008; Genilloud et al. 2011). Apart from their beneficial impact, actinomycetes also cause some harmful effects such as they cause diseases and spoil different useful materials (Waksman 1950). Other properties which make them an ideal candidate for developing inoculants are as follows: (1) their high genetic and metabolic versatility, (2) they can easily be genetically engineered (Pogell et al. 1991), (3) high growth rate and relatively faster colonization efficiency (Hsu and Lockwood 1975), and (4) ability to tolerate high salt concentration (Vassilev et al. 2012). Despite possessing such qualities, actinomycetes in general have been less explored organisms in agricultural technology, among variously distributed soil microflora (Qin et al. 2011).

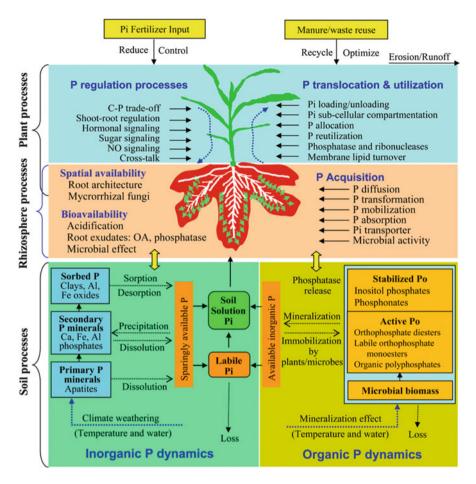


Fig. 6.2 Phosphorus dynamics in soil [Adapted from Shen et al. (2011)]

6.3.1 Isolation and Identification of Actinomycetes

Phosphate-solubilizing actinomycetes have been isolated from diverse environment (Matsumoto et al. 1998; Rai et al. 2007; Lee et al. 2008) such as soil (Xu et al. 1996; Debananda et al. 2009; Salcedo et al. 2014), rhizospheres (Norovsuren et al. 2007), phyllosphere (Gupta et al. 2010), and salt and alkaline environments (Jiang et al. 2005) (Table 6.1). The bioprospecting of actinomycetes from the usual or routine habitat is likely to result in rediscovery or the recovery of the same strain with the similar mode of actions (Jensen et al. 2005).

Actinomycetes represent one of the largest domains of bacteria involving 145 genera and more than 4,000 species (Berdys 2005). Their population has been reported highest at the soil surface which, however, decreases with increasing soil depth (Takahashi and Omura 2003). Prior to isolation, soil is pretreated in order

Habitat	Actinomycete genus isolated
Cultivated field	Streptomyces
Cultivated paddy field	Micromonospora
Plant matter	Microbispora
Pasture	Micromonospora
Lake sediment	Micromonospora
Mangrove rhizosphere soil	Actinobacteria
Tea field soil	Acidophilic and acid tolerant actinomycetes
Desert soil, marine sediment, seawater, and activated sludge	
Pretreatment	
Yeast extract (6 %, 20 min)	Enrichment of actinomycetes
Heating (100 °C, 1 h)	Streptosporangium, Microbispora, Microtetraspora
Phenol (1.0-1.5 %, 30 min)	Dactylosporangium, Microbispora, Microtetraspora
Pre-culture with CaCO ₃	Enrichment of actinomycetes
Chemotactic method	Motile actinomycetes, actinomycetes,
(KCl, v-collidin, xylose)	Dactylosporangium
Medium for isolation	
Addition of antibiotics	
Novobiocin	Actinoplanes, Kitasatospora
Tunicamycin	Micromonospora
Rifampicin	Actinomadura
Chlortetracycline	Nocardia
Macrolide or aminoglycoside	Macrolide or aminoglycoside producer
Addition of humic acid	Rare actinomycetes
Addition of proline	Enrichment of actinomycetes
Gellan gum (substitute for agar)	Actinobispora
Conditions for isolation	
High temperature	Thermophilic actinomycetes
Adapted from Takahashi and Omura (2	003)

Table 6.1 Different habitat and varying isolation methods for actinomycetes

Adapted from Takahashi and Omura (2003)

to avoid the emergence of bacterial and fungal contaminants. For this, soil samples are collected bulked, mixed, and allowed to dry heating (at 45 °C for 2 h or 50 °C for 10 min or 60 °C for 30 min) (Goodfellow 1971). The isolation medium is also suspended with antibacterial (penicillin 25 mg/ml) or antifungal (nystatin 0.1 % or cycloheximide 50 mg/ml) agents (Balagurunathan and Radhakrishnan 2007). A 0.1 ml of serially diluted (Nonomura and Ohara 1969) soil sample is spread plated onto different actinomycete isolation media, such as casein starch agar, the Czapek agar, and the oatmeal agar, and incubated at 28 °C for 15–30 days. The resulting colonies are then picked and identified using cultural, morphological (Pridham and Tresner 1974; Nonomura 1989; Sabaou et al. 1998), and physiological (Nonomura and Ohara 1969; Goodfellow 1971) characteristics. The actinomycetes are then identified to species level using fatty acid analysis, mol (%) G+C contents, DNA–DNA hybridization, and 16S rRNA sequencing.

6.3.2 In Vitro Screening of Phosphate-Solubilizing Actinomycetes

The phosphate-solubilizing activity of the actinomycetal strains is assessed generally on the Pikovskaya (PVK) medium (Pikovskaya 1948) which contains (g/l): glucose 10; $Ca_3(PO_4)_2$, 5; $(NH_4)_2SO_4$, 0.5; NaCl, 0.2; MgSO_4·7H_2O, 0.1; KCl, 0.2; yeast extract, 0.5; MnSO_4·H_2O, 0.002; and FeSO_4·7H_2O, 0.002, agar 15. The streaked/spot inoculated plates are incubated at 22 °C for 14 days and observed for halo formation. The development of a clear zone around the colony on the culture plates is taken as an index of P solubilization. Halo size is calculated by subtracting colony diameter from the total diameter (Valverde et al. 2006). Thereafter, the halo-forming actinobacteria is grown on SRSM-1 medium which has the same composition as that of the PVK medium but is supplemented with bromocresol purple as a pH indicator for determining the involvement of organic acid production in PS activity.

6.3.3 Role of Actinobacteria in Rock Phosphate Solubilization

Nearly 20 % of the actinomycetes including Streptomyces and Micromonospora possess profound phosphate-solubilizing potential (Barreto et al. 2008; El-Tarabily et al. 2008; Hamdali et al. 2008b). Actinomycetes as P solubilizer have received greater attention for two reasons: (1) ability to produce agro-active metabolites, for example, phytohormones, siderophores, and antibiotics, etc. (Hoster et al. 2005; Errakhi et al. 2007; Errakhi et al. 2009), and (2) ability to tolerate different stressor molecules (Fabre et al. 1988; Keiser et al. 2000; Hamdali et al. 2008d). Also, the inoculation with actinomycetal strains has been found to increase the plant growth (Hamdali et al. 2008c). Plants take up the orthophosphorus form of P as nutrient, the availability of which depends on soil characteristics (Nath and Borah 1983). However, Fe and Al at high pH and Ca at low pH fix the soluble form into insoluble form rendering it unavailable to the plants (Rengel and Marschner 2005; Johnson and Loepper 2006). The PS actinomycetes, however, through the release of organic acid (acidification) (Hoberg et al. 2005; Abdulla 2009) and by other mechanisms such as chelation, exchange reactions, and polymeric substances formation (Delvasto et al. 2006) convert the insoluble forms of P into soluble forms. The organic anions assist P solubilization mainly in two ways: (a) lowering pH and (b) ligand exchange reaction (Beunemann et al. 2011; Balemi and Negisho 2012). However, the organic anions in some cases have been found not to acidify the medium, probably because once they are released in soil, they already exist in the dissociated form due to their low acid dissociation constants (pKa). In fact, it is assisted by the proton extrusion accompanying respiration and NH⁴⁺ assimilation (Illmer and Schinner 1992), which compensates for the losses of negative charge and hence lowers the pH values. Organic anions are also involved in ligand exchange or ligand-enhanced desorption and/or chelation of Fe and Al oxides and Ca phosphates, thereby decreasing the positive surface potential on the metal oxide (Filius et al. 1991) and releasing the inaccessible P from the soil matrix (Raghothama and Karthikeyan 2005). The extent of solubilization by actinomycetes however depends on (1) the source of inorganic P (Pi), (2) the intrinsic PS ability of actinomycetal species, and (3) the types and composition of photosynthates available in the rhizospheres (Banik and Dey 1982). Interestingly, both solubilization (acidification of inorganic P) and mineralization (enzymatic dissolution of organic P) may coexist in the same PS actinomycetal strains (Tao et al. 2008).

Recently, numerous rhizoactinomycetes capable of solubilizing insoluble P have been identified (Franco-Correa et al. 2010; Pragya et al. 2012). For example, apart from soil actinomycetes, many marine actinomycetes have also shown the production of phosphatase which consequently enhanced the P availability (Sahu et al. 2007). In a similar study, Franco-Correa et al. (2010) reported that 20 % of the actinomycetes including *Streptomyces* and *Thermobifida*, isolated from the rhizosphere of Trifolium repens, grown in the fields of Sabana de Bogotá, Colombia, had quite active P-solubilizing activity. Even though all isolates produced acid phosphatase wherein 43 % isolates demonstrated alkaline phosphatase, which indicated that all the test actinomycete strains could mineralize the organic P sources (Richardson et al. 2009). Among phosphatases, acid phosphatases, a group of enzymes produced by plants/microbes in response to P stress, catalyze hydrolysis of phosphoric esters in a range of organic P complexes, thereby enhancing plant P uptake (Tarafdar and Claassen 1988; Tarafdar and Claassen 2001; Richardson et al. 2011) from unavailable P resources. There are also some other enzymes secreted by actinomycetes that take part in the dissolution process. As an example, Reza-Ghorbani-Nasrabadi et al. (2012) assessed 97 soil samples collected from different soil ecosystems and showed for the first time that actinomycetes had extracellular phytate-degrading activity. Phytases are a group of enzyme responsible for stepwise dephosphorylation of phytate, the most abundant inositol phosphate in nature. Of the total actinomycetal cultures, 46.3 % showed extracellular phytate-degrading activity in liquid culture medium supplemented with 4 g/l Na-phytate as sole P source. The two more closely studied phytase producers were identified according to 16SrRNA sequencing as Streptomyces (sp. isolate No. 43 showed 98 % similarity to Streptomyces alboniger and S. venezuelae, while isolate No. 63 exhibited 98 % sequence relatedness with S. ambofaciens and S. lienomycini).

6.3.4 Actinomycetes as a Potential Candidate for Increased Plant Growth and Yield

The agronomic use of PGPR at field scale in general has been hampered by poor understanding of mechanisms that facilitate plant growth, inability of bacterial strains to establish in various soils, rhizosphere incompetence, and changing environmental conditions. On the contrary, the actinobacteria with their ability to (1) sporulate, (2) transform various complex soil nutrients into simple and accessible forms, (3) extensively and efficiently colonize plant roots, (4) manage phytopathogens, and (5) secrete other plant-growth-promoting substances make these organisms as preferred choices for developing potential field bio-inoculants. Actinomycetes are metabolically flexible soil/rhizosphere-colonizing microorganisms (Miller et al. 1990; Euanorasetr et al. 2010; Lei et al. 2013) capable of producing a range of compounds of interest, including (1) antifungal compounds which have been found useful in controlling fungal root diseases (Rothrock and Gottlieb 1984; Zucchi et al. 2010; Bungonsiri et al. 2011; Sreevidya and Gopalakrishnan 2012; Francisco et al. 2013) (2) siderophores (Lee et al. 2012; Nakouti et al. 2012; Najwa et al. 2013), ACC deaminase (El-Tarabily 2008), and (3) plant-growth-promoting hormones (Hamdali et al. 2008a; Khamna et al. 2010). Generally, plant root exudates stimulate growth, proliferation, and rhizosphere colonization of actinomycetes that may act as a strong antagonist to fungal pathogens. The root exudates are utilized as a source of carbon and energy by the actinomycetes and, concomitantly, enhance the synthesis of antimicrobial substances (Crawford et al. 1993; Yuan and Crawford 1995). In addition, actinomycetes synthesize an array of biodegradative enzymes which includes chitinases (Blaak et al. 1993; Gupta et al. 1995; Pattanapipitpaisal and Kamlandharn 2012; Sowmya et al. 2012), glucanases (Hopwood 1990; Damude et al. 1993; Mahadevan and Crawford 1996; Harchand and Singh 1997; Thomas and Crawford 1998; Trejo-Estrada et al. 1998; Fayad et al. 2001; Huiling et al. 2014), peroxidases (Ramachandra et al. 1988; Djamila et al. 2011), and other enzymes possibly involved in mycoparasitic activity. Considering the potential role of actinomycetes in the management of plant diseases and plant growth promotion by certain other mechanisms, actinomycetes in recent times are considered as one of the important aspects in sustainable plant production (Palanivandi et al. 2013) as presented in Table 6.2.

The root-colonizing soil actinomycetes *S. lydicus* WYEC108, for instance, have been reported to influence pea root nodulation and increase the nodulation frequency possibly at the level of infection by *Rhizobium* spp. Following colonization, *S. lydicus* sporulate inside the surface cell layers of the nodules which in turn led to a massive increase in the nodules size. Subsequently, the forms and vigor of bacteroids were greatly improved due to enhanced assimilation of iron and possibly other soil nutrients within nodules. Moreover, bacteroid accumulation of the C storage polymer, poly- β -hydroxybutyrate (PHB), was reduced in colonized nodules (Solans 2007). The co-inoculation of rhizoactinomycetes *Streptomyces* MM40,

Table 6.2 Plant-growth-promoting traits exhibited by phosphate-solubilizing actinobacteria	oting traits exhibited by phosp	ohate-solubilizi	ng actinobacteria		
Actinomycetes	Source of isolation	Test plant	Test condition	Plant-growth-promoting traits	References
Micrococcus sp. NII-0909	Western ghat forest soil in India	Cow pea	In vitro	Auxin production, 1-aminocyclopropane-1- carboxylate deaminase activity, and siderophore production	Dastager et al. (2010)
Microbacterium azadirachtae sp. nov. AI-S262 ^T	Rhizoplane of neem seedlings	I	Plate assay	IAA production, P solubiliza- tion, ACC deaminase activity, and sulfur oxidation	Madhaiyan et al. (2010)
Streptomyces rochei IDWR19, Wheat rhizosphere Streptomyces carpinensis IDWR53, Streptomyces thermolilacinus IDWR81	Wheat rhizosphere	I	Solid-state fermentation	Soil enzyme production (invertase, cellulase alka- line protease, phytase chitinase)	Jog et al. (2012)
Streptomyces rochei IDWR19, Wheat rhizosphere Streptomyces thermolilacinus IDWR81	Wheat rhizosphere	Wheat	Plant growth experiment	Increase in biomass of 1×8 - and 2×3 -fold; increase of shoot length of plants	
Microbacterium sp. F10a	Oil-polluted soil	Wheat	Pot experiment	Significantly increased growth of wheat indoleacetic acid, siderophore, and 1-aminocyclopropane-1- carboxylate deaminase activity and solubilizing inorganic phosphate poly- cyclic aromatic hydrocar- bon degrading (phenanthrene and pyrene removal)	Sheng et al. (2009)
Streptomyces, Microbacterium	Inner tissues of a traditional Chinese folk medicine	I	In vitro		Qin et al. (2014)

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	Hamdali et al. (2008c)	Gopalakrishnan et al. (2013)		Gopalakrishnan et al. (2011)	Vasconcellos et al. (2010) (continued)
Indole-3-acetic acid (IAA), N ₂ -fixation, ACC deaminase	Stimulated aerial growth in more than 70 % of test tube and more than 30 % of RP soil as compared to non-inoculated control treatment	Enhanced plant height, leaf area, stem weight, leaf weight, root length, root surface area, root volume, and root dry weight over the control	Enhanced plant height, tillers, primary and secondary panicle number, panicle length, stover and grain yield, total dry matter, and test seed weight over the control. Root length, root volume, and root dry weight significantly enhanced in inoculated plots over the control	Biocontrol traits against Fusarium wilt	Indoleacetic acid and chitinases
	Test tube and rock phos- phate soil experiment	Greenhouse conditions	Field conditions		Plate assay
	Wheat (Triticum durum L.)	Sorghum	Rice	Chickpea	1
Limonium sinense (Girard) Kuntze	Benguerir phosphate mine, Morocco	Herbal vermicomposts			Rhizosphere of Araucaria angustifolia
	Streptomyces griseus related strain BH7	Streptomyces tsusimaensis, Streptomyces caviscabies, Streptomyces setonii, Streptomyces africanus, and an identified species of Streptomyces			Actinomycetes strains

Table 6.2 (continued)					
Actinomycetes	Source of isolation	Test plant	Test condition	Plant-growth-promoting traits	References
Streptomyces MCR10, Thermobifida MCR24, Nocardia MCR32, and other unidentified actino- mycete strains	Rhizosphere of Trifolium repens L.	Clover plants (<i>T. repens</i> L.)	Clover plants Pot experiment (<i>T. repens</i> L.)	Siderophore production but few unidentified strains also showed growth in N-free media suggesting that they could be nitrogen- fixing bacteria	Franco-Correa et al. (2010)
Streptomyces MCR9, Thermobifida MCR24, and Streptomyces MCR 26	Rhizosphere of <i>Trifolium</i> repens L.	Clover plants (<i>T. repens</i> L.)	Clover plants Co-inoculation with AM (<i>T. repens</i> fungi <i>Glomus mosseae</i> L.) without host; in vitro and in soil	Stimulated mycelial develop- ment from <i>G. mosseae</i> spores; MCR9 and MCR26 also showed stimulated germination of AM spores	Franco-Correa et al. (2010)
			Pot experiment with host; co-inoculated with AM fungi Glomus mosseae	Significant plant growth (par- ticularly shoot biomass at the end of the assay) Increased N and P acquisition by plants due to synergic effect Increased the total mycorrhizal root length of <i>Glomus</i> inoculated plants	Franco-Correa et al. (2010)
Streptomyces spp., Saccharo- polyspora spp., Actinopolyspora spp., Nocardia spp.	Tissue pieces of leaves, stems, and root of rice (Oryza sativa)	1	Plate assays	IAA and siderophore produc- tion, antifungal activity against Aspergillus niger, Alternaria brassicicola, Chaetomium globosum, Fusarium oxysporum, Phytophthora dresclea, Rhizoctonia solani, Botry- tis cinerea	Gangwar et al. (2012)

Actinoplanes ME3, and Micromonospora MM18 has been found to promote the growth of Discaria trinervis in symbiosis with Frankia; however, no plant-growthpromoting effect was observed when rhizoactinomycetes were applied alone to the plant (Solans 2007). In a similar study, Hamdali et al. (2010) isolated a population of PS actinomycetes spp. from Moroccan phosphate mines and tested their growthpromoting efficacy using wheat as a test plant. The most active RP-solubilizing strains had the highest stimulatory effect on the production of plant biomass. Of the various isolated actinomycetes strains, strain BH7 of Streptomyces griseus stimulated aerial growth of the plant by 70 % in test tubes and more than 30 % in RP soil compared to the non-inoculated control plants. In another study, the actinomycetes showed P solubilization activity $(1,916 \text{ mg } 1^{-1})$ and produced phytase (0.68 U ml^{-1}) , chitinase (6.2 U ml^{-1}) , IAA $(136.5 \text{ mg l}^{-1})$, and siderophore $(47.4 \text{ mg } 1^{-1})$. Furthermore, inoculation of *Streptomyces* mhcr0816 and mhce0811 with Triticum aestivum (wheat) significantly improved plant growth, biomass (33 %), and mineral (Fe, Mn, P) content in non-axenic conditions (Jog et al. 2014).

6.4 Conclusion

In order to reduce the environmental and economic stress due to massive use of synthetic fertilizers and to achieve food security goals, the use of actinomycete as biofertilizers in intensive agriculture practices appears to be a sound, inexpensive, and eco-friendly option. Even though the physiological functions and symbiotic roles of actinomycetes have been revealed under in vitro condition, the role of actinomycetes in sustainable crop production is not adequately explored. Considering the importance of actinomycetes in plant growth promotion via disease suppression and some other mechanisms, there is urgent need to popularize and maximize the use of actinomycetes in crop production in order to reduce dependence on chemical fertilizers and hence to preserve soil fertility without damaging the soil dwellers.

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