

## CHAPTER

## 5

# Streptomyces

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## 1. Introduction

*Streptomyces* is a Gram-positive bacterium, with a high guanine + cytosine (G + C) content, belonging to the family *Streptomycetaceae* and order Actinomycetales. It is found commonly in marine and fresh water, rhizosphere soil, compost, and vermicompost. *Streptomyces* plays an important role in the plant growth promotion (PGP), plant health promotion (crop protection), degradation of organic residues, and production of byproducts (secondary metabolites) of commercial interest in agriculture and medical fields. *Streptomyces*, in the rhizosphere and rhizoplane, help crops in enhancing shoot and root growth, grain and stover yield, biologic nitrogen fixation, solubilization of minerals (such as phosphorus and zinc), and biocontrol of insect pests and plant pathogens. There is a growing interest in the use of secondary metabolites produced by *Streptomyces* such as blasticidin-s, kusagamycin, streptomycin, oxytetracycline, validamycin, polyoxins, natamycin, actinovate, mycostop, abamectin/ivermectins, emamectin benzoate, polynactins and milbemycin for the control of insect pests and plant pathogens as these are highly specific, readily degradable, and less toxic to environment (Aggarwal et al., 2016). The PGP potential of *Streptomyces* is well documented in tomato, wheat, rice, bean, chickpea, pigeonpea, and pea. This chapter emphasizes the usefulness of *Streptomyces* in PGP, grain and stover yields, soil fertility, and plant health promotion.

## 2. Taxonomy of *Streptomyces*

*Streptomyces* is Gram-positive aerobic actinobacteria with high G + C DNA content of 69–78 mol % (Korn-Wendisch and Kutzner, 1992). The cell wall of *Streptomyces* is similar to that of any other Gram-positive bacteria as it contains a simple peptidoglycan mesh surrounding the cytoplasmic membrane (Gago et al., 2011). *Streptomyces* is the predominant genus among the actinobacteria followed by *Micromonospora*, *Mycobacterium*, *Actinomadura*,

*Saccharopolyspora*, *Microbispora*, *Nonomurea*, *Frankia*, *Actinoplanes*, *Verrucosispora*, and *Nocardia* (Martinez-Hidalgo et al., 2014). *Streptomyces* represents 50% of the total population of soil actinobacteria (Sathya et al., 2017). It is a filamentous bacteria representing the family *Streptomycetaceae* and order *Actinomycetales* that includes more than 500 species occurring in soil and water (Barca et al., 2016). *Streptomyces* was proposed by Waksman and Henricia (1943) and classified initially on the basis of morphology (color of hyphae and spores), chemotype, whole-cell sugars, fatty acid and phospholipid profiles, and composition of cell wall (peptidoglycan type) (Kroppenstedt et al., 1990) and later on the basis of phenotypic and genotypic traits (Anderson and Wellington, 2001). Previously, the taxonomic systems utilized phenotypic traits that helped to resolve the intergeneric relationships within the family *Streptomycetaceae*. However, the genotypic traits enabled considerable advances for genus delimitation within the *Streptomyces* (Stackebrandt et al., 1997). For instance, earlier *Streptomyces* and *Streptoverticillium* were considered two different genera; however, with the intervention of immunodiffusion and 16S and 23S rRNA analysis, these two genera were made synonyms of *Streptomyces* (Stackebrandt and Woese, 1981; Logan, 1994). Classification of *Streptomyces* based on morphologic, chemotaxonomic, and molecular studies is summarized in Table 5.1.

TABLE 5.1 Classification of genus *Streptomyces* based on morphologic, chemotaxonomic, and molecular studies.

Classification	Parameters	Characteristics	References
Morphologica classification	Mycelial morphology	Permanent and highly differentiated branched mycelia	Atlas (1997)
	Spore chain morphology	Spores grow out from the aerial mycelium	Cross and Goodfellow (1973)
	Spore chain length	Produce very long chains up to 100 spores. Spore chains classified as straight to flexuous, open loops, and open or closed spirals or verticillate	Pridham et al. (1958)
	Melanoid pigments	Produce brown melanin	Lechevalier and Lechevalier (1965)
Chemotaxonomic classification	Cell wall type	Contains LL-DAP, glycine, no sugar and chemo type 1	Lechevalier and Lechevalier. (1980)
	Taxonomic markers	Contains L-diaminopimelic acid, xylose, and madurose	Labeda (1987)
	Fatty acids	Contains Iso- and ante iso-branched chain fatty acids,	Hofheinz and Grisebach (1965), Lechevalier (1977), Kroppenstedt (1985)
	Biochemical tests	Using fluorogenic probe (targets hydrolysis enzyme)	Goodfellow et al. (1987)
	Whole-cell analysis	Curie-point pyrolysis mass spectrometry (PyMS)	Sanglier et al. (1992)
Molecular classification	Genome sequencing	16S and 23S r RNA analysis, DNA-RNA pairing	Gladek et al. (1985), Witt and Stackebrandt (1990)

### 3. Isolation of *Streptomyces*

*Streptomyces* is well known for its prolific production of useful bioactive compounds (Kekuda et al., 2014). More than 75% of all known naturally occurring antibiotics (such as chloramphenicol, cypemycin, grisemycin, neomycin, and bottromycins) and a wide range of structurally diverse compounds with various pharmaceutical applications were isolated from *Streptomyces* (Sathya et al., 2016a). *Streptomyces* has been the most preferred source of microbes for all types of bioactive secondary metabolites that have important applications in both human medicine and agriculture (Watve et al., 2001). Hence, isolation and identification of novel *Streptomyces* species is very important and the need of the hour.

For preferential isolation of *Streptomyces* from soil, various methods have been reported, which include a variety of selective media formulations, where *Streptomyces* colonies can be increased and other bacterial colonies effectively decreased. For instance, *Streptomyces* can be grown exclusively on Benedict's modification of the Lindenbein medium (Porter et al., 1959); pretreatment of soil suspension with yeast extract and sodium dodecyl sulfate followed by heat shock (dried in a hot air oven at 45°C for 1 h, this activated spores of *Streptomyces* and inhibited other soil bacteria; Williams et al., 1972); nalidixic acid (10–20 mg/L) has been proven effective when combined with humic acid vitamin agar, such as starch casein nitrate agar and glycerol arginine agar (Masayuki and Nideo, 1989); soil supplemented with sodium chloride and rifampicin enhanced the recovery of uncommon *Streptomyces* species (Duangmal et al., 2005); and pretreatment of the soil samples reduced the growth of ubiquitous microbial species (Singh et al., 2016a). Pretreatment of the soil suspension with 1.5% phenol (30°C for 30 min) denatures the proteins or disrupts the cell membrane of bacteria, fungi, and other common actinomycetes there by lowering their number but enhances the number of *Streptomyces* (Hayakawa et al., 1991).

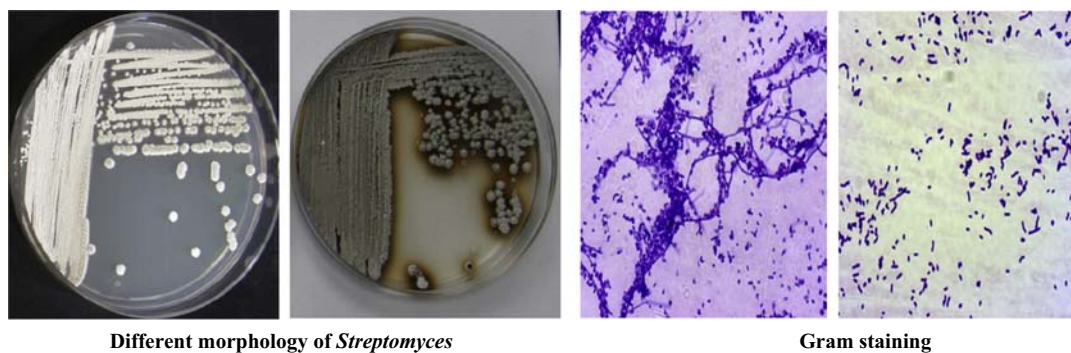
Hayakawa et al. (1991) reported a protocol for isolation of *Streptomyces* from rhizosphere soil. In brief, 1 g of soil sample was suspended in 10 mL of physiologic saline (0.85% of NaCl) and distributed in aliquots. One aliquot of the soil sample was treated with heat for 1 h at 120°C, and the other was treated with 1.5% phenol for 30 min at 30°C. The physicochemically treated soil samples were vortexed and left at room temperature for 30 min. The soil samples were serially diluted up to  $10^{-5}$ , and 0.1 mL of each dilution was plated on actinomycetes isolation agar (AIA), yeast malt glucose agar (M6), starch casein agar (SCA) and Czapek Dox agar. The prominent *Streptomyces* colonies were checked for purity and stored for characterization as desired. Gopalakrishnan et al. (2011a) reported a much simpler protocol for isolation of *Streptomyces* from soil/compost/vermicompost. In brief, 10 g of rhizosphere soil/compost/vermicompost were suspended in 90 mL of saline in a flask and placed on an orbital shaker (at 100–120 rpm) at room temperature ( $28 \pm 2^\circ\text{C}$ ) for 45–60 min. At the end of incubation, the soil/compost/vermicompost samples were serially diluted up to  $10^6$  dilutions with saline. Dilutions  $10^3$ – $10^6$  were plated (0.1 mL) on AIA, SCA, and/or Kenknight and Munaiers agar (KMA) by spread plate techniques. The plates were normally incubated at  $28 \pm 2^\circ\text{C}$  for 5–15 days. The most prominent colonies (the ones that produce pigments, found abundantly in the media plate and inhibiting the adjacent colonies) were isolated and maintained on AIA/SCA/KMA slants at 4°C for further studies.

## 4. Identification of *Streptomyces*

*Streptomyces* colonies can be easily identified by their small opaque, rough, nonspreading morphology and are usually embedded in agar medium (Fig. 5.1). *Streptomyces* is aerobic, slow growing, chalky or glabrous, and heaped. Its aerial and substrate mycelia are of different colors, and the spore mass color may vary from white, gray, red, yellow, green, and blue to violet (Pridham, 1965). It was also observed that some of the strains produced highly colored diffusible pigments on surrounding medium (Fig. 5.1). Color of the aerial mycelium is one of the prominent identification characters of *Streptomyces* isolates at the species level (Pridham and Tresner, 1974). An earthy odor is the typical characteristic feature of *Streptomyces* species. Earthy odor is due to the production of a secondary metabolite called, “geosmin” by *Streptomyces* (Zuo et al., 2010). A usual confirmatory identification of *Streptomyces* genus is long filamentous, Gram positive, and acid-fast negative (Fig. 5.1). *Streptomyces* degrade substrates including casein, tyrosine, and xanthine, but the rate varies with individual isolate. Some of them produce antibiotics, reflected by growth inhibition among other inhabitants of soil samples.

Pridham and Gottlieb (1948) and Shirling and Gottlieb (1966) characterized the *Streptomyces* for production of acid and utilization of different carbon sources including adonitol, sorbitol, dextrose, fructose, inositol, lactose, maltose, raffinose, rhamnose, sucrose, and xylose. Biochemical characterization of *Streptomyces* was done in detail by Hossain and Rahman (2014). According to this report, *Streptomyces* are nonmotile, catalase positive, oxidase positive, urease positive, and hydrogen sulfide positive. Detailed results of biochemical tests of *Streptomyces* are shown in Table 5.2.

Molecular identification of the *Streptomyces* is usually done by 16S rDNA analysis as per the methods explained by Gopalakrishnan et al. (2011a). In brief, pure cultures of *Streptomyces* were grown in starch casein broth until log phase (4 days), and genomic DNA was isolated according to the protocols of Bazzicalupo and Fani (1995). The amplification of 16S rDNA gene was done by using universal primer 1492R (5'-TAC GGY TAC CTT GTT ACG ACT T-3') and 27F (5'- AGA GTT TGA TCM TGG CTC AG-3') (Pandey et al., 2005). The PCR product was sequenced. The sequences obtained were compared with those from the GenBank



Different morphology of *Streptomyces*

Gram staining

FIGURE 5.1 Morphology of *Streptomyces* on starch casein agar and in Gram's staining.

TABLE 5.2 Biochemical characteristics of *Streptomyces*.

Traits	Reaction
Melanin pigment	D
Catalase	+
Oxidase	+
Urease	+
Hydrogen sulfide	+
Nitrate reduction	-
Methyl red	-
Voges-Proskauer	-
Citrate utilization	+
Hydrolysis of casein	+
Starch	D
Lipid	+
Utilization of carbon source	+
D-glucose	
D-mannitol	+
Fructose	+
Sucrose	D
Utilization of nitrogen source D-alanine	+
L-arginine	+
L-tyrosine	+

+, positive reaction; -, negative reaction; D, different isolates gave different reaction.

using the BLAST program, aligned using the Clustal W software and phylogenetic trees inferred using the neighbor-joining method in the MEGA version 4 program (Saitou and Nei, 1987; Alschul et al., 1990; Thompson et al., 1997; Tamura et al., 2007).

## 5. Beneficial role of *Streptomyces* in ago-ecology: in vitro PGP and biocontrol traits of the *Streptomyces*

*Streptomyces* are known to produce PGP and biocontrol traits including cellulase (on cellulose congo red agar; Bhattacharya et al., 2009), lipase (on tween 80 agar; Hendricks et al., 1995), protease (casein agar; Hendricks et al., 1995), chitinase (minimal media with 5% colloidal chitin; Hirano and Nagao, 1988),  $\beta$ -1,3-glucanase (on tryptic soy broth supplemented with 1% colloidal chitin; Singh et al., 1999), indole acetic acid (IAA; on starch casein

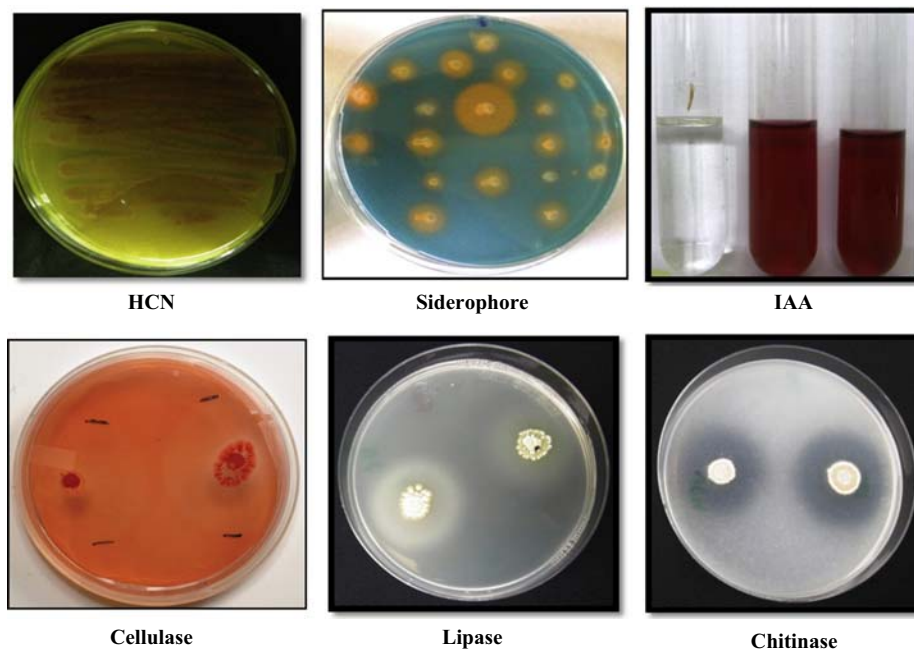


FIGURE 5.2 In vitro PGP and biocontrol traits of *Streptomyces*.

broth supplemented with L-tryptophan; Patten and Glick, 2002), siderophore (on King's B broth; Schwyn and Neilands, 1987), hydrocyanic acid (HCN; on starch casein broth amended with glycine by sulfocyanate method; Lorck, 1948) and 1-aminocyclopropane-1-carboxylate (ACC) deaminase (on starch casein agar using ACC as the sole nitrogen source; Penrose and Glick, 2003) under in vitro conditions (Fig. 5.2; Gopalakrishnan et al., 2011a, 2014; Vijayabharathi et al., 2018a, 2018b). The ability of the *Streptomyces* to produce hormones and enzymes is reported to play a significant role in PGP and plant health, particularly against insect pests and plant pathogens. For instance, protease- and cellulase-producing bacteria play an important role in the mineralization of major nutrients (NPK) and PGP (Lima et al., 1998);  $\beta$ -1,3-glucanase and HCN-producing bacteria plays a role in disease suppression (Haas et al., 1991; Singh et al., 1999); IAA-producing bacteria stimulate seed germination and root formation, so the host plant gets greater access to water and soil nutrients (Ahmad and Kibret, 2014); siderophore-producing bacteria solubilize iron from minerals under conditions of iron limitation (Indiragandhi et al., 2008).

## 6. In vitro physiologic traits of the *Streptomyces*

*Streptomyces* is an alkaline lover, growing well in alkaline soil conditions. They were widely reported to grow well with pH 5–11, NaCl concentration of 10%, and temperatures

between 20 and 40°C. *Streptomyces* were also found highly tolerant to fungicide bavistin, slightly tolerant to thiram and captan, but highly sensitive to radonil, benomyl, and benlate (Gopalakrishnan et al., 2012, 2014; Sadeghi et al., 2012). *Streptomyces* were also found highly resistant to ampicillin and trimethoprim (>800 ppm) and sensitive to chloramphenicol, kanamycin, and nalidixic acid (50–100 ppm) (Gopalakrishnan et al., 2012). It is concluded that *Streptomyces* may have the ability to survive in harsh environments including saline and acidic to alkaline pH soils and thus can be part of integrated disease management programs. Further, antibiotics such as ampicillin and trimethoprim (highly resistant; >800 ppm) and streptomycin and tetracycline (highly sensitive; <25 ppm) could be used as markers for their identification in the natural environment (field evaluation) studies.

### 7. *In planta* PGP traits of the *Streptomyces*

*Streptomyces* in the rhizosphere has been reported to enhance PGP traits including root volume, root growth (length), root hair development, nodulation, plant hormone concentrations, shoot growth, tiller number, panicle number, grain/fruit yield and stover yield on cereals, legumes, and horticultural crops. PGP *Streptomyces* promote plant growth through various mechanisms including biologic nitrogen fixation, solubilization of minerals (such as phosphorus, potassium, and zinc), chelation of iron, and secretion of plant growth hormones (such as auxin). The PGP potential of *Streptomyces* has been demonstrated under field conditions on tomato, wheat, rice, bean, pea, pigeonpea, and chickpea (Tokala et al., 2002; Nassar et al., 2003; Shaukat et al., 2006; El-Tarabily, 2008; Richardson et al., 2009; Sadeghi et al., 2012; Gopalakrishnan et al., 2012, 2014; 2015, 2016a,b; Sreevidya et al., 2016; Vijayabharathi et al., 2018a). *Streptomyces* colonization on the roots of host plants at the right place and time is important for PGP traits. Successful host–*Streptomyces* interaction happens only when they are present in sufficient population, their root-colonizing ability, PGP capability, and their rhizosphere competence (Lugtenberg and Dekkers, 1999).

### 8. *In planta* biocontrol traits of the *Streptomyces*

*Streptomyces* are widely reported to have plant disease suppression against wide variety of plant pathogens. For instance, *S. hygroscopicus* antagonized *Rhizoctonia solani* (causes pea root rot; Rothrock and Gottlieb, 1984); *Streptomyces* spp. were shown to control *Fusarium oxysporum* f. sp. *ciceri* (FOC; causes wilt in chickpea; Bashar and Rai, 1994); *Streptomyces* spp. inhibited *Fusarium oxysporum* f. sp. *cubense* (causes wilt in banana; Getha et al., 2005); *Streptomyces* spp., isolated from national parks in Kenya, were reported to inhibit FOC (Nonoh et al., 2010). A set of five *Streptomyces* strains were demonstrated to have antagonistic potential against FOC under both greenhouse and field conditions (Gopalakrishnan et al., 2011a). Singh et al. (2016b) reported antagonistic activity against *Rhizoctonia solani* on tomato by *Streptomyces* species including *S. coelicolar*, *S. girseus*, *S. albus*, *S. antibiotics*, and *S. champavatii* over the uninoculated control plants. Vijayabharathi et al. (2018b) reported a

set of three strains of *Streptomyces* and their consortia to have biocontrol potential against *Botrytis* gray mold disease in chickpea.

Endophytic *Streptomyces* also plays a role in plant disease suppression. For instance, endophytic *Streptomyces* from citrus were found to have antifungal activity against *Fusarium oxysporum*, *Colletotrichum sublineolum*, *Phytophthora parasitica*, *Guignardia citricarpa*, *Rhizoctonia solani*, and *Pythium* sp. (Quecine et al., 2008); endophytic *Streptomyces* were reported to control *Phytophthora* (Misk and Franco, 2011) and *Sclerotium rolfsii* in chickpea (Singh and Gaur, 2016, 2017); Selected endophytic *Streptomyces* were found to reduce the severity of *Botrytis cinerea* (causes *Botrytis* gray mold disease) both as single inoculation and consortium irrespective of the chickpea genotypes used (Vijayabharathi et al., 2018a). Antioxidant enzymes play an important role in plant disease suppression. *Streptomyces* are reported to enhance antioxidant enzymes. For instance, antioxidant enzymes such as polyphenol oxidase, catalase, superoxide dismutase, glutathione reductase, phenylalanine ammonia-lyase, ascorbate peroxidase, and guaiacol peroxidase were found induced in the chickpea leaves inoculated with endophytic *Streptomyces* over uninoculated control (Vijayabharathi et al., 2018a).

## 9. Secondary metabolite production traits of the *Streptomyces*

Actinobacteria are well known and widely reported for production of secondary metabolites, of which the genus *Streptomyces* is the major producer (39%) of them, including anti-fungal, antibacterial, antiviral, anticancer agents, insecticides, herbicides, antiparasitic agents, immune suppressants, antioxidants, and enzyme inhibitors (Bérdy, 2012). Among the known antibiotics, the majority of them (>60%) are produced by *Streptomyces* (Bérdy, 2005; Sharma, 2014). Secondary metabolites produced by *Streptomyces* include blasticidin-S (produced by *S. griseochromogenes*), kusagamycin (by *S. kasugaensis*), streptomycin (by *S. griseus*), oxytetracycline (by *S. rimosus*), validamycin (by *S. hygrosopicus*), polyoxins (by *S. cacaoi* var *asoensis*), natamycin (by *S. natalensis* and *S. chattanoogaensis*), actinovate (by *S. lydicus* WYEC 108), mycostop (by *Streptomyces* sp. K61), abamectin/avermectins (by *S. avermitilis*), emamectin benzoate (by *S. avermitilis*), polynactins (by *S. aureus*), and milbemycin (by *S. hygrosopicus* subsp. *aureolacrimosus*), which are used commercially as crop protection agents (Aggarwal et al., 2016). Owing to their high specificity, these novel secondary metabolites of microbial origin are much superior in safety for beneficial insect pests, mammalian animals, and humans. Therefore, *Streptomyces* can be a good alternative for the management of insect pest and plant pathogens of agriculturally important crops.

## 10. *Streptomyces* research at ICRISAT

International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), located at Patancheru, Hyderabad, Telangana, India, has been working on PGP and biocontrol research using *Streptomyces* since 2008 on crops including rice, sorghum, chickpea, and pigeonpea. A total of 19 *Streptomyces* strains such as CAI-13 (isolated from the foliage compost of *Allium sativum*), CAI-17 (from the foliage compost of *Chrysanthemum morifolium*), CAI-21 (from the



foliage compost of *C. morifolium*), CAI-24 (from the foliage compost of *Momordica charantia*), CAI-26 (from the foliage compost of *A. sativum*), CAI-68 (from the foliage compost of *Nerium indicum*), CAI-78 (from the foliage compost of *Parthenium hysterophorus*), CAI-85 (from the foliage compost of *Pongamia pinnata*), CAI-93 (from the foliage compost of *Azadirchta indica*), CAI-121 (from foliage compost of *C. morifolium*), CAI-127 (from foliage compost of *A. sativum*), CAI-140 (from foliage compost of *N. indicum*), CAI-155 (from the foliage compost of *Thevetia peruviana*), KAI-26 (from the foliage compost of *Oryza sativa*), KAI-27 (from the foliage compost of *O. sativa*), KAI-32 (from the foliage compost of *O. sativa*), KAI-90 (from the foliage compost of *O. sativa*), KAI-180 (from the foliage compost of *O. sativa*), and MMA-32 (from live soil) were reported (Gopalakrishnan et al., 2011b). The majority of these *Streptomyces* strains were found to produce PGP and biocontrol traits including cellulase, lipase, protease, chitinase,  $\beta$ -1,3-glucanase, IAA, siderophore, HCN, and ACC deaminase under in vitro conditions (Table 5.3). The 19 *Streptomyces* strains were also found to grow well with pH 5–11, NaCl concentration up to 10%, temperatures between 20 and 40°C, and tolerant to fungicides bavistin, thiram, and captan and sensitive to radonil, benomyl, and benlate (Table 5.4).

Under field conditions, the 19 *Streptomyces* strains were demonstrated to enhance tiller and panicle numbers (for rice in particular), root length, root volume, root weight, shoot weight, nodule number and weight (for chickpea and pigeonpea in particular), grain yield, and stover yield on rice, sorghum, chickpea, and pigeonpea on multiple years. These strains were found to enhance available P, total N, organic carbon (%), microbial biomass nitrogen and carbon, and dehydrogenase activity in the rhizosphere. The majority of these 19 strains were also demonstrated to enhance biofortification traits including iron and zinc under field conditions (Table 5.5), so these strains can be exploited in biofortification breeding programs. Colonization of these *Streptomyces* were demonstrated on chickpea roots by scanning electron microscopy analysis. Chickpea plants inoculated with *Streptomyces* exhibited significant surface colonization (Fig. 5.3). The data for PGP traits including shoot and root, grain and stover yields, the biologic activities of the rhizosphere soil, along with their root colonization under field conditions clearly demonstrate the PGP effects of the 19 *Streptomyces* strains. More details of the 19 strains can be found in these literatures (Gopalakrishnan et al., 2012, 2014, 2015, 2016a; Sathya et al., 2016a, 2017; Sreevidya et al., 2016).

Apart from their PGP traits, the 19 *Streptomyces* strains were also shown to have antagonistic traits against important pathogens of chickpea and sorghum, particularly against *Fusarium* wilt in chickpea and charcoal rot in sorghum (Fig. 5.4). A set of five *Streptomyces* strains (CAI-24, CAI-121, CAI-127, KAI-32, and KAI-90) were demonstrated to control *Fusarium* wilt in chickpea under field conditions (Gopalakrishnan et al., 2011a), and another set of eight strains of *Streptomyces* (such as CAI-17, CAI-21, CAI-26, CAI-68, CAI-78, KAI-26, KAI-27, and MMA-32) were shown to have biocontrol traits against charcoal rot in sorghum (Gopalakrishnan et al., 2011b). Vijayabharathi et al. (2018a,b) also reported a set of three strains of *Streptomyces* and endophytic *Streptomyces* and their consortia to have biocontrol potential against *Botrytis* gray mold disease in chickpea.

In addition to the aforementioned *Streptomyces* strains, ICRISAT also identified another series of 16 *Streptomyces* strains (BCA-508, BCA-546, BCA-659, BCA-667, BCA-689, BCA-698, CAI-8, CAI-13, CAI-70, CAI-85, CAI-87, CAI-132, CAI-133, CAI-155, and SAI-25) having the potential to control insect pests of chickpea and sorghum including *Helicoverpa armigera*, *Spodoptera litura*, and *Chilo partellus* (Vijayabharathi et al., 2014). A novel insecticidal

TABLE 5.3 In vitro enzymatic and antagonistic activities and secondary metabolite production by the 19 *Streptomyces* strains of ICRISAT.

Isolate	Scientific name	NCBI No.	PGP properties			Biocontrol properties					Antagonistic activity						
			IAA	Sid	HCN	Cel	Lip	Pro	Chi	$\beta$ 1-3-	Foc	MP	RB-6	RB-24	RB-115	Bot	Scl
CAI-13	<i>Streptomyces</i> sp.	KF770891	25.4	2	2	+	+	-	+	0.25	+	+	+	+	-	+	-
CAI-17	<i>Streptomyces</i> sp.	JQ682619	0.34	2	3	+	+	-	+	0.66	+	+	+	+	+	+	-
CAI-21	<i>Streptomyces</i> sp.	JQ682620	1.13	1	3	+	-	-	-	0	+	+	-	-	-	-	+
CAI-24	<i>Streptomyces</i> sp.	JN400112	5.9	3	3	+	+	-	+	0	+	-	+	-	-	+	-
CAI-26	<i>Streptomyces</i> sp.	JQ682621	1.17	2	2	-	-	-	+	0	+	+	+	-	-	-	-
CAI-68	<i>Streptomyces</i> sp.	JQ682622	0.22	3	3	+	+	-	-	0.66	-	-	-	-	-	+	-
CAI-78	<i>Streptomyces</i> sp.	JQ682623	0.95	0	2	+	+	-	-	2.92	+	-	+	+	-	+	-
CAI-85	<i>Streptomyces</i> sp.	KF770897	43.6	1	2	+	-	-	-	1.21	+	+	+	+	-	+	-
CAI-93	<i>S. fungicidicus</i>	KF742498	33.6	2	2	+	-	+	+	0	+	+	+	+	+	+	-
CAI-121	<i>Streptomyces</i> sp.	JN400113	43.7	3	2	+	-	-	+	0	+	-	-	-	-	+	-
CAI-127	<i>Streptomyces</i> sp.	JN400114	3.5	4	3	+	+	-	+	0	+	-	-	-	-	+	-
CAI-140	<i>S. coelicolor</i>	KF742497	15.4	1.3	3	+	+	+	-	0.353	+	+	+	+	-	+	-
CAI-155	<i>Streptomyces</i> sp.	KF770896	12.6	2	3	+	+	+	+	0.76	+	+	+	+	-	+	-
KAI-26	<i>Streptomyces</i> sp.	JQ682624	0.4	3	1	+	+	+	+	0.35	+	+	+	+	-	+	-
KAI-27	<i>Streptomyces</i> sp.	JQ682625	0.74	1	2	3	-	+	+	0.2	+	+	+	+	-	+	-
KAI-32	<i>Streptomyces</i> sp.	JN400115	2.3	3	3	+	-	-	+	0	+	+	+	+	-	+	-
KAI-90	<i>Streptomyces</i> sp.	JN400116	0	3	3	+	+	-	+	0	+	-	+	+	+	+	-
KAI-180	<i>Streptomyces</i> sp.	KF742499	30.1	0	2	+	+	+	+	0	+	+	+	+	+	+	-
MMA-32	<i>S. roseoviolaceus</i>	JQ682626	4.66	3	2	+	-	-	+	0	+	+	+	-	-	+	+

IAA ( $\mu\text{g/mL}$ ), Bot, *Botrytis cinerea*; Cel, cellulase; Chi, chitinase; Foc, *Fusarium oxysporum*; HCN, hydrocyanic acid; Lip, lipase; MP, *Macrophomina phaseolina*; Pro, protease; RB-6, 24 and 115, *Rhizotonia bataticola*; Scl, *Sclerotia rolfsii*; Sid, siderophore;  $\beta$ , 1-3,  $\beta$ , 1-3, glucanase.

TABLE 5.4 Effect of pH, temperature, salinity and fungicides on the growth by the 19 *Streptomyces* strains of ICRISAT.

Isolate	Physiologic			Fungicide				
	pH	T °C	S (%)	Bav	Ben	Cap	Rid	Thi
CAI-13	7-11	20-40	8	+	-	+	+	+
CAI-17	7-13	20-40	10	+	+	+	+	+
CAI-21	7-11	20-40	12	+	+	+	+	+
CAI-24	7-11	20-40	6	+	+	+	+	+
CAI-26	7-13	20-40	10	+	+	+	+	+
CAI-68	7-11	20-40	8	+	+	+	+	+
CAI-78	7-13	20-40	10	+	+	+	+	+
CAI-85	5-13	20-40	6	+	+	+	+	+
CAI-93	7-11	20-40	8	+	+	+	+	+
CAI-121	5-13	20-40	8	+	+	+	+	+
CAI-127	7-11	20-40	8	+	+	+	+	+
CAI-140	7-11	20-40	10	+	+	+	+	+
CAI-155	7-13	20-40	6	+	+	+	+	+
KAI-26	7-11	20-40	10	+	+	+	+	+
KAI-27	7-11	20-40	10	+	+	+	+	+
KAI-32	5-13	20-40	8	+	+	+	+	+
KAI-90	5-13	20-40	12	+	+	+	+	+
KAI-180	711	20-40	6	+	+	+	+	+
MMA-32	711	20-40	6	+	+	+	+	+

*Bav*, Bavistin; *Ben*, Benlate; *Cap*, Captan; *Rid*, Ridomil; *S*, Salinity; *T*, Temperature; *Thi*, Thiram.

TABLE 5.5 Biofortification potentials of the 19 *Streptomyces* strains of ICRISAT on chickpea.

Isolate	Fe	Zn	Ca	Cu	Mn	Mg
CAI-13	+	+	+	+	+	+
CAI-17	+	+	+	+	+	+
CAI-21	+	+	+	+	+	+
CAI-24	+	+	+	+	+	+
CAI-26	+	+	+	+	+	+
CAI-68	+	+	+	+	+	+

(Continued)

TABLE 5.5 Biofortification potentials of the 19 *Streptomyces* strains of ICRISAT on chickpea.—cont'd

Isolate	Fe	Zn	Ca	Cu	Mn	Mg
CAI-78	+	+	+	+	+	+
CAI-85	+	+	+	+	+	+
CAI-93	+	+	+	+	+	+
CAI-121	+	+	+	+	+	+
CAI-127	+	+	+	+	+	+
CAI-140	+	+	+	+	+	+
CAI-155	+	+	+	+	+	+
KAI-26	+	+	+	+	+	+
KAI-27	+	+	+	+	+	+
KAI-32	+	+	+	+	+	+
KAI-90	+	+	+	+	+	+
KAI-180	+	+	+	+	+	+
MMA-32	+	+	+	+	+	+

Ca, calcium; Cu, copper; Fe, iron; Mg, magnesium; Mn, manganese; Zn, zinc.

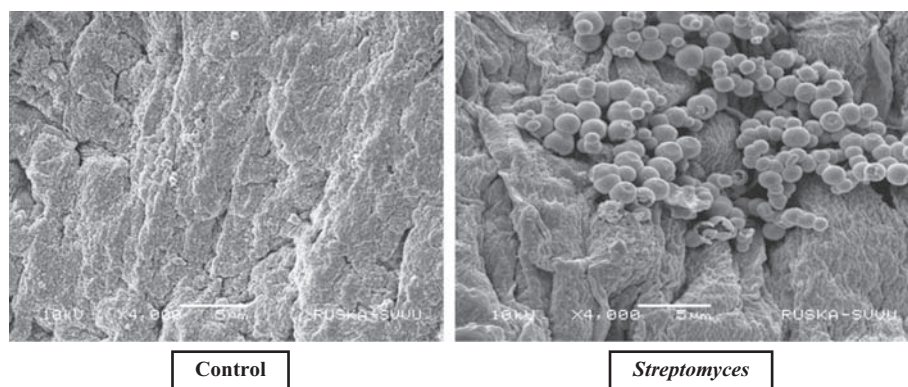
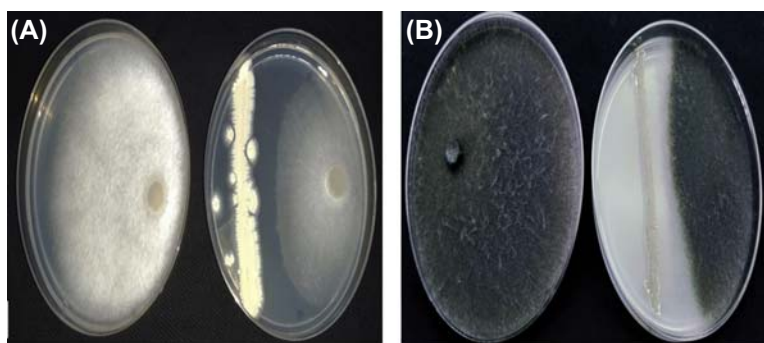


FIGURE 5.3 Scanning electron microscopy (SEM) photographs of *Streptomyces* showing colonization on the roots of chickpea.

metabolite (against *H. armigera*), N-(1-(2,2-dimethyl-5-undecyl-1,3-dioxolan-4-yl)-2-hydroxyethyl) stearamide, was purified from *Streptomyces* sp., CAI-155. The second insecticidal metabolite called cyclo(Trp-Phe) was purified from *S. griseoplanus* SAI-25. Both the metabolites were found to have antifeedant, larvicidal, and pupicidal properties against *H. armigera* (Vijayabharathi et al., 2014); Gopalakrishnan et al. (2016b), Sathya et al. (2016b).



*Streptomyces* showing inhibition against *F. oxysporum* (A) and *M. phaseolina* (B)



Metabolite production assay-against *M. phaseolina*

FIGURE 5.4 Antagonistic activity against fungal pathogens of chickpea and sorghum.

## 11. Conclusion

This chapter was focused on the taxonomy, isolation, identification, and beneficial role of *Streptomyces* and their secondary metabolites in the field of agriculture with more emphasis on their usefulness in PGP and biocontrol against both plant pathogens and insect pests of agriculturally important crops of semiarid tropics. It is concluded that *Streptomyces* have been an important source not only for enhancing PGP and biocontrol traits as inoculants but also for isolation and identification of potent compounds having both insecticidal and fungicidal properties. Hence, this important group of actinobacteria can be a useful component for integrated pest management and integrated nutrition management programs.

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