

Chapter 4

Plant Growth-Promoting Microbes from Herbal Vermicompost

Rajendran Vijayabharathi, Arumugam Sathya,
and Subramaniam Gopalakrishnan

4.1 Introduction

Overreliance on chemical pesticides and fertilizers has resulted in problems including safety risks, outbreaks of secondary pests normally held in check by natural enemies, insecticide resistance, environmental contamination, and decrease in biodiversity (Lacey and Shapiro-Ilan 2008). The increasing costs and negative effects of pesticides and fertilizers necessitate the idea of biological options of crop protection and production. This includes the use of animal manure, crop residues, microbial inoculum (*Rhizobium*, *Azotobacter*, *Azospirillum*, and blue green algae), and composts. They provide natural nutrition, reduce the use of inorganic fertilizers, develop biodiversity, increase soil biological activity, maintain soil physical properties, and improve environmental health (Hue and Silva 2000; Vessey 2003).

On the other hand, a progressive increase in world's population, intensive industrialization of food and beverage processing, and animal husbandry production leads to the generation of large volumes of organic wastes. As per the estimation of World Bank, municipal solid waste alone from the urban areas of Asia is projected to be 1.8 million tonnes/day in 2025 (Chandrappa and Das 2012). These can be disposed by landfilling, pelletization, incineration, biomethanization, and composting. Organic wastes act as a major source of environmental pollution and create serious disposal problem, release odor and ammonia into air, contaminate groundwater, and thereby pose health risks (Inbar et al. 1993). This problem can be solved by vermicomposting, a process of decomposing organic wastes into a valuable product of organic fertilizer and soil conditioner by the use of earthworms.

R. Vijayabharathi • A. Sathya • S. Gopalakrishnan (✉)
International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) Andhra Pradesh,
India, Patancheru 502 324, Andhra Pradesh, India
e-mail: s.gopalakrishnan@cgiar.org

Vermicomposting is an enhanced bio-oxidative and nonthermophilic organic decomposition process by the joint action of earthworms and microorganisms which involves a wide range of organic wastes such as horticultural and agricultural residues, weeds, dry leaves, cow dung, animal droppings, brewery wastes, sericulture wastes, municipal sewage sludge, industrial wastes, paper mills and dairy plants sludge, and domestic and kitchen wastes (Kumar 2005; Chitrapriya et al. 2013). The resultant product of vermicomposting is a stabilized, uniformly sized substance with a characteristic earthy appearance known as “vermicast/vermicompost.” Vermicompost exhibits better performance on various plants during field application due to its enrichment with various macro- and microelements, enzymes, hormones, plant growth regulators, and antibiotics (Makulec 2002; Tilak et al. 2010). Detailed methods of vermicomposting have been documented by many authors (Domínguez 2004; Nagavallemma et al. 2004).

Vermicomposting accelerates decomposition rates which further leads to higher nutrient turnover (Mikola and Setälä 1998; Sampedro and Domínguez 2008) than the traditionally prepared compost which involves the action of microorganisms alone. Though microorganisms act as primary partner for the biochemical decomposition of organic matter, the earthworms, as secondary partner, are crucial drivers for the process and they are broadly grouped into three ecological categories: (1) anecics such as *Lumbricus terrestris*, *L. polyphemus*, and *Aporrectodea longa* are geophagous in nature and live in deep soils; (2) endogeics such as *A. caliginosa*, *Octolasion cyaneum*, *Pontoscolex corethrurus*, and *Aminthas* sp. reside just below the soil surface and feed the organic materials in soils, which were further subdivided into polyhumic, mesohumic, and oligohumic endogeic earthworms; and (3) epigeics such as *Eisenia foetida*, *L. rubellus*, and *Eiseniella tetraedra* live in the upper surface of soils and feed mainly on plant litter and other organic debris available on the soil surface. The details about different earthworm species, their ecological niches, characteristic features, and beneficial actions on decomposition have been reviewed by Pathma and Sakthivel (2012). Since the epigeic earthworms are consumers of a variety of organic matters, they are most suitable for vermicomposting process; however, the use of anecic and endogeic earthworms has also been reported (Lavelle and Martin 1992).

Each earthworm has its own characteristic features on decomposition of organic matter, and they are sensitive to fluctuating climatic and environmental conditions. For instance, *Eudrillus eugeniae* known as the “African night crawler” can decompose large quantities of organic wastes rapidly as it has higher growth and reproduction rates. Hence it is applied widely for vermicomposting and also in combination with other earthworms such as *E. foetida* and *Perionyx excavates* (Pattnaik and Reddy 2010); *P. excavates*, a commercially produced tropical earthworm known as “blues/Indian blues,” is useful for vermicomposting in tropical and subtropical regions (Chaudhuri and Bhattacharjee 2002); *L. terrestris*, an introduced species of North America, is a long-living, cold-tolerant species which makes deep burrows beneath the frost line (Joschko et al. 1989). Domínguez (2004) reported different earthworm species, the factors affecting earthworm

survival (moisture content, temperature, pH, aeration, and ammonia), and also the process of vermicomposting.

Earthworms harbor a variety of decomposer microbes in their gut and excrete them along with nutrients in their excreta, and both are found to be mutual partners. Various enzymes and intestinal mucus in the earthworm’s intestinal tract play a key role in the breakdown of organic macromolecules, which in turn results in a greater increment of the available surface area for microbial colonization, their biological activity, and higher nutrient retention. So, vermicompost is a hotspot for the isolation of beneficial microorganisms, including saprophytic bacteria and fungi, protozoa, nematodes, and microarthropods. Maintenance of mesophilic conditions throughout the entire process is another contributing factor (Domínguez et al. 2010). These microorganisms directly or indirectly offer many agriculturally favorable traits to the vermicompost, but exploration of those microbes has not been studied in detail, though enough reports are available for the microbial diversity of vermicompost (Huang et al. 2013; Pathma and Sakthivel 2012). An overview on the effect of vermicompost and associated microbes on agriculturally useful traits is depicted in Fig. 4.1.

Microbes with agriculturally favorable traits were categorized as plant growth-promoting (PGP) microbes—a heterogeneous group of beneficial bacteria/fungi/actinomycetes which promotes plant growth either directly (nitrogen fixation, phosphate solubilization, iron chelation, and phytohormone production) or

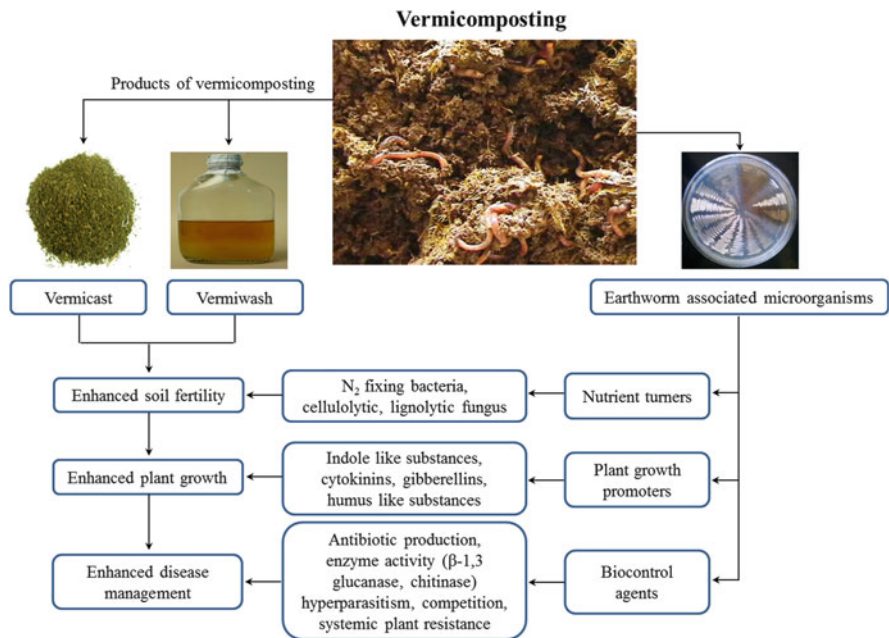


Fig. 4.1 Overview of vermicompost and its associated microbes on plant growth

indirectly (suppression of plant pathogenic organisms, induction of resistance in host plants against plant pathogens, and abiotic stresses).

PGP microbes include *Bacillus*, *Pseudomonas*, *Erwinia*, *Caulobacter*, *Serratia*, *Arthrobacter*, *Micrococcus*, *Flavobacterium*, *Chromobacterium*, *Agrobacterium*, *Hyphomycrobium*, and free-living nitrogen-fixing bacteria and also the members of the family Rhizobiaceae such as *Rhizobium*, *Bradyrhizobium*, *Sinorhizobium*, *Azorhizobium*, *Mesorhizobium*, and *Allorhizobium*. The practice of using such PGP microorganisms to agriculturally important crops as inoculants is getting attraction as it has a wide range of applications including the substantial reduction of the use of chemical fertilizers/pesticides, increased soil health, inhibitory activity against phytopathogens/insects, and enhanced crop yield (Bhattacharyya and Jha 2012; Mehboob et al. 2012). Hence, in this chapter, we intend to deliberate the usefulness of vermicompost and the associated microorganisms in enhancing soil health and agricultural productivity.

4.2 Microbial Diversity of Earthworms and Vermicomposts

Microbial communities including bacteria, actinomycetes, filamentous fungi, and yeast have been reported in earthworms such as *L. terrestris*, *Allolobophora caliginosa*, and *A. terrestris* (Parle 1963a, b), and most of them are mesophilic bacteria, fungi, and actinomycetes (Benitez et al. 1999; Sen and Chandra 2009; Vivas et al. 2009), which have been illustrated in Table 4.1. It is noticed that, earthworm's age hasn't showed any influence on microbial community (Fernández-Gómez et al. 2012), but the microbial counts between the earthworm species may vary due to their different ability to digest and assimilate microbial biomass, their ecological group, food, and environmental conditions in which earthworms live (Brown and Doube 2004). These factors make the vermicompost a hotspot of microbes. Unique indigenous gut-associated microflora has been documented in *E. foetida* (Toyota and Kimura 2000). In contrary, microbes living in traditional compost undergo a selection process during the heating phase, where the organic material is decomposed by specially adapted thermophilic bacteria (Dees and Ghiorse 2001). The microbial community which resides in the finished traditional compost are the facultative thermophiles, which form spores during the hot phase and recolonize during the mesophilic stage.

Microbial count in the ingested material of earthworms can be increased up to 1,000-fold while passing through their gut (Edwards and Fletcher 1988). Devi et al. (2009) have given a distinction on the microbial count of vermicomposts and of normal composts of fruit and vegetable waste, cow dung, and groundnut husk for bacteria, fungi, and actinomycetes. A similar trend of supporting evidence has been given by many research groups (Pedersen and Hendriksen 1993; Devliegher and Verstraete 1995). Microbial biomass and activity were also

Table 4.1 Microbial diversity of earthworms

S. no	Microorganisms	Earthworm	References
1	An oxalate-degrading <i>Pseudomonas oxalaticus</i>	<i>Pheretima</i>	Khambata and Bhat (1953)
2	Anaerobic N ₂ -fixing bacteria— <i>Clostridium butyricum</i> , <i>C. beijerinckii</i> , and <i>C. paraputrificum</i>	<i>E. foetida</i>	Citernesi et al. (1977)
3	<i>Streptomyces lipmanii</i> and <i>Streptomyces</i> spp.	<i>E. lucens</i>	Contreras (1980)
4	Actinobacteria	<i>L. rubellus</i>	Křišťůfek et al. (1993)
5	Fluorescent pseudomonads	<i>L. terrestris</i>	Devliegher and Verstraete (1997)
6	<i>Aeromonas hydrophila</i>	<i>E. foetida</i>	Toyota and Kimura (2000)
7	Gammaproteobacteria, firmicutes, and actinobacteria	<i>L. rubellus</i>	Furlong et al. (2002)
8	<i>Pseudomonas</i> , <i>Paenibacillus</i> , <i>Azoarcus</i> , <i>Burkholderia</i> , <i>Spiroplasma</i> , <i>Acaligenes</i> , and <i>Acidobacterium</i>	<i>L. rubellus</i>	Singleton et al. (2003)
9	Novel nephridial symbiont, <i>Verminephrobacter eiseniae</i>	<i>E. foetida</i>	Pinel et al. (2008)
10	Gammaproteobacteria	<i>L. rubellus</i>	Knapp et al. (2009)
11	Acidobacteria, actinobacteria, bacteroidetes, chloroflexi, cyanobacteria, firmicutes, Gemmatimonadetes, nitrospirae, planctomycetes, proteobacteria, tenericutes, and verrucomicrobia	<i>L. terrestris</i>	Wüst et al. (2011)
12	Aeromonadaceae, comamonadaceae, enterobacteriaceae, flavobacteriaceae, moraxellaceae, “paenibacillaceae,” pseudomonadaceae, rhodocyclaceae, sphingobacteriaceae, and actinobacteria	<i>A. caliginosa</i>	Ihssen et al. (2003), Horn et al. (2003)

significantly increased in vermicasts over composts (Brown and Doube 2004; Aira et al. 2006; Monroy et al. 2009). Earthworms’ interaction with physical, chemical, and biological components affects the structural features of the microflora and microfauna in vermicompost (Domínguez et al. 2003; Lores et al. 2006; Monroy et al. 2009).

A recent study by Huang et al. (2013) on the bacterial communities of the earthworm *E. foetida* showed different phyla including Bacteroidetes, Firmicutes, Actinomycetes, Chlorobi, Planctomycetes, and Proteobacteria in vegetable waste compost, in which Bacteroidetes were predominant. Enrichment of Bacteroidetes (anaerobic group of microorganisms) in the vermicompost is probably due to the anaerobic conditions in the earthworm’s gut (Karsten and Drake 1995). In contrast, Pathma and Sakthivel (2013) noticed *Bacillus* as the dominating genus followed by *Pseudomonas* and *Microbacterium* in goat manure compost. Bacterial diversity analysis of commercial composts (poultry litter, sewage sludge, and municipal solid waste) and homemade composts (vermicompost from food wastes) has been

registered with the groups such as Firmicutes: *Bacillus benzoovorans*, *B. cereus*, *B. licheniformis*, *B. megaterium*, *B. pumilus*, *B. subtilis*, and *B. macroides*; Actinobacteria: *Cellulosimicrobium cellulans*, *Microbacterium* spp., and *M. oxydans*; Proteobacteria: *Pseudomonas* spp. and *P. libaniensis*; ungrouped genotypes: *Sphingomonas* spp. and *Kocuria palustris*; and yeasts: *Geotrichum* spp. and *Williopsis californica* (Vaz-Moreira et al. 2008). Fischer et al. (1995) observed variations in the bacterial community of vermicasts and guts (including foregut, midgut, and hindgut) of earthworms in which the bacterial count of α , β , and γ subgroups of proteobacteria increased significantly toward the end of the gut and remained high in the cast. Among the subgroups, α -proteobacteria was higher in the hindgut and casts, and β - and γ - proteobacteria were predominant in the fore- and hindgut. Similar studies conducted by Nechitaylo et al. (2010) revealed the presence of Bacteroidetes, Alphaproteobacteria, Betaproteobacteria, and representatives of classes Flavobacteria, Sphingobacteria (Bacteroidetes), *Pseudomonas* spp., and unclassified Sphingomonadaceae (Alphaproteobacteria) and *Alcaligenes* spp. (Betaproteobacteria) in earthworm (*L. terrestris* and *A. caliginosa*), casts, and soil.

In addition to bacteria, several studies have also been reported for fungal diversity in vermicompost and earthworms. The phyla of Saccharomycetes, Lecanoromycetes, and Tremellomycetes dominated in the initial substrate of vermicompost (Bonito et al. 2010). The compost without earthworm was reported to have less fungal diversity, whereas during earthworm treatment, the fungal diversity has increased with Sordariomycetes, followed by Agaricomycetes, Pezizomycetes, Eurotiomycetes, Saccharomycetes, and Orbiliomycetes (Bonito et al. 2010; Huang et al. 2013). Besides this, other beneficial fungi in the vermicompost have also been noticed and some of the identified populations include *Paecilomyces* spp. and *Dactylaria biseptata* (Siddiqui and Mahmood 1996), *Cephalophora tropica* (Morikawa et al. 1993), and *Trichoderma* spp. (Harman 2006). A study by Anastasi et al. (2005) also revealed the differentiation of fungal diversity in compost and vermicompost. Among the 194 fungal species isolated, 66 were common to both the compost and vermicompost, whereas 118 were obtained from compost and 142 from vermicompost. This concludes that fungal diversity is found more in vermicompost than in compost.

Next to bacteria, actinomycetes are the major gut flora of earthworm and have been reported widely in the literature (Parle 1963a, b; Ravasz et al. 1987; Ravasz and Tóth 1990; Jayasinghe and Parkinson 2009). It is noticed that vermicompost has higher actinomycetes than fungus in the final product, which might be due to the antagonistic activity of the former group against the latter group (Jayasinghe and Parkinson 2009). For instance, Yasir et al. (2009) and Huang et al. (2013) detected *Streptomyces* and *Rhodococcus*, the genera which have the ability to kill plant pathogens from vermicompost and fresh sludge. The actinomycetes present in the form of cell aggregates or individual cells and most of them belong to *Streptomyces* spp., the well-known antibiotic producers (Krištúfek et al. 1993, 1994, 1995). Other actinomycetes such as *Micromonospora* spp. were also recorded (Krištúfek et al. 1990; Polyanskaya et al. 1996).

Earthworms have food preference for substances colonized by certain fungal (Tiwari and Mishra 1993; Moody et al. 1995; Marfenina and Ishchenko 1997) and bacterial species (Wright 1972). Their food preference for actinomycetes has been demonstrated by Polyanskaya et al. (1996) on *E. foetida*, which actively consumed the spores of *S. caeruleus* than other actinomycete spores. Even though a substantial quantity of actinomycetes is digested in the foregut of the earthworms, the undigested remaining actinomycetes are able to develop rapidly in the earthworm's mid- and hindgut. Hence, the chances of survival for actinomycetes were found to be higher in earthworm's hindgut (Křišťufek et al. 1992; Polyanskaya et al. 1996; Zenova et al. 1996). These ingested actinomycetes inhibit the growth of other microorganisms particularly litter-decomposing and pathogenic fungi and Gram-positive bacteria in the earthworm's gut. This leads to the predominance of other actinomycetes and other antibiotic-resistant microorganisms and hence the bio-control properties against various phytopathogens (Doube et al. 1994a, b; Stephens et al. 1994). Though the microbial community of bacteria/fungi/actinomycetes varies with the earthworm species/vermicompost, it also depends on the initial substrate of vermicompost.

4.3 Nutritional Values of Vermicompost

The nutritional quality of the vermicompost depends on the type of the initial substrate, earthworm species (epigeic, endogeic, and anecic), microbial population (cellulolytic, lignolytic, and N_2 -fixers), and environmental conditions like aeration, humidity, pH, and temperature. The nutrient composition of vermicomposts has been documented with organic carbon 9.2–17.9 %, total nitrogen 0.5–1.5 %, available phosphorus 0.1–0.3 %, available potassium 0.1–0.2 %, calcium and magnesium 22–70 mg/100 g, copper 2–9.3 ppm, zinc 5.7–11.5 ppm, and available sulfur 128–548 ppm (Kale 1995). Vermicompost has higher concentrations of exchangeable Ca^{2+} , Mg^{2+} , and K^+ than the initial substrate, which indicates the conversion of nutrients to plant-available forms during the passage in the earthworm's gut and associated microorganisms. Apart from the nutritional indices, the earthworm's activity also enhances the soil's physical qualities like bulk density, pore size, water infiltration rate, soil water content, and water-holding capacity (Edwards 1998).

A detailed study on the effect of substrate (cow dung, grass, aquatic weeds, and municipal solid waste), liming (enhances earthworm activity and microbial population), and microbial community (*Trichoderma viride*, *Phenacoccus cryosporium*—lignolytic fungus and *Bacillus polymyxa*—free-living nitrogen-fixing bacteria) on the nutritional status of vermicompost has been reported by Pramanik et al. (2007). They found that the usage of cow dung, *B. polymyxa*, and lime concentration of 5 g/kg was found to be the best combination in increasing NPK values, humic acid content, and enzyme activities like urease and phosphatase; however, *T. viridae* has shown equal nutrient effects irrespective of the lime content. Ghosh et al. (1999) demonstrated the difference in composting of organic

wastes such as cow dung, poultry droppings, kitchen wastes, municipal wastes, and dry leaves with and without *E. foetida* and observed higher availability of macro- and micronutrients in vermicast than compost without earthworms. Similarly, three- to fourfold increased NPK and micronutrient content on cow dung vermicompost than the noncomposted parental material was also noticed. Recent studies also concluded the nutritional enrichment of vermicompost over normal compost (Atiyeh et al. 2000; Hashemimajda et al. 2004; Lazcano et al. 2008). Hence, it can be concluded that the extensive usage of vermicompost can reduce the application of chemical fertilizers without affecting crop yield.

Vermicast has been documented with various enzyme activities including cellulase, amylase, invertase, protease, peroxidase, urease, phosphatase, and dehydrogenase in which the maximum enzyme activity is contributed by gut microbes (Sharpley and Syers 1976; Edwards and Bohlen 1996; Devi et al. 2009). Though vermicomposts have a wide range of enzyme activities, fluctuations are there during the composting period that the maximum enzyme activities were observed during 21–35 days in vermicomposting, whereas in conventional composting it was noticed on 42–49 days (Devi et al. 2009). This might be due to higher microbial count and activity in vermicomposts than the conventional composts. Since earthworms influence soil physical, chemical, and biological properties, they have been considered as soil engineers and as indicators of soil quality (Muys and Granval 1997; Jouquet et al. 2006).

4.4 Plant Growth Promoters of Vermicompost

Vermicompost was found to increase the growth of various vegetable, fruit, flower, and food crops not only by their macro- and microelement composition of the vermicast but also by their plant growth-promoting substances like growth hormones and enzymes. Microbes residing in the earthworm are the major contributors of such known and other unknown growth-promoting elements. *Rhizobium*, one of the PGP bacterium in soil that fixes nitrogen, was reported to disperse in soil by the earthworm *A. trapezoids* (Bernard et al. 1994). The first report on the identification of plant growth-promoting substances in earthworms was done by Nielson (1965). He identified indole-like substances in the tissue extracts of *A. caliginosa*, *L. rubellus*, and *E. foetida* and observed enhanced growth rate of garden pea. Various researchers reported substantial quantities of plant growth promoters such as auxins, gibberellins, cytokinins of microbial origin (Grappelli et al. 1985, 1987; Krishnamoorthy and Vajranabhaiah 1986; Tomati et al. 1988; Muscolo et al. 1999), and humic acids (Masciandaro et al. 1997; Atiyeh et al. 2002) in vermicomposts.

Vermiwash, the aqueous extracts of vermicompost, is a collection of excretory compounds of earthworms and also the associated microbes. It serves as a fertilizer and also a biocide due to the presence of macro- and micronutrients and antibiotics compounds. Hence, the use of vermiwash also registered increased plant growth on

a par with the use of hormones such as auxins, gibberellins, and cytokinins on plants such as *Petunia*, *Begonia*, and *Coleus* (Grappelli et al. 1987; Tomati et al. 1987, 1988). Nagavallema et al. (2004) showed a marked difference in the plumule length of maize seedlings dipped in vermiwash than normal water. Comparative studies on the impact of vermiwash and urea solution on seed germination and on root and shoot length in cluster bean, *Cyamopsis tertagonoloba*, demonstrated the enhanced growth in vermiwash solution which might be due to hormone-like substances (Suthar 2010). HPLC and GC-MS analyses of the vermiwash of cattle waste-derived vermicompost showed the presence of significant amounts of indole acetic acid (IAA), gibberellins, and cytokinins (Edwards et al. 2004). Thus, it was demonstrated that both vermicompost and vermiwash are rich source of plant growth-promoting substances.

4.5 Biocontrol Properties of Vermicompost

Microbial population in vermicompost acts as powerful biocontrol agents due to the production of antibiotics and secretion of extracellular enzymes such as chitinase and lipase which cause the lysis of fungal and bacterial phytopathogens. Vermicompost is a valuable source of antagonistic bacteria and/or actinomycetes; several research reports are available to augment the biocontrol properties of vermicompost against phytopathogens such as *Botrytis cineria* (Singh et al. 2008), *Fusarium* spp. (Yeates 1981; Moody et al. 1996), *Gaeumannomyces* spp. (Clapperton et al. 2001), *Rhizoctonia* spp. (Doube et al. 1994a; Hoitink et al. 1997; Stephens et al. 1994; Stephens and Davoren 1997), *Phytophthora* (Ersahin et al. 2009), *Plasmodiophora brassicae* (Nakamura 1996), and *P. infestans* (Kostecka et al. 1996). Control of powdery mildew in barley (Weltzien 1989), balsam, and pea by vermicompost application has been demonstrated under field conditions (Singh et al. 2003). Pathogen control has been demonstrated in other crops like clover, cabbage, cucumber, grapes, tomatoes, radish, and strawberry (Jack 2011). Besides the biocontrol properties of vermicompost, vermiwash was also found to have biocontrol traits against *B. cineria*, *Sclerotinia sclerotiorum*, *Corticium rolfsii*, *R. solani*, *F. oxysporum* (Nakasone et al. 1999), *Erysiphe cichoracearum*, and *E. pisi* (Singh et al. 2003). Systemic plant resistance, microbial competition, antibiosis, enzyme activity, and hyperparasitism are the suspected reasons for pathogenic control (Hoitink and Grebus 1997). Yasir et al. (2009) documented the presence of chitinolytic bacteria *Nocardioides oleivorans*, *Streptomyces* spp., and *Staphylococcus epidermidis* from vermicompost with inhibitory activity against phytopathogens such as *R. solani*, *Colletotrichum coccodes*, *Pythium ultimum*, *P. capsici*, and *F. moniliforme*. Similarly, antibiotic heliomyacin-producing *S. olivocinereus* has been isolated from *E. foetida*'s gut (Polyanskaya et al. 1996). The dispersed actinomycetes from earthworms act as potential biocontrol agents against plant pathogenic fungi (Doube et al. 1994a, b; Stephens et al. 1994) due to their production capacity for a wide range of secondary

metabolites and antibiosis compounds. Besides pathogen control, insects or pests such as jassids, aphids, spider mites, mealy bugs, sucking pests, caterpillars, and beetles have also been controlled by vermicompost application (Edwards et al. 2007; Biradar et al. 1998; Rao et al. 2001; Rao 2002, 2003) under greenhouse and field conditions.

4.6 PGP Research at ICRISAT

ICRISAT has identified over 1,500 microbes including bacteria and actinomycetes, isolated from various composts and rhizospheric soil, in which at least one out of six has documented either single or multiple agriculturally favorable traits. Our research group has a collection of 137 actinomycetes isolated from 25 herbal vermicomposts prepared from *Jatropha curcas*, *Annona squamosa*, *Parthenium hysterophorus*, *Oryza sativa*, *Gliricidia sepium*, *Adhatoda vasica*, *Azadirachta indica*, *Capsicum annuum*, *Calotropis gigantea*, *Calotropis procera*, *Datura metal*, *Allium sativum*, *Zingiber officinale*, *Ipomoea batatas*, *Momordica charantia*, *Moringa oleifera*, *Argyranthemum frutescens*, *Nerium indicum*, *Allium cepa*, *Curcuma aromatica*, *Pongamia pinnata*, *Abacopteris multilineata*, *Nicotiana tabacum*, *Tridax procumbens*, and *Vitex negundo* using the epigeic earthworm *E. foetida* (Gopalakrishnan et al. 2013a) and demonstrated plant growth-promoting and biocontrol properties under laboratory, greenhouse, and field conditions.

Among them, actinomycetes, *Streptomyces* spp., *S. caviscabies*, *S. globisporus* sub sp. *caucasicus*, and *S. griseorubens* isolated from herbal vermicomposts, have registered in vitro PGP traits such as IAA and siderophore production and also documented their positive effect on the upregulation of PGP genes such as IAA and siderophore-producing genes. They proved these in vitro potentials by enhanced growth performance on rice under field conditions via increased tiller numbers, panicle numbers, filled grain numbers and weight, stover yield, grain yield, total dry matter, root length, root volume (Fig. 4.2), and root dry weight. In addition, they significantly enhanced rhizospheric total nitrogen, available phosphorous, % organic carbon, microbial biomass carbon, microbial biomass nitrogen, and dehydrogenase activity over the uninoculated control. Apart from the PGP traits, they also have the capacity to act as biocontrol agents due to the production of hydrogen cyanide and enzymes such as lipase, chitinase, and β -1,3 glucanase (Gopalakrishnan et al. 2012, 2013b, 2014). PGP actinomycetes such as *Streptomyces* spp., *S. tsusimaensis*, *S. caviscabies*, *S. setonii*, and *S. africanus* isolated from herbal vermicomposts have proved this by their inhibitory activity against *Fusarium oxysporum* f. sp. *ciceri* (FOC) (Gopalakrishnan et al. 2011a) and *Macrophomina phaseolina*, a causative agent for the charcoal rot of sorghum (Gopalakrishnan et al. 2011b) under greenhouse conditions. Antagonistic activity of these actinomycetes on *Fusarium* wilt-sick fields has also been demonstrated.

Besides the biocontrol activity of microbes isolated from herbal vermicomposts, washings of vermicompost, “vermiwash or biowash,” were also demonstrated to

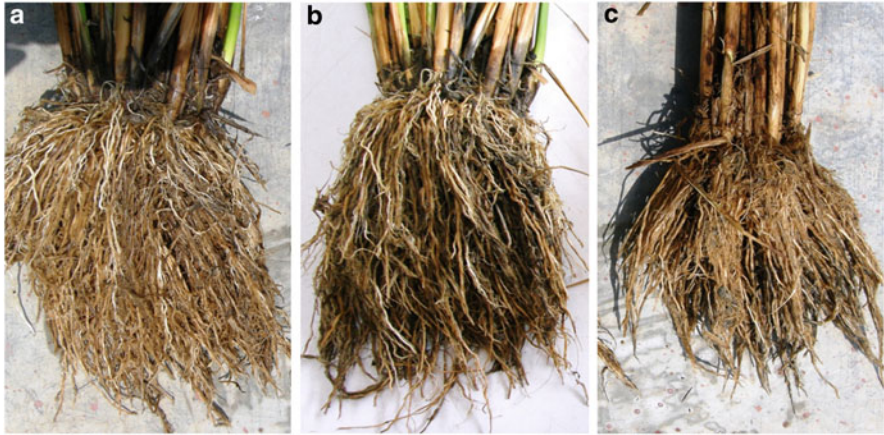


Fig. 4.2 Effect of PGP actinomycetes (a) *S. caviscabies* and (b) *S. globisporus* sub sp. *caucasicus* on root development of rice over (c) uninoculated control (Gopalakrishnan et al. 2014)

have inhibitory activity against phytopathogens. Crude biowash and partially purified extracts of vermicompost prepared from *Jatropha curcas*, *Annona squamosa*, and *Parthenium hysterophorus* marked their fungicidal activity on FOC, *S. rolfsii*, and *M. phaseolina* (Gopalakrishnan et al. 2010). Additionally, insecticidal activity has been registered by biowash and microbes isolated from herbal vermicomposts. Our investigation proved this via the biowash of *Annona*, *Datura*, *Jatropha*, Neem, *Parthenium*, *Pongamia*, and isolated PGP bacteria *B. subtilis*, *B. megaterium*, *Serratia mercerscens*, and *Pseudomonas* spp.; fungus *Metarhizium anisopliae* and actinomycetes *S. cavourensis* sub sp. *cavourensis*, *S. albolongus*, *S. hydrogenans*, *S. antibioticus*, *S. cyaneofuscatus*, *S. carpaticus*, *S. bacillaris*, and *Streptomyces* spp. which were found to have broad-spectrum insecticidal properties against lepidopteran pests such as *Helicoverpa armigera*, *Spodoptera litura*, and *Chilo partellus* (Gopalakrishnan et al. 2011c; Vijayabharathi et al. 2014).

Besides the contribution of actinomycetes, bacteria have also been registered with PGP activity. Phosphate-solubilizing bacteria and *Azotobacter* have been isolated from the vermicompost of cow dung and saw dust with earthworms *E. eugeniae* and *P. excavatus* (Chitrapriya et al. 2013). Similarly, *Bacillus*, *Pseudomonas*, *Rhizobium*, and *Azotobacter* with in vitro PGP traits such as IAA, ammonia, and siderophore production were isolated from the vermicompost of paper mill sludge, leaf litter, and press mud with *E. foetida* (Prakash and Hemalatha 2013). A detailed study by Pathma and Sakthivel (2013), on vermicompost produced from straw and goat manure with *E. foetida*, identified 193 bacteria with antagonistic and/or biofertilizing potential. The dominance of identified bacteria was found to be in the order of *Bacillus* (57 %) > *Pseudomonas* (15 %) > *Microbacterium* (12 %) > *Acinetobacter* (5 %) > *Chryseobacterium* (3 %) with the other members such as *Arthrobacter*, *Pseudoxanthomonas*, *Stenotrophomonas*, *Paenibacillus*, *Rhodococcus*, *Enterobacter*, *Rheinheimera*, and *Cellulomonas*. Functional analyses of

these microbes have registered in vitro PGP traits such as phosphate solubilization, nitrate reduction, assimilation of different carbon sources, and production of IAA, siderophore, 1-aminocyclopropane-1-carboxylate (ACC) deaminase, chitinase, lipase, and HCN. Besides this, they have also been reported with the production of commercially important enzymes protease, cellulase, amylase, xylanase, and Dnase. These studies thus conclude that vermicomposting organisms and biowash have the potency to promote plant growth, control the infectious diseases, and restrict pest attack. Hence, these PGP microorganisms are expected to replace inorganic fertilizer, pesticides, and artificial plant growth regulators which have numerous side effects to sustainable agriculture.

4.7 Conclusions

This chapter was intended to summarize the current knowledge on plant growth-promoting microbes associated with vermicompost. Vermicompost, vermiwash, and earthworm, in specific earthworm gut, nephridia and alimentary canal, have complex group of beneficial microorganisms. These microorganisms directly or indirectly contribute to the beneficial properties of vermicompost and vermiwash in enhancing soil health, plant growth, and hence agricultural productivity. Plenty of literatures are available for the presence/diversity of bacteria, fungi, and actinomycetes in vermicompost and earthworm and also for the enhanced plant growth by vermicompost application. However, studies related to the exploration of such potential microbes with plant growth-promoting properties are scarce. So, investigation on the isolation, identification, and characterization of plant growth-promoting microbes and their active metabolites from vermicompost will be useful for sustainable agriculture.

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