Soil Microflora – An Extensive Research

Sameen Ruqia Imadi, Mustafeez Mujtaba Babar, Humna Hasan, and Alvina Gul

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Abstract Soil is the most complicated biomaterial present on earth. It is composed of a variety of substances and provides a habitat to various organisms. Different chemical reactions take place in soil that ensures the sustainability of life. Microorganisms including bacteria, fungi, actinomycyes and algae are widely distributed in soil. These natural micro flora have several advantages. They contribute to the growth and development of plants, decomposition of organic materials, nutrient cycling, soil nitrification, sustenance of pedological system and production of bioactive compounds. Soil fungi develop mutualistic associations with plants and increase their surface area for absorption. Rhizosphere of soil, the area in which

S.R. Imadi • A. Gul
Atta-ur-Rahman School of Applied Biosciences, National University of Sciences and Technology, Islamabad, Pakistan

e-mail: alvina_gul@yahoo.com

M.M. Babar
Shifa College of Pharmaceutical Sciences, Shifa Tameer-e-Millat University, Islamabad, Pakistan

H. Hasan
Quaid-I-Azam University, Islamabad, Pakistan

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Soil micro flora is present, is rich not only in diverse micro flora but also plant roots and nutrients. Soil pollution and anthropogenic activities used for higher yield of agricultural crops negatively affect the soil micro flora. Pesticides kill micro flora and reduce soil biodiversity. The focus of this chapter is on the advantages of natural flora of soil and various factors causing their degradation. The chapter also sheds light on the changes in micro floral communities due to changes in environment. Towards the end, the future perspectives in which soil micro flora can be used for further benefit of mankind have also been discussed.

**Keywords**  Soil microflora • Symbiosis • Microbe-environment interaction • Microflora applications • Agriculture improvement

1 Introduction

Soil is considered as one of the most complicated biomaterial on earth. It not only provides home to a variety of microorganisms but also serves as a natural laboratory where different kinds of experiments can be performed. In general, soil is formed by the weathering of parent rock material. Thereafter, microbes adapt this soil as their habitat, hence, being characterized as ‘soil micro flora’. The upper 16–17.6 cm plow layer is considered to be the most active in terms of the population density of these micro flora (Suneetha and Khan 2010). Of the different layers of soil, rhizosphere is the region that is directly under the influence of various biological and physiological factors including plant roots and microbes (Chaudhry et al. 2012; Hakeem et al. 2016).

Soil micro flora have been observed to play a fundamental role in the organic matter mineralization in the soil. Plant nutrition and growth in agro systems is also largely dependent on soil micro flora. These microbial processes take place in the rhizosphere (Bérrard et al. 2015). Among the various soil microorganisms, the most prevalent fungi worldwide include species of genus Aspergillus, Fusarium, Verticillum, Penicillium, and Trichoderma. Largest fungal colonies residing in the soil are that of Aspergillus niger (Rajik et al. 2011). A number of experiments have revealed that upon the application of the microbial fertilizers, the number of soil bacteria and actinomycetes is significantly increased. This, ultimately, results in an increase in the crop yield (Wang et al. 2012).

A number of factors contribute to the microbial diversity in the rhizosphere of various biogeographical regions. It has been observed that the soil microbial communities are quite similar among the different cultivated soils, for instance the microbes in the cultivated deciduous forests and mowed grasslands are generally the same. Though there are major differences in soil properties and vegetation conditions in various cultivated lands yet the soils share similar microbial population. There are, however, a few reports in which the conversion of deciduous forest to tilled croplands resulted in a change in the soil properties leading to an alteration in the soil micro flora as well. It can, hence, be assumed that history of land use is a major determinant of soil micro flora along with the vegetation and soil properties.
(Jangid et al. 2011). Moreover, other biological factors, like the species of the earthworm, have also been found to contribute significantly to the nature and amount of bacteria and actinomyces present in soil (Lin et al. 2016).

Quantities of soil bacteria and actinomyces have been observed to significantly decrease by conversion of plantation from evergreen broad leaved forests to Chinese fir plantation (Xia et al. 2012). Similarly, in a recent study, the effect of fertilizers was observed on soil micro flora of watermelon fields. It was seen that the treatment with both organic and inorganic fertilizers resulted in a decrease in the concentration of normal fungi levels in soil. On the other hand, the amount of bacteria and actinomyces was significantly increased as a result of fertilizer treatment (Zhu et al. 2013). Moreover, it has been observed, through paraffin baiting technique based experiments that several species of *Nocardia* genus are present in the soil that also contributes to a diverse microbial community in many cultivated regions (Habibnia et al. 2015).

Effects of application of different organic manures were observed on soil micro flora of mulberry in another recent study. On the treatment of roots of mulberry plants with yard manure biofertilizers, it was observed that root tips had communities of *Azotobacter chroococcum, Gluconacobacter diazotrophicus, Bacillus sonoren sis, Bacillus pumilis, Bacillus coagulans* and *Pseudomonas putida*. The root tips of plants treated with *Azospirillum* biofertilizer were enriched in *Azospirillum brasilense, Bacillus coaculans, Bacillus subtilis, Azotobacter chroococcum* and *Azotobacter vinelandii*. Root tips of phosphobacteria-fertilized mulberry contained *Streptomyces thermonitrificans, Pseudomonas putida, Brevibacillus borslelansis* and *Bacillus stea rothermophilus*. Vermicompost biofertilized root tips had *Bacillus sonoren sis, Bacillus megatherium, Brevibacillus borslelansis, Bacillus subtilis, Bacillus stea rothermophilus, Glucanobacter diazotrophicus, Azotobacter vine landii, Pseudomonas putida* and *Azotobacter chroococcum* (Mary et al. 2015; Hakeem et al. 2016). Similarly, *Listeria monocytogenes* is a food borne pathogen and is the causative agent of listeriosis in humans. It has been observed that *L. monocytogenes* naturally occupies a place in soil as micro flora. This pathogen has been isolated from a variety of garden vegetables including carrots, tomato, lettuce, spinach and radish (Locatellia et al. 2013).

### 2 Effects of Environment on Soil Micro Flora

The variation in the soil microbial community can partly be explained by the alteration in soil chemistry and plants growing in the region (Mitchell et al. 2012). In addition, the environment is also observed to affect the micro floral conditions of the soil. It has been reported that as the moisture content in the environment is decreased, the decomposing activity mediated by the microbes is also diminished. Soil micro flora, hence, undergo stress due to reduction in environmental moisture. Therefore, it can be safely concluded that changes in rainfall pattern result in a change in the carbon and nutrient cycling, owing to the role of soil micro flora in these cycles.
Moreover, the micro flora have an influence on the levels of zinc, copper and total sulphur in soil which ultimately affects the soil pH as well (Sullivan et al. 2013).

Tree species’ transition also has a large impact on the biomass, in general, and soil microflora, in particular (Huang et al. 2013). Application of integrated agricultural management has shown that the amounts of beneficial bacteria, like *Bacillus*, *Streptomyces*, *Paenibacillus* and *Arthrobacter*, are significantly increased in response to these transitions. Contrarily, the colonization of *Ralstonia* is considerably decreased (Wu et al. 2014). Park grass experiment, which has been ongoing in United Kingdom since 1856, helps in measuring the long term response of soil microflora to variations in soil pH and other environmental parameters. The most prevalent microfloral species in these experiments are *Clostridium*, *Rhodoplanes*, *Bacteriodes*, *Paenibacillus*, *Bradyrhizobium*, *Ruminiococcus* and *Mycobacterium*. Based upon these experiments, it has been reported that the soil micro floral diversity is directly dependent upon the plant species richness (Zhalnina et al. 2015).

With respect to the temperature, the warmer the environmental conditions, the greater is the concentration of *Actinobacteria* in soil. Increase in the concentration of ectomycorrhizal fungi is also observed with an increased atmospheric temperature (Deslippe et al. 2012). However, an increase in temperature is negatively related to the concentration of *Gimmatimonadaceae* and *Proteobacteria* in soil. Seasonal and inter-annual variability in chemical resources, temperature and precipitation results in alteration of micro floral fungal community in soil (Burke 2015). Normal field dose of pesticides possesses minor effects on soil micro flora whereas a 10-fold increased dose of pesticides has shown to be detrimental on the microfloral populations (Sofo et al. 2012).

Among the other biological factors, different effects of transgenic plants have been observed on the soil microflora. The natural flora is differentially affected by plants that have been modified to express traits considered beneficial to food industry, phytoparasite resistance and phytopathogen resistance (Turrini et al. 2015). Similarly, adverse effects of pesticides on soil microflora have been observed in a number of studies. Complete degradation of some species of soil micro flora are seen as a result of pesticide application (Singh et al. 2015). On applying cadmium and pyrene on soil, it has been observed that the metabolic quotient, basal respiration and microbial biomass were influenced. The microflora are highly sensitive towards heavy metals including zinc and lead. It has been observed that actinomycses are most sensitive to these metals followed by fungi and bacteria (Jin et al. 2015). These studies helped in concluding that different physicochemical characteristics of soil change the physiological response of soil micro flora (Lu et al. 2013).

In another study, the effect of intercropping of sugarcane and soybean cultivation on soil micro flora was tested. It was observed that in comparison to the monocultures, microbial concentrations in polycultures were relatively quite high. In lands where only sugarcane was cultivated, after microbial enrichment, the bacterial, fungal and actinomycses population was increased by 42.62%, 14.5% and 78.5% respectively.
respectively. However, bacteria, fungi and actinomycyes recorded a growth of 188%, 183% and 73% in sugarcane-soybean intercropped systems. Therefore, it can be established that the growth of sugarcane with soybean is an excellent agricultural management practice (Li et al. 2013).

Furthermore, the application of photosynthetic bacterial inoculum in agricultural lands tends to increase both the quantity and diversity of soil micro flora. The number of fungi, bacteria and actinomycyes was significantly high in soil treated with photosynthetic bacteria in comparison to the untreated soil (Fang et al. 2014). Recently, the effect of grafted tomato on the quantity of soil microflora was studied. The study showed that the rhizospheric microbial population including soil bacteria, cellulose decomposing bacteria, actinomycyes, aerobic abigenous azotobacter, ammonifying bacteria and nitrifying bacteria was significantly increased. However, the fungal population was decreased (Na et al. 2014). Treatment of coal mine disturbed soil with arbuscular ecto-mycorrhizal fungi has also been explored. The study has led to establish the symbiotic relationship of these fungi with plants. Moreover, an increase in the number of other soil bacteria, actinomycyes and fungi has also been reported (Jin et al. 2014). Based upon these studies, it can be safely concluded that a number of environmental factors, both biotic and abiotic, help in the establishment of the complex, yet highly beneficial, relationship between various plants and microbes.

3 Microflora of Soil

A number of experiments have been carried out to ascertain the relative population of various microbes in soil. In one such study, the soils of tomato greenhouse were collected to determine the microflora in it. It was observed that the bacteria were the highest in number whereas fungi had the lowest number. Similarly, soil experiments were conducted over a long term to measure the changes in soil microflora as a function of time. A significant decrease was seen in these microfloral communities over a period of 10–20 years. Similarly, on studying the effect of the soil thickness, microflora was largely present in upper 20 cm thick layer. Concentrations of actinomycyes and fungi were observed to decrease with increasing depth of soil (Tang 2012). When quantity of soil micro flora was determined in different types of tobacco soil, it was seen that quantities of bacteria, actinomycyes and fungi as well as nitrogen fixing microbes was higher in yellow brown soil as compared to purple soil (Xu et al. 2013). Majority of micro flora found in soil rhizosphere is bacteria but significant number of fungi are also present (Bhatt et al. 2015; Hakeem et al. 2016). The major types of microbes and various factors affecting their relative abundance have been discussed in the subsequent sub-sections.
3.1 Actinomyces

Actinomyces, especially *Streptomyces*, are largely present in soil as natural microflora. It has been observed that the ultrasonic treatment of soil samples resulted in an increase in the quantity as well as the type of actinomyces. Quantity of actinomyces in soil increased by 280% whereas that of *Streptomyces* increased by around 375%, as a result of 50 s of treatment with ultrasonic. In comparison, the ultrasonic treatment for 20 s resulted in an increase in actinomyces concentration by merely 12.07%. However, increasing the treatment duration to 100 s also had a negative effect on the Actinomyces species where a population growth of only 40% was noted (Mao et al. 2013).

Root exudates of Chinese onion cultivar were applied to study their effects on cucumber seedling growth. It was reported that there was a significant increase in the concentration of actinomyces in the treated soil (Yang et al. 2013). Application of biological organic fertilizer, also, causes an increase in the number of actinomyces and the structure of soil microbial community (Yuan et al. 2011). Actinomyces also stimulate the soil functionality by enhancing the concentration of different soil enzymes as they are a great source of keratinases, lipases, xylanases and pectinases (Suneetha and Khan 2010).

3.2 Bacteria

Bacteria present in soil play an essential role in maintaining a number of biogeochemical cycles. They occupy small spaces present between the pores in soil. The impact of bacteria and their function is dependent on their spatial arrangement (Archana et al. 2015). Much similar to the actinomyces, the application of root exudates of Chinese onion cultivar enhances the concentrations of bacteria in the soil. The different bacterial species increased as a result of this treatment include various strains of genus *Anaerolineaceae*, *Actinobacteria* and *Proteobacteria* (Yang et al. 2013). Soils treated with organic fertilizers are observed to possess higher quantities of beta lactam resistant bacteria in comparison to those treated with inorganic fertilizers. Similarly, the number of *Pseudomonas* species is also increased in organic fertilizer treated soil. Manure treatment also increases the concentration of *Psychrobacter pulmonis* and *Janthinobacterium* species. This helps in hypothesizing that the application of organic fertilizers can lead to an increase in the concentration of antibiotic resistant strains of bacteria (Udikovic-Kolic et al. 2014). However, further experimentation in this regard is needed. The soil is also enriched in bacterial species including *Burkholderia terrestris*, *Burkholderia humi*, *Burkholderia udeis*, *Burkholderia choica* and *Burkholderia telluris* (Vandamme et al. 2013).
3.3 Fungi

The most common form of fungi found in soil are the arbuscular mycorrhizal fungi. These fungi are known to form mutualistic ecto-mycorrhizal associations with plants aiding their growth and development. Fungi have a prominent role in the ecosystem as they serve as mutualists, decomposers and, oftentimes, as pathogens (Taylor et al. 2014). The soil population of fungi and fusarium is significantly increased in the rhizosphere after treatment with root exudates of Chinese onion cultivar in cucumber fields (Yang et al. 2013). Similarly, on analyzing the soil samples from bat hibernacula in United States, it was reported that *Geomycetes* species of fungi was the most significant among all the fungal isolates from the respective soil. The fungi account for almost 33% of all fungal species found in soil (Lorch et al. 2012). Nearly 90% of terrestrial plants are in symbiotic relationship with soil fungi (Miransari 2011). These fungi play an important role in the degradation and decomposition of organic matter and formation of soil infrastructure. Moreover, it has been reported that when these symbionts are inhibited, other beneficial soil microbes serve the same function to maintain the food web (Helfrich et al. 2015).

Arbuscular mycorrhizal fungi are well known soil organisms. They develop mutualistic relationship with plants by promoting the phosphorus uptake by the plant roots (Sharma and Buyer 2015). In a recent study, different types of fertilization treatment were provided to soil cultivated with sage to determine the distribution of arbuscular mycorrhizal fungi (AMF) in soil ecosystem. It was observed that the AMF colonization of sage was higher in case of plants treated with manure fertilization as compared to those treated with mineral fertilization. Moreover, *Olpidium* species and dark septate endophytes have also been isolated from the manure-treated fields (Zubek et al. 2012).

One of the most common genus of soil fungi is *Aspergillus*. It has been found that even after the treatment of soil with 600 ppm of cypermethrin, a highly toxic pesticide, at least 10 fungal species of *Aspergillus* tend to survive in the treated soil. *A. flavus* and *A. candidus* are the major isolates among these varieties. At 1000 ppm of cypermethrin, *A. flavus* is the single most dominant species. However, *A. terrae*, *A. awamori* have also been found to inhabit these soils (Sethi et al. 2015). Similarly, twelve different fungal species were isolated from crop fields of Mysore district in India. These included *A. niger*, *C. lunata*, *A. terreus*, *F. solani*, *A. fumigatus*, *F. oxysporum*, *A. flavus*, *T. harzianum*, *R. stolani*, *T. viride*, *P. chrysogenum* and *P. fumiculosum*. In general, *Aspergillium* species and *Penicillium* species are most dominant fungal species in agricultural soil. The primary reason for their high population density is their defense and survival mechanisms. *Aspergillus* produces toxins and *Penicillium* produces penicillin antibiotics which cause a retardation in the growth of other microbes. Moreover, they are also involved in the biodegradation of compounds and enhancement of soil nutrients (Sharma and Raju 2013).
3.4 Algae

A diversity of cyanobacteria have been isolated from rice fields. In a recent study carried out in Andhra Pradesh, India, at least 38 different species of the algae were isolated from rice paddies. They included both heterocystous and non-heterocystous. Heterocystous are specialized cells that are produced under nitrogen-stressed conditions. In rice fields, cyanobacteria are the most abundant nitrogen fixing organisms (Danaboyina and Sivakumar 2013). Unicellular, filamentous and colonial, all types of cyanobacteria have been found in these fields. However, a number of physiological parameters affect the nature and concentration of cyanobacteria in soil. These include temperature, moisture content and soil pH.

4 Advantages of Microflora

The microflora of soil is rich in a variety of organisms. These can be exploited to attain various benefits including production of enzymes, industrial products, secondary metabolites and biopharmaceuticals.

4.1 Living Soil

Living soil is a term coined for the soil that contains diverse microflora. Microorganisms especially filamentous fungi are one of the most important constituents of a well living soil. These fungi are prominent sources for of food as well as biosynthesis of a variety of products including antibiotics and other medicinal agents (Sethi et al. 2015). The soil microorganisms contribute to the modification of biological, physical and chemical properties of the soil. The health of soil is, hence, maintained by this microflora. The characteristic earthy smell of soil is also a gift of bacterial diversity present in it (Bhatt et al. 2015).

4.2 Growth and Development of Plants-Agricultural Sustainability

The sustainability of the environment and agriculture depends upon the soil microflora as well. Microflora have been observed to enhance the productivity of agricultural system in a natural and organic way. Soil microbial communities, hence, have multiple positive impacts on growth and development of plants (Singh et al. 2011). Mycorrhizal fungi are one of the groups of organisms living in rhizosphere. These fungi form associations with plants and contribute to their growth by symbiosis. It
is due to mycelium of these fungi that the absorption of nutrients and water from soil is enhanced. With this plant-fungal association, soil micro flora plays a significant role and bind to these fungi to complement their development. The interaction which is formed between ectomycorrhizal fungi and other microbes of the soil flora leads to the development of a sustainable microbial complex ecosystem which helps in proper growth and development of plants. This, ultimately, enhances yield and quality of crops (Duponnois et al. 2012).

Fungi improve the availability of soil nutrients for plants. It also helps in the degradation of cellulose to release carbohydrates including pectin, hemicelluloses and starch (Bhatt et al. 2015). Soil micro flora have a direct relation with soil fertility (Marathe et al. 2012). Soil bacteria are the only known life forms which perform nitrogen fixation during nitrogen cycle (Bhatt et al. 2015). They are, hence, excellent candidates to increase zinc bioavailability to plants. Improvement of microbial activity in soil yields free zinc which can then be easily absorbed by plants (Imran et al. 2014). The soil microbes are involved in a number of biochemical transformations and mineralization activities taking place in soil (Yadav 2014).

4.3 Decomposition of Organic Matter

Soil microorganisms are known to possess a primary role in environment because they degrade plant and animal residues. Hence, global cycling of nutrients is possible due to diverse micro flora of soil (Sonia and Saksham 2013). Soil natural flora like Pseudomonas and Trichoderma are, also, involved in the control of a large variety of harmful plant pathogens including Rhizoctonia, Pythium, Fusarium and Phytophthora (Jagtap 2012).

4.4 Bioremediation of Soil

Soil micro flora have been observed to convert pesticidal toxic compounds to nontoxic forms. Bacteria present in soil, for instance Bacillus stearothermophilus, are involved in detoxification of largely used pesticides including chlorpyrifos (Savitha and Raman 2012). Pristine and oil contaminated soils naturally have micro flora which utilize hydrocarbons and possess heavy metal resistance. Bacterial species including Chromobacterium orangum, Bacillus subtilis, Enterobacter aerogenes, Corynebacterium pseudotuberculosis, Nocardia paraffinea, Nocardia coralline, Micromonaspora chalcea, Streptomyces flavovirens, Alcaligenes faecalis and Brevibacterium linens are of prime importance in this respect. These species degrade aromatic hydrocarbons and yield nutrients that can be utilized by plants. Indigenous soil micro flora are quite diverse. Due to these facts, the development of genetically modified micro floral organisms for bioremediation of soil is not needed (Ali et al. 2012). Addition of organic fertilizers in soil enhances the amount of soil micro flora.
The process, hence, causes a decrease in the soil pollution. Bioremediation of polluted soil can, therefore, be performed using microbe-mediated organic fertilization process (Osaigbovo et al. 2013).

5 Deteriorating Effects on Soil Micro Flora by Anthropogenic Activities

Increased herbicide concentrations decrease the microbial concentrations in soil. However, with the degradation of applied herbicides, microbial populations are greatly enhanced. After 60 days of application of herbicides, soil micro flora gain their pre-treatment concentrations (Bera and Ghosh 2013a, b). Frequent long term application of pesticides on agricultural lands might cause severe detrimental effects on soil micro flora. Organophosphorus pesticides are one of the most widely used pesticides. They have long term degrading effects on soil microbes. Application of pesticides, like atrazine, greatly reduces the concentration of cellulose producing bacteria. Similar adverse effects have been noted with the use of other pesticides including paraquat and trifluralin (Sethi et al. 2015).

Though nearly all kinds of herbicides possess detrimental effects on soil microbes yet their persistent application has been associated with the development of resistance in these microbial species. This, ultimately, results in the recovery of microbial count in the soil (Bera and Ghosh 2013a, b). Chemical methods of controlling insects, fungi and herbs are gaining fame because they are highly efficient. They increase the crop yield by protecting them from pests. Repetitive, high-concentration treatment and the resulting accumulation of these chemicals in soil, however, poses significant harmful effects on micro flora even leading to their extinction in certain cases (Das et al. 2014). Among the biological threats, the soil micro flora are also degraded by continuous cultivation of Bt cotton in a particular area. Bt toxin which leaches in to the rhizosphere kills the microorganisms in soil (Marathe et al. 2012). Similarly, bacterial population in soil cultivated with soybean is inhibited by pesticide alachlor. Excessive use of chemical fertilizers and compounds might contribute to generating a biological imbalance. This makes the soil prone to degradation. Rate of biodegradation of soil is directly related to the diversity and density of microbial community in it (Yadav 2014).

6 Conclusion and Future Prospects

A variety of microbes are found in soil. It can be rightly said that microbial communities are the most significant component of soil. The microfloral diversity in soil is more diverse than any other environmental habitat (Bhatt et al. 2015). Richness of soil micro flora is dependent on cropping pattern, soil moisture and organic
matter present in soil (Marathe et al. 2012). Integrated crop management techniques use fungicides, insecticides and herbicides in a manner that once they deteriorate soil micro flora, the concentration of micro flora rise again with the passage of time. These approaches can be used for the maintenance of the living soil (Jagtap 2012). Modern day agriculture is not possible without exploiting the soil micro flora. Hence, this microbial community needs to be protected. Therefore, biological methods need to substitute the currently prevalent chemical techniques. This would help in prevention of soil degradation and preservation of microbial communities residing in the soil. Moreover, composition of soil microfloral communities also needs to be monitored. Further research on soil microflora is required to get maximum benefits from them. They can then be exploited to attain pharmaceutical drugs and super foods. They can also be employed for the biofertilization process. Therefore, the scientific expertise of botany, microbiology, molecular biology, molecular genetics, biotechnology, pharmaceutics and systems biology need to be exploited for attaining the maximum benefits from the plant-microbe interactions.

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