Chapter 12
Beneficial Microorganisms for Sustainable Agriculture

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Abstract There was a desperate need for food to recover the economy of the 1950s and 1960s. Farmers all over the world were advised to rely on intensive production methods and synthetic pesticide inputs to increase the productivity. No doubt, these chemical-based agricultural practices substantially increased crop yield. However, indiscriminate use of agrochemicals have contributed significantly to the environmental pollution and adversely affected human and animal health. In addition, the increasing cost of these agrochemicals has continued to lower the farmer's net cash return. The global use of synthetic pesticides at the start of this millennium exceeded 2.5 million tons per year. A growing worldwide concern for these problems has motivated researchers, administrators, and farmers to seek alternatives to chemical-based, conventional agriculture. One such product is effective microorganisms (EM) developed by Japanese scientists. Effective microorganisms are a mixed culture of beneficial and naturally occurring microorganisms, such as species of photosynthetic bacteria (*Rhodopseudomonas palustris* and *Rhodobacter sphaeroides*), lactobacilli (*Lactobacillus plantarum*, *L. casei*, and *Streptococcus lactis*), yeasts (*Saccharomyces* spp.), and Actinomycetes (*Streptomyces* spp.). These beneficial microorganisms improve crop growth and yield by increasing photosynthesis, producing bioactive substances such as hormones and enzymes, controlling soil diseases, and accelerating decomposition of lignin materials in the soil. Experiments conducted on various agricultural crops in different parts of the world have shown good prospects for the practical application of these beneficial microorganisms in improving crop yield and soil fertility. Application of beneficial microorganisms generally improves soil physical and chemical properties and favors the growth and efficiency of symbiotic microorganisms such as nitrogen fixing rhizobia and arbuscular mycorrhizal (AM) fungi. Nonetheless experiences of some researchers revealed that the effect of these microorganisms on crop growth and yield was usually not evident or even negative in the first test crop. However, this adverse effect can be overcome through repeated

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applications of these microorganisms. Research on these microorganisms has shown that crop yields tend to increase gradually as subsequent crops are grown. Foliar application of beneficial microorganisms avoids many of the biotic and abiotic factors and constraints of the soil environment, and thus increases the crop growth and yield significantly. Application of beneficial microorganisms also reduces seed bank of weeds in agricultural soils by enhancing the rate of weed seeds germination. There are reports of management of various fungal and bacterial pathogens as well as insect pests due to application of beneficial microorganisms. These microorganisms have shown a great promise in dairy wastewater treatment. They can reduce NH_3 concentration in poultry manure up to 70% possibly by transforming NH_4^+ to NO_3^-. Research conducted so far concludes that benefits of beneficial microorganisms can be best exploited through their repeated applications for few years in combination with organic amendments and applying them as foliar spray. Integrated use of organic matter plus beneficial microorganisms with half mineral NPK can yield equivalent to that of full recommended NPK fertilizers dose. Beneficial microorganisms can also be used for wastewater treatment, pest and disease management, and to reduce the abiotic stresses on crop growth and yield.

Keywords Biofertilizer • effective microorganisms • nature farming • sustainable agriculture

12.1 Introduction

Fertilizers as a source of plant nutrients and pesticides as plant protection measures are being used to increase the crop production. The global use of synthetic pesticides at the start of this millennium exceeded 2.5 million tons per year (Bhanti and Taneja 2007). However, imbalance and frequent use of these agrochemicals have polluted the environment to a great extent. There is a growing concern that food produced under such farm management may not be safe or of good quality. This has shifted the scientific approach toward some alternative measures (Shaxson 2006). In the recent past, some successful efforts have been made to at least partially substitute agrochemicals with natural substances to minimize the bad effects of the former (Kannaiyan 2002). One such effort was made by Dr. Teruo Higa, Professor of Horticulture, University of the Ryukyus, Okinawa, Japan, who conducted pioneering work in advancing the concept of “Effective Microorganisms” (EM) (Higa 1991). EM consists of mixed cultures of beneficial and naturally occurring microorganisms that can be applied as inoculants to increase the microbial diversity of soils and plants. EM is not a substitute for other management practices. It is, however, an added dimension for optimizing our best soil and crop management practices such as crop rotations, use of organic amendments, conservation tillage, crop residue recycling, and biocontrol of pests. If used properly, these beneficial microorganisms can significantly enhance the beneficial effects of these practices (Higa and Wididana 1991).

12.2 Beneficial Microorganism's Cultures

First solution of beneficial microorganisms contained over 80 microbial species from 10 genera, isolated in Japan. However, with time the technology was refined to include predominant populations of lactic acid bacteria and yeast and smaller numbers of photosynthetic bacteria, actinomycetes, and other types of microorganisms. All of these are mutually compatible with one another and can coexist in liquid culture (Higa and Parr 1994). The functions of principle microorganisms in EM are as follows.

12.2.1 Photosynthetic Bacteria

Photosynthetic bacteria include *Rhodopseudomonas palustris* and *Rhodobacter sphaeroides.* These bacteria are a group of independent, self-supporting microbes. These are considered the pivot of EM activity and support the activity of other microorganisms in EM culture. They synthesize useful substances from secretions of plant roots, organic matter, and harmful gases such as hydrogen sulfide, by using sunlight and the heat of soil as sources of energy (Kim et al. 2004). The useful substances produced by these bacteria include amino acids, polysaccharides, nucleic acids, bioactive substances and sugars, all of which promote plant growth and development (Higa 2000). The metabolites developed by these microbes are absorbed directly by plants (Kim and Lee 2000; Ranjith et al. 2007).

12.2.2 Lactic Acid Bacteria

Lactic acid bacteria include *Lactobacillus plantarum*, *L. casei*, and *Streptococcus lactis*. They produce lactic acid from sugars and other carbohydrates produced by photosynthetic bacteria and yeasts (Hussain et al. 2002). Lactic acid is a strong sterilizing compound, suppresses harmful microorganisms such as *Fusarium* and enhances decomposition of organic matter (Higa and Kinjo 1991). These bacteria promote the fermentation and decomposition of materials such as lignin and cellulose, thereby removing the undesirable effect of undecomposed organic matter (Gao et al. 2008; Valerio et al. 2008).

12.2.3 Yeasts

Yeasts include *Saccharomyces cerevisiae.* Yeasts synthesize useful substances required for plant growth from amino acids and sugars secreted by photosynthetic bacteria, organic matter, and plant roots (Higa 2000). The bioactive substances such as hormones and enzymes produced by yeast promote active cell and root division. The secretions are also useful substrates for other microorganisms in EM culture viz. lactic acid bacteria and actinomycetes (Hussain et al. 2002).

In the beginning, five formulations of beneficial microorganisms were developed starting from EM_1 to EM_5. EM_1 had predominantly filamentous fungi that are heat resistant and hasten the decomposition of organic amendments. It was initially developed for preparing compost quickly but it is not produced any more. EM_2 is used primarily to protect plants from soil-borne pathogens, diseases, and insects. It contains predominantly the species of genus *Streptomyces*, which produces antibiotics that suppress harmful microorganisms. It also contains smaller numbers of photosynthetic bacteria, yeast, and molds. EM_3 is comprised predominantly of photosynthetic bacteria with smaller numbers of yeast and actinomycetes. It enhances the growth, yield and quality of crop, and improves soil physical properties (Anonymous 1995). EM_4 consists mainly of the lactobacilli with smaller number of photosynthetic bacteria, *Streptomyces* spp. and yeast. It increases the availability of nutrients to plants by enhancing the decomposition of organic wastes and residues. It also suppresses the activity of harmful insects and pathogenic microorganisms (Sajjad et al. 2003). EM_5 is prepared by mixing EM_2, EM_3, and EM_4. It is used to suppress pathogens and to ward off harmful insects. It is especially used for cultivating fruit trees and vegetables (Anonymous 1995).

EM is often inoculated to organic matter fermented and the mixture is called EM Bokashi. It can improve the ability of microorganisms to break down organic matter, thereby providing plant nutrients to make better yield and quality (Xu et al. 2000; Yan and Xu 2002).

In Pakistan, EM is being produced at Nature Farming Research Center, Faisalabad and is available in the form of EM-Bioab, EM-Biovet, and EM-Biocontrol. EM-Bioab is used in agricultural crops along with organic manures as a substitute of chemical fertilizers. EM-Biovet is used in livestock and poultry production while EM-Biocontrol is used in crops, vegetables, and orchards for prevention and remedy of diseases and insect pest attack (Hussain et al. 2002).

12.3 Soil Application of Beneficial Microorganisms

12.3.1 Positive Effects on Plant Growth

Experiments conducted throughout Japan as well as in many other countries have shown good prospects for the practical application of beneficial microorganisms. Research has shown that the inoculation of these beneficial microorganisms cultures to the soil/plant ecosystem can improve soil quality, soil health, and yield and quality of crops.

12.3.1.1 Cereal Crops

Most of the research work on beneficial microorganisms technology has been conducted on cereals crops, particularly rice and wheat. Hussain et al. (1999, 2000) conducted a long-term field experiment in Pakistan to determine the agronomic and economic merits of beneficial microorganisms in a rice–wheat cropping system. They found that when NPK fertilizers and organic manures were combined with EM, higher straw and grain yields were obtained as compared to corresponding treatments without EM application for both the test crops. Beneficial microorganisms applied in combination with NPK, green manure, and farmyard manure caused a significant increase in nutrient uptake by the grains and straw of each crop. The average net profit from rice and wheat production using beneficial microorganisms was US$ 44.9 and 62.35 ha^{-1}, respectively. Corales et al. (1997) studied the effect of beneficial microorganisms in transplanted and wet direct seeded rice. Results showed that the reduced inorganic fertilizers (by 25–50%) in combination with EM or EM Bokashi gave comparatively better results in increasing yield of rice as compared to inorganic fertilizers alone. Xu (2000) conducted a study under glass house to determine the effects of beneficial microorganisms and various organic amendments on the growth, photosynthesis, and yield of sweet corn, compared with chemical fertilizers. Beneficial microorganisms applied with organic fertilizers promoted root growth and activity, and enhanced photosynthetic efficiency and capacity, which resulted in increased grain yield. This was attributed largely to a higher level of nutrient availability facilitated by beneficial microorganisms application over time. Similar experiments conducted in many other countries including China, Japan, Vietnam, Korea, Nepal, and Bangladesh have also revealed increase in crop growth and yield of rice and/or wheat when beneficial microorganisms application was carried out in combination with various organic amendments and different doses of inorganic fertilizers (Lee 1994; Ta and Chanh 1996; Iwaishi 2000; Sherchand 2000; Chowdhury et al. 2002).

12.3.1.2 Vegetable Crops

The demand for organically grown food crops has increased markedly in the recent years as consumers have become more concerned about pesticide residues in the human diet. Consequently, organic crop production system is gaining popularity worldwide. Generally, farmers utilize available crop residues, animal manures and off-farm vegetative materials as organic amendments to supply plant nutrients and maintain soil productivity. However, the yields of food crops grown in these systems are generally low. Use of beneficial microorganisms has been shown to increase the yield and quality of food crops in organic farming systems. Daly and Stewart (1999) investigated the effect of beneficial microorganisms on vegetable crops on organic farms in Canterbury, New Zealand. They found that beneficial microorganisms plus molasses increased the onion yield by 29%, pea yield by 31%, and sweet corn cob weights by 23%. Daiss et al. (2008) reported that Swiss chard

(*Beta vulgaris* L.) treated EM plus EM Bokasi had higher phosphorus and magnesium contents than control plants. Enhanced nutrient utilization efficiencies of the plants in beneficial microorganisms-applied treatments have also been reported in capsicum (*Capsicum annuum* L.) and cowpea (*Vigna unguiculata* L.) by Sangakkara et al. (1998). Sangakkara (1998) suggested that yields of sweet potato (*Ipomoea batatas* L.) and bush bean (*Phaseolus vulgaris* L.) were significantly increased by the application of beneficial microorganisms to traditional organic system in Sri Lanka. Increase in yield due to beneficial microorganisms application has also been reported for certain other vegetables, viz. radish, cabbage, and lettuce (Lee 1994; Naseem 2000).

12.3.1.3 Fruit Crops

Higa (1988) conducted experiments on various horticultural crops and concluded that beneficial microorganisms can increase the yield and quality of fruits. Similarly, Paschoal et al. (1998) have reported an increase in the yield of oranges owing to the application of beneficial microorganisms. Xu et al. (2000) reported that beneficial microorganisms inoculation to both Bokashi and chicken manure increased photosynthesis and fruit yield of tomato plants. Application of beneficial microorganisms also increased vitamin C concentration in fruits from all fertilization treatments. They concluded that both fruit quality and yield could be significantly increased either by inoculation of beneficial microorganisms to the organic fertilizers or application directly to the soil. According to Joo and Lee (1991), with beneficial microorganisms application, yield of citrus increased significantly as compared to traditional method of farming. Fuel cost index was also declined with beneficial microorganisms farming system. Wibisono et al. (1996) obtained a significant effect on plant height, number of shoots, and number of leaves in *Citrus medica* when beneficial microorganisms were applied along with rice straw at 2.5 t ha^{-1}. Tokeshi and Chagas (1997) studied the hormonal effect of beneficial microorganisms on citrus germination. They concluded that beneficial microorganisms have hormonal action as gibberellic acid. The potential of seedling survival and vigo r was measured by emergence speed. There was rapid emergence speed with beneficial microorganisms as compared to control.

12.3.1.4 Other Crops

Application of beneficial microorganisms is also known to enhance crop growth and yield in certain other crops of economic importance such as cotton, coffee, and sugarcane. Khaliq et al. (2006) conducted a field experiment to determine the effects of integrated use of organic and inorganic nutrient sources with and without beneficial microorganisms on growth and yield of cotton. They observed that organic material and beneficial microorganisms did not increase the yield and yield-attributing components significantly but integrated use of both resulted in a 44% increase over control. Integrated use of organic matter plus beneficial microorganisms with half mineral NPK yielded equivalent to that of full recommended NPK fertilizers dose. Economic analysis suggested the use of half mineral NPK with organic matter and EM saves the mineral N fertilizer by almost 50% compared to a system with only mineral NPK application. Increased yield in sugarcane due to beneficial microorganisms application in combination with various organic materials have been reported. However, the increase in yield over control was not as much pronounced as in other crops (Punyaprueg et al. 1993; Zacharia 1995). Chagas et al. (1997) suggested that coffee plant propagation could be substantially improved by replacing the chemical fertilizers with Bokashi plus EM. Beneficial microorganisms application in combination with cow dung significantly increased the germination and physical growth parameters including shoot and root length and biomass, vigor index, collar diameter, and leaf number of *Albizia saman*, a medium-to-large-sized tree native to Central America, West Indies, and Guyana and is widely distributed in the tropical forests of Asia (Khan et al. 2006).

12.3.2 Negative or No Effects on Plant Growth

Majority of the scientists who are engaged in promoting EM technology have no doubt that plant growth is just as good or better and the quality of plant products is superior to conventional farming (Daly and Stewart 1999; Hussain et al. 1999; Yamada and Xu 2000; Iwaishi 2000; Khaliq et al. 2006; Khan et al. 2006). In contrast to that, experience of some workers revealed that the effect of beneficial microorganisms on crop growth and yield was usually not evident or even negative particularly in the first test crop. Rashid et al. (1993) noted that beneficial microorganisms applied along with farmyard manure did not improve wheat grain yield and N uptake in wheat over farmyard manure alone and standard dose of chemical fertilizers. Similarly, Yousaf et al. (1993) did not find any significant effect on crop growth in maize when beneficial microorganisms were applied along with different soil amendments such as farmyard manure, poultry manure, sewage sludge, and NPK fertilizers. Bajwa et al. (1998a) noted an inhibitory effect of EM application on plant growth in *Brassica campestris* L. In another study, Bajwa et al. (1999a) observed similar effect of beneficial microorganisms application on root and shoot biomass production in *Trifolium alexandrinum* L. Priyadi et al. (2005) conducted a field experiment to elucidate the effect of chicken manure and beneficial microorganisms on the yield of corn and chemical and microbial properties of two types of acidic wetland soils. The results showed that the interaction between soil types and chicken manure application affected the corn yield, while beneficial microorganisms had no effect. Bajwa et al. (1995a) showed that application of beneficial microorganisms in heat-sterilized soil induced a significant positive effect on crop growth and yield in wheat while adversely affected crop growth in non-sterilized soil, indicating that microorganisms in EM solution had to face a competition with soil indigenous microflora. However, in a recent study, Javaid et al. (2008) reported negative or no effects of EM application on yield of wheat both in heat-sterilized and unsterilized soils.

It has been found that in beneficial microorganisms treated soils, generally crop yields tend to increase gradually as subsequent crops are grown. Sangakkara and Higa (1994a) studied the effect of beneficial microorganisms application on growth and yield of eggplant (*Solanum melongena* L.), capsicum (*Capsicm annum* L.), and tomato (*Lycopersicum esculentum* L.) for two seasons. During the first season, the effect of beneficial microorganisms application was insignificant while in the second season significant effect of these microorganisms for increasing crop growth and yield was evident. Sangakkara et al. (1998) conducted a 3-year study to evaluate the efficiency of beneficial microorganisms and organic matter on crop growth and nutrient uptake in capsicum and cowpea (*Vigna unguiculata* L.). They reported that the effect of beneficial microorganisms application was pronounced in the second and third year as compared to the first one. Similar responses of crop growth to beneficial microorganisms application have also been reported in wheat (Javaid et al. 2000a), *Phaseolus vulgaris* L. (Javaid et al. 2002) and pea (Javaid and Bajwa 2002). According to Kinjo et al. (2000) the lack of consistency in results of the experiments regarding beneficial microorganisms application may be due to variable cultural conditions employed in previous studies. They tested this hypothesis by applying beneficial microorganisms and chemical fertilizers in soils with different cultural practices. One soil was collected from an organic farm and the other from a conventional farm. They observed useful effects of beneficial microorganisms on yield of radish in soil collected from organic farm but these effects were not evident in the soil collected from conventional farm. Imai and Higa (1994) stated that the observed decline in crop yields can often be attributed to the fact that soils, where conventional farming is practiced, have become disease-inducing or putrefactive soils from long-term use of pesticides and chemical fertilizers. Consequently, it takes time to establish a disease-suppressive or zymogenic soil. Until this conversion process is completed, it is virtually impossible to exceed crop yields that were obtained with conventional farming methods.

12.3.3 Effect on Soil Properties

Studies have shown that beneficial microorganisms significantly improved certain physical and chemical properties of the soil. Higa (1989) found that the application of beneficial microorganisms to soil increased NO_3^- concentration from 4.5 to 5.1 mg 100 g^{-1} dry soil. Zhao (1998) noted that application of beneficial microorganisms significantly increased the available nutrients, organic matter, and total nitrogen and lowered the C:N ratio in the soil. Paschoal et al. (1998) showed that beneficial microorganisms application in *Citrus* agro-ecosystem significantly increased soil organic matter content, level of some macronutrients including Ca, Mg, and K; soil cation exchange capacity, and lowered soil base saturation. Park (1993) reported that application of manure and beneficial microorganisms improved the topsoil by reducing bulk density and dispersion ratio thus made the soil less compact and more resistant to erosion. Improvements in soil physical properties like bulk density and hydraulic conductivity due to beneficial microorganisms application has also been reported by Hussain et al. (1994). Lee (1994) reported the increased levels of available P_2O_5, Ca, and Mg in the soil due to EM solution and EM plus compost application. Beneficial microorganisms application is known to enhance nodulation and nitrogen fixation efficacy of soil rhizobia (*Rhizobium/Bradyrhizobium*) in leguminous crops (Sangakkara and Higa 1994b; Javaid et al. 2000b; Yan and Xu 2002; Javaid 2006). Similarly, beneficial microorganisms application also stimulated development and functioning of other soil-borne symbiotic microorganisms such as arbuscular mycorrhizal fungi and thereby enhance soil fertility and plant nutrient acquisition (Javaid et al. 1995; Mridha et al. 1997; Bajwa et al. 1999b).

12.4 Foliar Application of Beneficial Microorganisms

Foliar application of beneficial microorganisms avoids many of the biotic and abiotic factors and constraints of the soil environment. Farmers of Indonesia have learnt to use beneficial microorganisms on vegetable crops much like a foliar fertilizer, akin to the foliar application of micronutrients. On-farm tests and demonstrations have shown that foliar applications of beneficial microorganisms can increase the growth and yield of vegetable crops in a relatively short time, even though no organic amendment is added to the soil (Widdiana and Higa (1998). These authors conducted a field study to determine the effects of foliar-applied beneficial microorganisms on the production of garlic, onion, tomato, and watermelon, compared with the recommended application of chemical fertilizers. Foliar solutions of beneficial microorganisms at a concentration of 0.1%, 0.5%, and 1% were applied at 1- and 2-week intervals. Most vegetable yields were generally higher with foliar-applied beneficial microorganisms compared with the chemical fertilizer control. The highest yield of garlic was obtained with beneficial microorganisms at 0.1% applied at 1-week intervals, and was 12.5% greater than the fertilized control. The highest yield of onion and tomato resulted from weekly applications of beneficial microorganisms at 1%. Yield for these two crops were 11.5% and 19.5% higher than the fertilized control. However, there was no significant increase in watermelon yield from foliar application of beneficial microorganisms at any dilution level.

Xiaohou et al. (2001) conducted various studies in China to investigate the effect of foliar application of beneficial microorganisms on yield and quality of various crops. He reported that in field trials, sprinkling of 0.1% beneficial microorganisms solution improved the quality and enhanced yields of tea, cabbage, and sugar corn by 25%, 14%, and 12.5%, respectively. Yousaf et al. (2000) investigated the effect of seed treatment and foliar application of beneficial microorganisms on growth and yield of two varieties of groundnut (*Arachis hypogaea* L.). They recorded an 18% and 17% increase in yield in varieties CG-2261 and CGV-86550 due to seed treatment, and 58.1% and 58.3% increase due to combined application of seed treatment plus foliar application over control, respectively.

The type of soil amendment also affects the performance of foliar applied beneficial microorganisms. However, the mechanism is not known so far. Javaid (2006) compared the effect of foliar and soil application of beneficial microorganisms on growth and yield of pea (*Pisum sativum* L.) in soils amended with NPK fertilizers, farmyard manure, and green manure. The results showed that soil and soil plus foliar application of beneficial microorganisms either exhibited insignificant effect or suppressed the plant growth and yield. However, foliar application alone significantly enhanced shoot biomass by 70% in NPK treated soil. Similarly, foliar application of beneficial microorganisms significantly increased the number and biomass of pods by 157% and 266%, and 126% and 145% in NPK fertilizers and green manure amended soils, respectively.

12.5 Effect of Beneficial Microorganisms on Symbiotic Microorganisms

12.5.1 N_2-Fixing Rhizobia

Legumes are unique among crop plants in that they are capable of contributing a limiting resource to the agroecosystem by fixing N_2. The history of crop husbandry is replete with examples of yield enhancement of a non-legume crop by legumes grown either in rotation (Voss and Shrader 1984) or as multicrops (Heichel and Henjum 1991). Historically, these management practices were the mainstay of N replacement in cropping systems until the advent of economical commercial N fertilizers (Heichel and Barnes 1984). Soil bacteria belonging to the family *Rhizobiaceae* and falling in the genera *Rhizobium*, *Bradyrhizobium*, *Sinorhizobium*, *Mesorhizobium*, *Allorhizobium*, and *Azorhizobium* are capable of forming nodules on leguminous plants (Wei et al. 2008). The species of *Azorhizobium* form nodules on the stems of tropical legumes *Sesbania* and *Aeschynomene* while species of other genera form nodules on roots of leguminous plants. While colonizing the nodules, the bacteria develop into N_2-fixing bacteroids providing the host plant with NH_4^+ as nitrogen source. In return, the plant supplies the bacteroids with vital organic compounds. During the establishment of active symbiosis, a well-coordinated exchange of molecular signaling between the legume host and bacterial partner occurs leading ultimately to the formation of nodules (Lakshminarayana and Sharma 1994).

Application of beneficial microorganisms is known to have variable effects on the development of nodulation and nitrogen fixation in legumes. It caused a significant reduction in nodule number but increased the size and biomass of nodules in *Trifolium alexandrinum* (Bajwa et al. 1999a). In a similar experiment Javaid et al. (2000c) noted a significant increase in nodulation in *Vigna radiata* due to beneficial microorganisms application. Javaid et al. (2002) have reported similar effects of long-term beneficial microorganisms application and organic manures on nodulation in *Phaseolus vulgaris* L. Sangakkara and Higa (1994b) studied the effect of EM on nodulation parameters of vegetable beans (*Phaseolus vulgaris*) and mungbean [*Vigna radiata* (L.) Wilczek] in soils with low and high population of rhizobia. Application of beneficial microorganisms significantly increased the most probable number counts of bacteria in the soils. The greatest change was observed in soil with low inherent microbial populations. Nodulation and nitrogenase activity, characteristics of both the legumes, were also significantly enhanced by beneficial microorganisms application especially when grown in nutrient-depleted soil. Addition of fertilizer decreased the process of biological nitrogen fixation. However, this adverse impact was reduced with beneficial microorganisms application. Application of EM Bokashi significantly increased both the nodule numbers per plant and fresh weight per nodule in peanut (Yan and Xu 2002). Recently Javaid (2006) conducted a pot experiment to evaluate the efficacy of foliar and soil application of EM on nodulation in pea in soil amended with NPK fertilizers, farmyard manure, and *Trifolium* green manure. Results indicated that at flowering stage, beneficial microorganisms application depressed the nodulation and maximum number of nodules were recorded in uninoculated control in all the three soil amendments. Difference between control and microbial inoculated treatments was more pronounced in NPK and farmyard manure amendments than in green manure amendment. At maturity, foliar spray of effective microorganisms significantly enhanced the number of nodules in NPK fertilizers amendment. A similar but insignificant increase in the number of nodules was also recorded due to foliar spray in green manure amendment. Soil and soil plus foliar application of effective microorganisms depressed nodulation in all the three soil amendment systems at this growth stage. Effect of foliar spray and soil application of effective microorganisms on nodules biomass in different soil amendment systems was generally similar to that of nodules number**.** This variation in nodulation could be attributed to various factors including the soil physical and chemical properties, soil amendment, cropping and agricultural practices history, soil indigenous rhizobial and other microbes population, environmental conditions of the area, and concentration of beneficial microorganisms.

12.5.2 Arbuscular Mycorrhizal Fungi

The fungi that are probably most abundant in agricultural soils are arbuscular mycorrhizal (AM) that account for 5–50% of the biomass of soil (Olsson et al. 1999). These fungi are multifunctional in ecosystems. Colonization of roots by AM fungi has been shown to improve growth and productivity of several field crops (Javaid et al. 1994; Kapoor et al. 2004; Cavagnaro et al. 2006; Chen et al. 2007) by increasing nutrient element uptake (Al-Karaki 2002; Pasqualini et al. 2007); enhanced tolerance to various biotic (Khaosaad et al. 2007) and abiotic stress factors (Arriagada et al. 2007); and improving physical, chemical, and biological properties of soil (Rillig and Mummey 2006).

Few studies, mostly by our research group, have been conducted to assess the effect of beneficial microorganisms application on mycorrhizal colonization, and effect of dual inoculation of beneficial microorganisms and mycorrhizae on crop growth and yield of test species. Variable results were obtained in these interactive studies. The variation in response of crop growth, yield, and mycorrhizal colonization to beneficial microorganisms application or co-inoculation of beneficial microorganisms and mycorrhizae was generally associated with the nature of the test species, soil amendment, and history of beneficial microorganisms application. Bajwa and Jilani (1994) studied the interaction of beneficial microorganisms and mycorrhizal fungi in sterilized pot soil and found that beneficial microorganisms significantly enhanced the mycorrhizal colonization in maize and the combined inoculation resulted in significantly increased crop growth and yield. Bajwa et al. (2002) found that beneficial microorganisms application increased maize growth in both farmyard and green manure amended soils while mycorrhizal colonization was favored by beneficial microorganisms only in farmyard manure amended soil. Similarly Bajwa et al. (1995b) assessed the usefulness of dual inoculation of beneficial microorganisms and two mycorrhizal species viz. *Glomus mosseae* and *G. fasciculatum* in improving growth and yield of tomato. They noted a significantly greater shoot dry biomass and fruit yield in *G. mosseae* plus beneficial microorganisms and *G. mosseae* plus *G. fasciculatum* plus beneficial microorganisms as compared to respective sole mycorrhizal treatments. In another experiment, Bajwa et al. (1995c) studied the effect of beneficial microorganisms application on crop growth, mycorrhizal colonization, and nutrient uptake in soybean by introducing extra-mycorrhizal spores. They reported that indigenous mycorrhizal flora of field soil did not respond positively to beneficial microorganisms application and mycorrhizal infection failed to develop properly with subsequent adverse effects on host plant growth. However, beneficial microorganisms significantly supported externally introduced mycorrhizal inoculum and marked influence was observed on crop growth and nutrient uptake. Javaid et al. (1995) showed that application of beneficial microorganisms in unsterilized field soil enhanced mycorrhizal colonization in the roots of pea, resulting in increased growth, yield, nodulation, and nitrogen nutrition in host plant. Similarly, Bajwa et al. (1998b) observed a significant increase in root and shoot growth, and shoot P and N content in chickpea due to co-inoculation of beneficial microorganisms and mycorrhiza. Beneficial microorganisms application also favored mycorrhizal development in root cortex of host chickpea plants. In another study, the authors also noted similar effects of beneficial microorganisms and mycorrhizae under allelopathic stress caused by aqueous leaf extract of *Syzygium cumini* (L.) Skeels (Bajwa et al. 1999b). Javaid et al. (1999) conducted a field study to evaluate the effectiveness of beneficial microorganisms application on mycorrhizal colonization and subsequent growth and yield in sunflower, at two growth stages viz. 40 and 70 days after sowing. In 40-day-old plants, beneficial microorganisms supported mycorrhizal association, which resulted in a parallel increase in number and biomass of leaves as well as stem length while stem biomass remained unaffected. However, beneficial microorganisms application failed to induce any remarkable change in extent of mycorrhizal colonization at 70 days growth stage. However, the number of arbuscules was enhanced by beneficial microorganisms application at this growth stage that resulted in a parallel increase in vegetative growth and yield of the host plant. By contrast, Bajwa et al. (1999a) while studying the effect of beneficial microorganisms on mycorrhizal colonization, nodulation, and crop growth in *Trifolium alexandrium* L., in soils amended with farmyard manure and green manure, noted that EM significantly enhanced mycorrhizal colonization but exhibited an inhibitory effect on crop growth. Javaid et al. (2000b) conducted pot experiment in farmyard and green manure amended soils with two different histories of beneficial microorganisms application using *Vigna mungo* as test species. They observed a better response of crop growth, nodulation, and mycorrhizal colonization to beneficial microorganisms application in soil 1 where beneficial microorganisms application was started 6 months prior than the soil 2. In a similar experiment, Javaid et al. (2000c) noted a significant increase in mycorrhizal colonization in *Vigna radiata* due to beneficial microorganisms application. In another study, Javaid et al. (2000a) assessed the effects of long-term application of beneficial microorganisms and organic manures on mycorrhizal colonization, crop growth, and yield of wheat in soils with three different histories of beneficial microorganisms application. Beneficial microorganisms proved more effective in increasing mycorrhizal colonization and yield in wheat in soil with oldest history of application of these microorganisms. Similarly, Javaid et al. (2002) have reported similar effects of long-term application of beneficial microorganisms and organic manures on crop growth and mycorrhizal colonization in *Phaseolus vulgaris* L. Mridha et al. (1997) reported that dual inoculation of arbuscular mycorrhizae and EM resulted in significant better plant growth in *Sesbania rostrata* as compared to either beneficial microorganisms or mycorrhizal inoculation. Furthermore, beneficial microorganisms application enhanced percentage root colonization and mycorrhizal spores.

12.6 Pest Management with Beneficial Microorganisms

12.6.1 Weed Management

Weeds compete with crops for nutrients, available moisture, space and sunlight, which results in yield reduction. Weeds also deteriorate the quality of farm products and hence reduce the market value. Since weeds are present in all food crop systems irrespective of the intensity of the crop management (Schroeder et al. 1993), thus their control is vital to achieving high yields. Weeds have traditionally been controlled by manual and cultural methods. With the development of synthetic agrochemicals in the 1940s, the reliance on chemicals to obtain weed-free cropping systems increased. However, the indiscriminate use of these synthetic chemicals has led to pollution problems in most agricultural systems, and more importantly the development of herbicide-resistant and problematic weed species (Chhokar et al. 2008; Doole and Pannell 2008). Modern biological agricultural systems do not permit the use of synthetic herbicides for weed control because of their potential to become environmental pollutants and harmful residues in the food chain. There is a growing interest in nonchemical weed control methods worldwide.

Application of beneficial microorganisms is known to stimulate seed germination and early growth of food crops (Sangakkara and Higa 1994a) and can create a more favorable root surface-rhizosphere environment for crop plants that improves plant growth and protection (Sangakkara 1996). It is likely that these documented beneficial effects of beneficial microorganisms on crop plants would also be extended to weeds, and could enhance weed seed germination, early growth and development, and their level of infestation. Consequently, there is considerable interest in whether beneficial microorganisms through this process, over time, could reduce soil weed seed-bank. Maramble and Sangakkara (1998) conducted a study to determine the effect of beneficial microorganisms on weed population and weed growth grown with organic amendments during the dry season of 3 consecutive years in Sri Lanka. The application of organic amendments alone suppressed weed growth, although the variation among the years was insignificant. Beneficial microorganisms applied with organic amendments enhanced weed growth during the first year which then declined significantly during the succeeding years. In a similar study, Maramble et al. (1996a) investigated the influence of beneficial microorganisms and organic matter on weed populations of two annual food legumes in consecutive wet and dry season over 2 years. The lowest weed populations and crop yields were obtained from plots not receiving beneficial microorganisms or chemical fertilizers. Application of beneficial microorganisms especially with organic matter having a low C:N ratio enhanced weed populations in the first year. The impact was more pronounced in dry than in wet season. In the second year, beneficial microorganisms increased crop yields and reduced weed populations and biomass significantly. The authors suggested that long-term studies are required to get more benefits of beneficial microorganisms as an alternative method for controlling weeds. Maramble et al. (1996b) reported that application of beneficial microorganisms significantly increased tuber germination and subsequent growth of purple nutsedge (*Cyperus rotundus* L.) plants. However, beneficial microorganisms significantly lowered the number of tubers and tuber biomass at the time of flowering. The authors suggested the poor tuber formation in beneficial microorganisms treated nutsedge plants during the first season would reduce the weed infestation during the following season.

12.6.2 Control of Fungal Pathogens

Different species of turfgrass are widely used worldwide for golf courses, athletic fields, and landscaping. *Sclerotinia homoeocarpa* (Lib.) Korf & Dumont, causal agent of dollar spot disease is considered the most prevalent turfgrass pathogen in North America, Central America, Australia, New Zealand, and Europe. Fungicides are a major input for controlling this disease. To evaluate effective alternative approach, Kremer et al. (2000) conducted in vitro laboratory bioassays to determine the effects of beneficial microorganisms on growth and development of *S. homoeocarpa*. The results showed that beneficial microorganisms amendment in potato dextrose agar medium at 1.0% and 4.0% significantly inhibited hyphal growth of *S. homoeocarpa.* Following in vitro bioassays, greenhouse study was conducted to investigate the effect of beneficial microorganisms on disease development by *S. homoeocarpa* in turfgrass and turf quality. They found that beneficial microorganisms treated compost treatment had significantly less disease than the standard golf green substrate. According to Tokeshi et al. (1998) soils treated beneficial microorganisms were found to be suppressive to the soil-borne plant pathogen *Sclerotinia sclerotiorum.* Similarly Jonglaekha et al. (1995) reported that root rot in strawberry, caused by *Rhizoctonia fragariae*, can be considerably controlled either by mixing beneficial microorganisms compost in the soil or applying beneficial microorganisms solution for 4–6 times at weekly intervals. Application of beneficial microorganisms also reduced the incidence of wilt disease of potato (Jonglaekha et al. 1993). Encouraging results have also been recorded in the management of anthracnose of sweet potato caused by *Colletotrichum gloeosporioides*, suppressing populations of soil-borne phytopathogenic fungus *Phytophthora cinnamomi*, and control of black sigatoka disease of bananas caused by *Mycospherella fijiensis* (Tokeshi and Chagas 1996; Aryantha and Guest 1997; Elango et al. 1997). Control of fungal pathogens may be attributed to the activity of lactic acid bacteria in the beneficial microorganisms mixture that produce lactic acid, a strong sterilizing compound (Higa and Kinjo 1991; Higa 2000).

12.6.3 Control of Bacterial Pathogens

Few studies have been conducted to investigate the effect of beneficial microorganisms on bacterial diseases. Castro et al. (1996a) conducted an in vitro study and found that beneficial microorganisms inhibited the growth of *Xanthomonas campestris* pv. *vesicatoria* and *Pseudomonas solanacearum.* They extended the study to evaluate the potential of these microorganisms for control of *X. campestris* pv. *vesicatoria* in sweet pepper (*Capsicum annum* cv. *margareth*) under field conditions and obtained promising results in the management of the disease (Castro et al. 1996b). The suppressive effects of beneficial microorganisms have also been reported against bacterial leaf blight of rice caused by *Xanthomonas oryzae* pv. *oryzae* (Myint et al. 1996).

12.6.4 Control of Insect Pests

Very little work has been carried out regarding the effectiveness of beneficial microorganisms against insect pests. Nasiruddin and Karim (1996) conducted a field trial to test the efficacy of one formulation of beneficial microorganisms (EM_5) in reducing the damages caused by the red pumpkin beetle (*Aulacophora foveicollis*) and the melon fly (*Bactrocera cucurbitae*) in cucurbitaceous vegetable crops. EM_5 reduced the beetle infestation by 38% and melon fly infestation by 28.3–35.8% over the untreated control. Chemical insecticides showed more than 80% reduction of infestation of the two insect pests. Pickleworm (*Diaphania nitidalis*) is a serious pest of cucumber and other vegetables of the Cucurbitaceae family. The conventional control of this pest calls for excessive use of synthetic pesticides which pollute both product and environment. Wood et al. (1997) reported that incidence of disease and damage by the pickleworm was significantly reduced by foliar applications of beneficial microorganisms fermented plant extracts in combination with EM_5.

12.7 Role of Beneficial Microorganisms under Abiotic Stresses

Few reports are available in the literature regarding the role of beneficial microorganisms under abiotic stress factors such as allelopathy, acidity, and salinity. Bajwa et al. (1999a) reported that application of beneficial microorganisms significantly reduced the adverse impact of aqueous leaf extract of an allelopathic tree *Syzygium cumini* (L.) Skeels on plant growth, yield, and shoot nitrogen content of chickpea (*Cicer arietinum* L.). The efficacy of beneficial microorganisms was further enhanced by dual inoculation with arbuscular mycorrhizal fungi.

Pairintra and Pakdee (1994) suggested that beneficial microorganisms treated compost can be used as an efficient soil amendment in ameliorating a slightly saline soil. Aluminum toxicity is considered to be the most important growth limiting factor in many acid soils in Malaysia, especially those having pH levels below 5.0. Anuar et al. (1997) conducted a field experiment to evaluate the use of beneficial microorganisms in reducing the aluminum toxicity of an acid soil, its effect on the yield of sweet potato and selected chemical changes of the soil. The results showed that exchangeable aluminum was reduced with the application of beneficial microorganisms. Furthermore, these microorganisms increased the yield of sweet potato and also influenced the chemical characteristics of the acid soil.

12.8 Treatment of Dairy Wastewater with Beneficial Microorganisms

Wastewater originated from dairy operations may contain certain human pathogens including *Escherichia coli*. In addition, excess nutrients present in dairy wastewater can also pollute surface and ground waters. Beneficial microorganisms and duckweed have shown a great promise in dairy wastewater treatment. According to

Rashid and West (2006) combined application of beneficial microorganisms and duckweed growth significantly reduced the ammonium nitrogen, total phosphorus, total suspended solids, and biological oxygen demand after 3 months and is a very efficient way of dairy wastewater treatment. Li and Ni (2000) reported 42–70% reduction in NH_3 concentration in poultry manure when beneficial microorganisms were added to both drinking water and feed. The mechanism involved suggests that as EM is a mixed culture of many species of microorganisms, some of which can transform NH_4^+ to NO_3^-, thereby decreasing the potential for N-fraction.

12.9 Conclusion

Extensive studies carried out in various countries of Asia Pacific Region have shown that beneficial microorganisms enhance the growth, yield, and quality of various agricultural and horticultural crops possibly through rapid decomposition of organic matter, production of biogenic substances, improved soil quality, and enhanced growth and efficacy of symbiotic. However, the affectivity of beneficial microorganisms varies with soil type, source and amount of soil nutrients, and test crop species. The negative or no effects of beneficial microorganisms as reported by some workers can be overcome through repeated applications of these microorganisms. For the best exploitation of advantages of beneficial microorganisms, they should be used in combination with organic matter plus half dose of NPK fertilizers. Furthermore, they should be used as foliar spray as this practice avoids many of the biotic and abiotic factors and constraints of the soil environment.

Most of the studies with beneficial microorganisms limited to their effects on crop growth and yield. However, in other areas of agriculture such as management of pests like weeds and insects, and control of plant pathogens including bacteria and fungi, only few studies have been undertaken. These few studies, however, exhibited very encouraging results in the management of pests and diseases with beneficial microorganisms. Similarly, few studies conducted so far to manage the environmental problems like treatment of wastewater and poultry manure revealed that the benefits of beneficial microorganisms can also be exploited in these areas. However, more intensive and systematic studies are required in these areas to provide a better understanding of the usefulness of beneficial microorganisms technology in various farming systems and environmental issues to provide safe food products to the consumers.

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