Chapter 16 Biofertilizers as Microbial Consortium for Sustainability in Agriculture



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Abstract In the entire world, the aggregate effect of climate change is ceaselessly expanding deteriorated lands creating pressure on agricultural output and food security. The use of biofertilizers instead of chemical fertilizers can improve crop productivity and food quality under different environmental conditions. The use of bioagents minimizes the deposition of toxic agrochemicals in soil without altering its biological and functional characteristics. Biofertilizers are mainly comprised of a single or combination of microorganisms which can be endophytic or rhizospheric in nature. The communities of plants are directly or indirectly impacted by rhizospheric microorganisms which influence the structure and yield capacity. Considerable data is presently accessible on the composition and different aspects of plants along with microbial population residing in the rhizosphere and their functional capabilities. Hence, belowground microbiota is regarded as a forecaster of variations in plants and overground yield efficiency. Different approaches for microbial population improvement exist, and the use of microbial consortium (MC) as biofertilizer is one of them. Farming practices, environmental factors, and plant genotypes harbor distinct and diverse microbial communities and their functions. Currently, biofertilizer products having individual or combination of microbes exhibit restricted efficiency in specific environmental regimes. MC as a biofertilizer contributes a lot to help the plant to cope up against numerous strains (abiotic and biotic stresses) in different environmental conditions. Therefore, the selection of an appropriate MC for a particular agroecosystem and/or genotype of crops is in the direction of improving interactions between crop and the introduced microbes and is considered to be the way forward for enhancing profitability. However, the benefits of using MC

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over the use of individual microbes lie in their multifunctionality unmistakably demonstrated by researches. However, limited attention is being paid by the manufacturers in maintaining quality norms. In the current chapter, we focused on the progress made in the development of biofertilizers comprising MC and their quality, microbiome engineering of biofertilizers, and their impact on plants under various environmental conditions.

Keywords Rhizosphere \cdot Biofertilizer \cdot Microbial consortia \cdot PGPR \cdot Microbial inoculants

16.1 Introduction

Green revolution or the third agricultural revolution had significantly enhanced agricultural food grain production (especially wheat and rice) worldwide, to meet the demand of diet at the beginning of the late 1960s. The rapid increase in agricultural output came from the green revolution because of the enhanced utilization of different chemical inputs (fertilizers, pesticides, and herbicides). This excessive utilization of chemicals caused deleterious impacts on the fertility of the land in addition to the well-being of mankind (Alori and Babalola 2018). The continuous rising of such environmental issues not only enforced researchers to solve the hazardous effect of these chemical fertilizers on the ecosystem but also encouraged farmers for cultivation involving sustainable approaches (Malusá et al. 2012). Globally, the population of mankind at the end of 2050 is anticipated to upsurge nearly 9.6 billion (Yadav et al. 2017). However, on the way to provide food to every individual, two challenges are identified: i) the reduction in arable land due to land acquisition for building residences and ii) availability of quality food to everyone. However, excessive utilization of chemical fertilizers to enhance agricultural production is not feasible in the context of the environment and the wellness of mankind. Under this circumstance, biofertilizer can be implemented as plant growth-promoting agents that would help in reducing the use of chemical fertilizers making the lands more fertile which ultimately will increase yield in addition to reducing different diseases of crop plants (Patel et al. 2016; Singh et al. 2019a). These biofertilizers include live beneficial microorganisms associated with the host and which could provide direct or indirect gain to the host by adapting various mechanisms that lead to enhanced crop production (Fuentes-Ramirez and Caballero-Mellado 2005). Application of these biofertilizer agents either with seed or soil as an inoculant enriches the soil with various important nutrients (micro- and macronutrients) via several ways like nitrogen fixation, nutrients solubilization, and mobilization, secreting compounds, and antibiotics involved in plant development (Singh et al. 2011). Some of these biofertilizers could also help in degrading organic matter and enhancing soil nutrient availability to the plants (Sinha et al. 2010).

In agricultural fields, most of the chemical enrichers applied are leached out (approx. 60-90%), and only the remaining (~10-40%) is available to plants. Yet,

these chemicals are not readily available to plants as such due to the formation of complexes with other compounds. The application of biofertilizers enhances agricultural productivity playing a vital part in integrated nutrient management (Bhardwaj et al. 2014; Singh et al. 2019b). Moreover, the implementation of these biofertilizers in soil reduces the chemical input leading to organic farming practices. The demand for organic input in place of a chemical has been recommended for agricultural crop production to improve nutrient supply and maintaining soil fertility. The organic farming system is helpful to ensure food security and enriches soil biodiversity (Yadav et al. 2017). Microbes are found in their natural habitat in communities. Microbial communities in their habitat refer to the formation of microbial consortium (MC) that offers multiple actions like enhancing plant growth and minimizing abiotic and biotic stresses, viz., drought, chilling, temperature variation, pests, and disease infection in plants leading to food safety and security (Sekar et al. 2016). MC is synergistically associated with the host and mimics with the natural condition and plays a diverse role in the rhizospheric zone by solving the most challenging issues raised around the rhizosphere and creates an eco-friendly environment among soil-plant-atmosphere (Jain et al. 2013). In addition to increasing their populations, microbes also provide multiple benefits that support plants for tolerating several abiotic strains (Singh et al. 2013). Subsequently, co-inoculation or soil amendment with consortia based on compatible microbes has very high significance over a single application (Sekar et al. 2016).

The present trend has shown more focus on the application of MC on a small scale to control phytopathogens and improve plant health. Positive outcomes from such studies have attracted more researchers to experiment with MC rather than using single microbial inoculant (Sarma et al. 2015). Furthermore, alternative ways to enhance crop yield is by maintaining soil health and engineering of rhizomicrobiome. The integration of biotechnological approaches over bio-formulations in the cropping system has been adopted globally and would cope with the several challenges raised in plant growth (Odoh 2017). In the current scenario, the colossal application of microbes-based bio-formulations as a biofertilizer in agricultural fields is increasing because of its capacity to preserve the healthiness of soil and lowering down of the environmental concerns. Besides, it can cut down the utilization of inorganic chemicals in agricultural practices. Additionally, the use of biofertilizers is more effective in rainfed agriculture, mainly for the marginal farmers, who cannot afford the high cost of chemical fertilizers (Barman et al. 2017). Biofertilizer application is an ideal, cost-effective, and sustainable approach in farming as it conserves long-term soil fertility (Shelat et al. 2017). Agriculturally, important microorganisms used as biofertilizers include fungal mycorrhiza, cyanobacteria involved in the fixation of nitrogen, and PGPRs (plant growth-promoting rhizobacteria), biocontrol agents, biopesticides, endophytes, and bio-degrading microbes (Singh et al. 2011, 2018). Indeed, microbes are used as supplementary components in the soil which is helpful in promoting cropping practices like crop rotation, crop residue recycling, tillage, and organic manure maintenance. Long-term use of such important microbes in the soil can sustain enhanced yield in many commercial crops (cotton, jute, oilseed, sugarcane, sun

hemp, tobacco, tea, coffee, etc.) (Bhardwaj et al. 2014). Therefore, the application of bio-formulations of compatible microbial consortium as biofertilizers around the rhizospheric region could be an efficient tactic for promoting plant growth and development. Similarly, the co-metabolism application of MC might also be a superior approach over single inoculum. This process is manifested during an interaction between microbes, where secreted specific metabolites serve as restricting elements to different communities of microbes within the network (Odoh et al. 2020). This facilitates the availability of limiting nutrients by mineralization of the by-products in addition to augmenting capacities of arable lands.

16.2 Development of Multifunctional MC

Crop productivity can be affected by two major environmental stresses, i.e., abiotic and biotic stresses. Several findings are coming out with tools to minimize these stresses and improve crop productivity. PGPRs are serving an important part against these stresses (Yang et al. 2009). Essential nutrient sources are present in the soil in sufficient amounts, but most of the time they are unavailable to plants. Rhizospheric bacteria present in the soil are majorly known to solubilize these nutrients for plants. Nitrogen availability in the environment is 78%, and plants cannot take it directly from the environment. Here comes the role of microorganisms to fix atmospheric nitrogen for plants and helps to maintain the nitrogen cycle (Rasche and Cadisch 2013). Microbes can solubilize naturally occurring nutrients present in bound form in the environment and maintain the nutrient cycle from source to sink, such as phosphorus (P), sulfur (S), potassium (K), magnesium (Mg), and calcium (Ca). Artificial application of selected microbes could be a strategy to make the soil rich in nutrients and also to minimize the use of chemical fertilizers and thus maintain soil nutrient balance (Ahemad and Kibret 2014).

Microbes may be utilized like biofertilizers or biocontrol agents either individually or in consortia. Microbes should be characterized and well tested scientifically for their specific biofertilizer activity before use. These microbes should fulfill several specific criteria to be a candidate for field use. The use of several microbes having different specific characters could be a strategy to use their potential efficiently. Several reports (Jain et al. 2012; Singh et al. 2013; Patel et al. 2016, 2017) display the use of compatible MC (either 2–3 bacteria or bacteria and fungi together) for enhancement of plant resistance against stressful factors besides improving the development of the plant. Utilizing different microbes belonging to different rhizosphere and environments can enhance biocontrol efficiency as well as minimize the competition among them. The synergistic effect of these compatible microbes for a plant trait such as crop yield or availability of nutrients resulting in improved plant growth and yield has been reported earlier. The development of potential MC for making the microbes more effective is very important, and it can be a substitute for using harmful pesticides and chemical fertilizers to a great extent. Enhanced uptake of phosphorous and nitrogen along with protection against soil-borne phytopathogens has been reported for the consortium of *Trichoderma*, *Rhizobium*, and PSB, i.e., phosphate-solubilizing bacteria (Rudresh et al. 2005). Sarma et al. (2015) have observed individual microbial components of consortia, and their importance for the protection of plants counters to numerous phytopathogens.

Consortia of fungal mycorrhiza, PGPR, and bacteria living as endophytes have been reported for significantly enhancing plant protection and reducing reliance on chemical fertilizers (Pérez et al. 2007). Utilization of PGPR and bacteria living as endophytes may be considered to use in combination and as it would be a good and highly effective tactic for ensuring sustainability in agriculture and integrated pest management practices. Such a combination can control pests such as fungal pathogens, insects, and weeds effectively. The combined application of PGPR and Bacillus sp. has been suggested by Prabhukarthikeyan et al. (2014) for the biocontrol of tomato Fusarium wilt as well as tomato fruit borer without using any other chemical pesticides. Rhizobacteria possess the spatial ability to control different plant pathogens. A report for inducing induced systemic resistance (ISR) by Pseudomonas fluorescens has been also reported by Bandi and Sivasubramanian (2012) to control damage by an insect pest Thrips (Thrips tabaci L.). Researchers are continuously working to understand the roles of microorganisms in the agricultural system, but several puzzles remain unresolved. These microorganisms sustain capacity for improving growth and productivity of plants via different ways such as plant resistance induction and acclimatization to the environment and develop tolerance toward diverse abiotic strains (increased salinity, heavy metal, in addition to high pH) (Ahmad et al. 2018). We should consider the adaptability of MC being inoculated in different kinds of soil other than their natural environment. Earlier studies have not emphasized this point as it will be interesting to note whether the efficiency of the MC is enhanced in non-native soil. Consortia of microbes should be developed by using compatible microbes. It is, therefore, very necessary to test the compatibility of microbes before developing the consortia. If the microbes are not showing a synergistic effect on the targeted traits of plants, the basic concept behind the use of microbial combination will not be fulfilled (Sarma et al. 2015).

The development of microbial combinations that can improve several functions of the plants is a hot topic for current research. The Fig 16.1 highlights the basic methodoligies involved in development of MC for utilizing them as biofertilizers for reducing the chemical use in agriculture practices. Agricultural research is shifting toward the diminishing use of chemical fertilizers and pesticides without compromising production and quality of the produce. Recently, Backer et al. (2018) have shown the potential of PGPR for sustainable agriculture. The strategy of using MC is very old but it has been used for legumes and cereal crops significantly only in the last few years (Sessitsch and Mitter 2015). PGPR can use several mechanisms like deaminase action of ACC, enhanced fixation of N, and solubilization of calcium, besides phosphate solubilization for improvement in wellness of the plants and their yield capacity (Backer et al. 2018). MC activities should be thoroughly studied initially under laboratory conditions to maximize the effect of a consortium to its optimum level (Odoh et al. 2019).



Fig. 16.1 Basic steps for the development of microbial consortia as biofertilizers

Microbial inoculants should be accessed for their shelf life in the particular formulation. Multilocation field trials should be conducted and approved for commercialization. Such testing and approval are important to release any microbe in a particular environment. A recent report (Backer et al. 2018) has shown the necessity to know the microbial load to be inoculated in an agricultural field for efficient colonization in the rhizosphere. The optimal spore dose of *Trichoderma asperellum* varied for different vegetable crops as determined by the growth and germination of the vegetable seeds (Singh et al. 2016a, b). The specific role (such as effect on plant growth, the effect on nutrient uptake, development of host resistance) of each component of MC must be well known including the type of soils suitable for them (Macouzet 2016; Baez-Rogelio et al. 2017). Furthermore, training of staff and farmers is needed for efficient use of these bio-inoculants concerning knowledge about soil specificity, the effect of environmental factors, and complexity of the individual components (Parnell et al. 2016; Bashan 2016; Itelima et al. 2018).

16.3 Impact of MC as Biofertilizer in Different Environmental Conditions

Over the period of evolution, plants are constantly evolving based on their relationship with the associated microorganisms which regulates the well-being and development of plants. These plant-associated microbes, i.e., plant holobiont termed as plant microbiota; plant microbiome which comprises the microbes associated in the different portions of the plants, viz., rhizosphere, phyllosphere, and endospheres; and such microorganisms straight or circuitously have links with the plant's growth in addition to their healthiness (Vorholt 2012; Brader et al. 2017; Lemanceau et al. 2017). For maintaining proper relation, floras actively recruit microbes from various reservoirs, i.e., rhizospheric (soil), phyllospheric (leaf surface and its surrounding environment), the anthosphere (flowers), the spermosphere (seed germination), and the carposphere (fruit area) (Hardoim et al. 2015). Limited information is available related to the structure and different aspects about plant-associated microbes. However, the abundance and species richness information are most commonly revealed by many researchers and tried their best to identify the structural basis of their composition in their community. These microbiomes serve an efficient part to fulfill the requirements of emerging challenges during the production of crops and an emergency prerequisite to constructing innovation in microbial technologies regarding their adaptation to productive agriculture. Plant microbiota has potentiality to reduce farmer's income by utilizing microbes for soil enrichment, nutrient uptake, managing biotic and abiotic stresses, weed management, improving crop nutrient status, and ultimately increasing crop yields (Jangra et al. 2018). Prior to use in agricultural practices, it is necessary to study cultural characteristics and the nature of adaptability of these microbes to know their behavior in different soils (Jiao et al. 2018). However, environmental factors are the principal character in deciding the role of applied microbes and their nature of adaptation concerning soil. Improved approaches for the application of microbes as a group of related strains or their blends could be standardized taking account of soil variability and external parameters. By looking into the crop status of the past years, it is realized that a smart knowledge-based choice of microbes is required to put forward the delivery approach and formulations. Instead, agricultural methods and crop varieties could impact the abundance of plant microbiota and its role in agriculture. Therefore, planning of suitable agricultural techniques beforehand could improve plant microbiome association during and after the cropping season and eventually provide benefit to better adaptability of plant microbiota.

Root microbiota mainly known as rhizobiome harbors a limited group of microbes based on the soil type that surrounds them which can be mostly horizontally transferred, i.e., the difference in soil type and their respective environment. Rhizobiome is extremely complex driven and consists of various microbes. Soil microbes can also target the ecosystem through the biogeochemical cycling of available elements along with the formation of soil surface/sub-surface particles, pollutant degradation, and water quality (Li et al. 2014; Eilers et al. 2012). However, sometimes it can also be vertically transferred via seeds, host plant, available nutrients, and organic matter (Jiao et al. 2018). Seeds also represent a central foundation of microbes as microbes are associated either intrinsically or extrinsically and serve as the initial region for multiplication in the roots in the seedling (Liu et al. 2012). Rhizospheric zone has the ability to provide unique ecological niche and metabolites that help in the attraction of microbiota which consequently provides their effect on the remaining plant parts (Hartmann et al. 2009). However, understanding the rhizobiome with the domesticated plants does not represent the status of native plants as they recruit various microbes during their growth and development (Bulgarelli et al. 2013). Various reports explained the higher richness of bacterial species in root microbiota in the rainforest when compared to other soils. The highest taxonomic ranks for the microbe diversity (bacteria) are given to alphaproteobacteria <actinobacter<acidobacter in various root-associated studies (Yeoh et al. 2017). Still, the beta-proteobacteria hold better species richness in root association when compared to rhizobiome status suggesting the recruitment process and enrichment of the root environment attract the nearby microbes (Lundberg et al. 2012). Recently, Donn et al. (2015) studied wheat rhizosphere to understand the root-driven bacterial abundance resulting in tenfold increased abundance of actinobacteria, and other microbes, i.e., pseudomonads, oligotrophs, and copiotrophs, in comparison to bulk soil further suggest an alteration of rhizosphere and rhizoplane microbe structure without affecting the bulk soil population.

However, the difference in plant genotypes and relative species can also influence the structure of rhizospheric microbes. The variation in bacterial community is not only affected by rhizosphere or surrounding environment, but the difference in host genetic content can also alter the diversity in microbiodata. Bouffaud et al. (2012) studied the richness of the microbiome in an inbred line of maize landraces using microarray analysis of rhizospheric samples. The dent corn group produced higher discriminating signals targeting the beta-proteobacteria genera, but the flint corn received higher signals from alpha-proteobacteria. Delta-proteobacteria and betaproteobacteria group bacteria were able to produce high signal intensity in tropical and stiff stalk corn group (Bouffaud et al. 2012), which states the qualitative difference in rhizodeposition and other exudates composition may raise the difference in bacterial community association (Bressan et al. 2009). Zarraonaindia and Gilbert (2015) demonstrated the above-ground microbiome profiling in grapevine and found Sphingomonas and Pseudomonas were abundant in vine leaves and grapes due to availability of nutrient source and water-limited condition for growth and development of microbes by giving an advantage to crops for disease suppression besides guarding counter to water stress. Flora roots are also colonized internally (root endospheres) by abundant endophytic microbes. The microbe entry takes place through a passive process, root cracks; wounds in roots and emergence of new lateral roots provide the access to a diverse range of microbes (Compant et al. 2005). Correa-Galeote et al. (2018) studied the maize root endophytes showing the predominance of Proteobacteria, Firmicutes, and Bacteroidetes due to soil cultivation practices history. Whereas in rice roots, the microbes belongs to family Rhizobiaceae, Comamonadaceae, Streptomycetaceae, and Bradyrhizobiaceae are occupying the diverse status (Edwards et al. 2015). Similarly, in grapevine, the fullness of Acidobacteria, Proteobacteria, Actinobacteria, Bacteroidetes, and Firmicutes was found in various studies (Burns et al. 2015; Faist et al. 2016; Samad et al. 2017). Apart from the adaptation of microbes to various plant parts and root zone, microbes too serve a vital part against the living and environmental constraints. During this process, the tendency of variation in microbial physiological characters and metabolic pathways occurs due to response from the stress generated. Application of single microbe or consortia is gaining more interest among the researchers and government launching a lot of initiatives to this kind of work by keeping in mind about the reduction in chemical use in agriculture. Similarly, food crops are facing threat due to the climate change scenario created by the downfall of crop production (Odoh et al. 2020). However, the application of microbes in such areas can help in mitigating various biotic and abiotic stresses that result in less crop loss. One such example revealed the meta-transcriptomics analysis resulted in the production of the polyketides, osmotic stress, and cold shock genes in suppressive soils due to the occurrence of *Stenotrophomonas* spp. and *Buttiauxella* spp., whereas oxidative stress genes along with antibiotic synthesis genes were more prevalent in non-suppressive soil in which *Pseudomonas* spp. and *Arthrobacter* spp. were highly present (Hayden et al. 2018). Hence, microbes have the potential to be used as biofertilizers, biopesticides, bioherbicides, and decomposers, and many had already arrived in the market as substitutes of chemicals with wider adaptability in a different environment (Mitter et al. 2016). MC which means mixing of two or more microbes based on the mode of work, i.e., biofertilizer-biopesticides, nodulation-growth enhancer, decomposer-growth promotion, nutrient use-crop protection, etc., is new market product strategies to reach more audiences with the same effect when applied in a single form (Yadav et al. 2019). Furthermore, collection of microbes from the extreme habitat and integrating various agri-microbial biotechnology tools to transfer the extreme habitat property to the locally adapted microflora will be a sustainable solution for the microbe adaptation without disturbing their community (Timmusk et al. 2017). Different studies proved the significance of microorganisms alleviating abiotic stresses (Kumar et al. 2019; Patel et al. 2017; Srivastava et al. 2015) and biotic stresses (Jain et al. 2012; Singh et al. 2013; Kumar et al. 2017) pretty well. Consortium development integrating microorganisms belonging to varied ecological backgrounds could provide crop protection in different environmental regimes. Thus, proper designing of MC as biofertilizers for particular environmental conditions and crops might prove to be a great move that can help in

enhanced crop productivity with reduced chemical uses and environmental damages.

16.4 Microbiome Engineering of Biofertilizers

The historic events during the green revolutions had initiated the cultivation of high yielding varieties. Indeed, an increase in the application of inorganic fertilizers and chemicals created a drastic impact on soil health status leading to depletion of useful

microbial diversity. This has led to the extinction or reduction in population dynamics of potential microbes which are working together for sustainable agriculture. However, in the present era, efforts are being made to conserve the potential microbes and engineer them ecologically to meet our requirements and applying those in other fields to meet our needs. However, the microbiome constitutes of a diverse group of microbes which have direct and indirect roles in the ruling soil ecosystem. During the interaction, soil microbes carry out various events in increasing quantitative food production, recycling of biogeochemical cycle, and maintaining soil health status (Hansel et al. 2008). During this process, these biological entities may have positive/negative influence on living and nonliving parameters (Odoh et al. 2019). Advanced studies in the medical field showed the importance of engineered microbes for fast and reliable production of antibiotics (Cycon et al. 2019), and food stains developed from microbes (Sen et al. 2019) by understanding the mechanisms, growth, and development pattern and complexity (Kumar 2016).

Plant-microbe interactions can take place with variation in relationships, i.e., beneficial, neutral, or completely negative. The beneficial interaction between plants and microbes is a matter of interest and is exploited extensively (Farrar et al. 2014). In this category of plant-microbes interaction, the role of arbuscular mycorrhizae (AM) holds a significant position in benefiting the soil and plants grown (Smith and Smith 2011). Similarly, fixation of N in legumes interacts with nodule-forming rhizobacteria (Oldrovd et al. 2011) and pathogenesis (Dodds and Rathjen 2010; Kachroo and Robin 2013; Wirthmueller et al. 2013). This system of symbiosis association between flora and microbes remains well-characterized providing clear information of gene expression, signaling pathway, and many more. However, understanding the plant evolution and adaptation to climate change scenario has made the scientific community think further in such studies (Hirsch 2004). In addition, plants are in interaction with other microbes (bacteria, fungi, algae) in an ecosystem either to get benefits or parasitize them in soil by producing the reciprocal signals during their interactions with other rhizospheric microbes or plants themselves (Badri and Vivanco 2009; Evangelisti et al. 2014). During interactions, microbes instead of acting individually potentially mingle with other microbes as consortia to exhibit the performance (Hirsch 2004). Sometimes the opportunistic microbes integrate with dynamic microbial communities posing threat to plant or humans because of pathogenic behaviors (Berg et al. 2005). MC can be administered by considering the practical parameters rather than selecting the specific microbe species and may undergo tripartite interactions (Bonfante and Anca 2009; Dames and Ridsdale 2012). Progress in procedures is necessary for manipulating the microbiome engineering process through various tools and techniques. Historically, research studies state the information of easily culturable microbial genera/species providing information about growth and medium parameters (Stewart 2012; Vartoukian et al. 2010). Still, some microbes especially endosymbionts are unable to grow in the absence of a living host as mycorrhiza needs a host plant to interact endophytically in the root system (Hildebrandt et al. 2002). Advancements in fluorescent tagging methodologies enable the visualization of the endophytic bacteria community (Elbeltagy et al. 2001) but make the microbe unculturable. This makes the microbiome research more interesting in understanding the microbial need to make it culturable and get benefitted from it. In some cases, microbes stay together in the entire life cycle as "obligate endophyte" which opens a new area of research for resilience in agriculture.

A considerable amount of information about plant microbiome is available now. Similarly, the reports are also available regarding the plant-microbe interactions. Every beneficial microbe does not have all the properties that are linked with structural progress of plant, development, nutrient solubilization besides mobilization, capability for tolerating various abiotic stresses, and biocontrol against various pathogenic microbes. Thus, the information about the microbiome of the plant and the particular genotype of the plant will be helpful to design the MC as a biofertilizer for the various crops in different environmental conditions. Various biotechnological interventions are available to edit the genome of microbes as per the requirement. These tools and techniques will also be helpful to create the compatibility of various incompatible beneficial microbes aimed at the expansion of MC as biofertilizers. By using the knowledge of plant microbiome and biotechnological advancement, microbiome engineering could have great potential for the development of MC as biofertilizers. In this regard, designing of MC must be free from opportunistic pathogenic microbes. Nithya et al. (2014) reported the food poisoning outbreak from lettuce and fresh fruits grown after microbial treatment. However, the engineering in microbiome provides major beneficial properties in the soil for continuous growing crops in rotation (Farrar et al. 2014). Thus, microbiome engineering might be helpful to open a new paradigm shift in sustainable agriculture for better crop production with food safety and security.

16.5 The Standard Norms for Biofertilizers Based on MC

It is very evident that profit due to the application of MC is massive. MC has logged encouraging achievements in different fields like ventures of food production, use in agricultural activities, medical uses, and ecological curative in comparison with a single strain. A challenging and multifaceted network of the prototypical microbial agent is shaped via metabolic modeling and specific strain recreation which is intended for ideal execution and creation of required biochemical agents and biomass (Faust 2019). Due to technical expertise in a few Asian countries like India and China, America, Africa, and Europe have a slight streak between fruitful uses of biofertilizers. The developed countries are focusing on thorough research on biotechnological methods for bioproducts designing, expanding mindfulness on their use although battling for relatively less utilization of chemical fertilizers. Sufficient consideration is yet to be given by developing countries on biofertilizers use as their benefit to the agricultural system is obvious. However, only a few farmers in such countries are using biofertilizers in their cropping practices. This is rather very much non-uniform as in Brazil where nearly the entire harvest protein is produced using

biological nitrogen fixation (BNF), but the use of biofertilizer is <1 percent in east and southern Africa. Normally, introduced biofertilizers are formed in accordance with or personalized to its origin nation remembering their regional circumstances like climatic and storing state. Moreover, these constraints assume an immense job in deciding their timeframe of realistic usability and practicality. Through enhanced manpower advancement via training and expanded awareness, research, and innovations, regional impacting circumstances will turn into a significant aspect during the production of indigenous biofertilizers specific to a particular area. This would, in turn, help limit viability loss detected in few biofertilizers available in the marketplace (Jefwa et al. 2014) in which state of storing in addition to management serves as a vital job. However, due to lack of insufficient and comprehensive studies on formulation development which is needed for the spatial crop responses, no country will have the ability to procure profit from the complete capacity of biofertilizers. In nations like India, it would be helpful in the conveyance of betterquality produce where support from the government has improved the production of biofertilizers (Odoh et al. 2020). The quality norm of biofertilizers should incorporate specification which could be recorded in the label or for marketing authorization that would be needed to be given. Known basic features of biofertilizers are considered to be crucial, and they are minimum number of living cells/propagules, nutrient solubilization efficacy and fixation in bacteria, plant inoculation competence in mycorrhizal fungi, time span of usability and/or date of expiry, level of contamination, the pH, the physical structure, and amount of carbon and water. Taking the technical prospects from the producer's part and the facts via researches, a range of standards must be setup for few parameters like minimum number of living cells/ propagules and for the efficacy statistics (Malusa and Vassilev 2014). PGPM strains used for commercial purpose should be precisely identified, and it is one of the most essential components. Based on the molecular biology approaches and strain's distinctive features documented in the registration dossier, identification of strains should be done. For conveyance to support commercialization of biofertilizers and conveyance of PGPM in proper physiological state and a reasonable quantity, the choice of the inoculant's carrier is very decisive (Malusá et al. 2012). Thus, other than the regular organic, inorganic, and polymeric complexes utilized as a carrier, presently an innovative way of utilizing biofilm of bacteria or nanocarriers as the carrier is a biological tactic which is under progress (Jayasinghearachchi and Seneviratne 2004; Qureshi et al. 2005; Seneviratne et al. 2007). Efficiency assessment can be very tough for MC biofertilizers. Consortium formed by the combination of a number of PGPM strain stimulates the growth of plant at various developmental phases, and they may also display different mode of activities which may occasionally include a mechanism(s) of plant defense. Improvement in nutrient efficacy by species consortia has been evidenced, for instances, combined inoculation of PSM + AMF, or *Rhizobium* + AMF in single gel preparation could likewise display plant defense characteristics (Vassilev et al. 2006). Moreover, multipurpose goods are more liked by farmers for utilization, and producers favor advertising products having numerous actions as they show higher impact and pull in users.

16.6 Ordinance and Commercialization of MC as Biofertilizer

The authentic definition of a commercialized product like biofertilizer is crucial for the producer's interest to manufacture them. In the United State of America (USA) and the European Union (EU), there is no any authenticate descriptions of biofertilizers or any lawful specifications to explain its features. In the EU, microbes (bacteria, viruses, and fungus) are incorporated as promising contributions in EU Commission Regulation No. 889/2008 on organic manufacture and solitary limited aimed at controlling pests and diseases employing biological approaches (Malusa and Vassilev 2014). A biofertilizer could in this manner be characterized as the developed item containing at least one microorganism that increases the status of plant nutrients (better development and yield) through supplementing soil nutrients additionally leading nutrients further accessible to floras as well as via expanding floras to get nutrients (Malusa and Vassilev 2014).

Worldwide the utilization and interest for biopesticides are ascending because of expanded attention to crops having no or less amount of pesticide residues. The worldwide level estimate for the microbe-based goods in 2014 was US\$ 2183 million, and by 2019, it is anticipated twofold by US\$ 4556 million with 15.3% of CAGR. Of the few microbial strains, the bacterial portion represented the biggest share (US\$ 1.6 billion). Like biopesticides, by 2020 the biofertilizers market worldwide is anticipated to reach US\$ 1.88 billion at 14% of CAGR between 2015 and 2020 (Markets and Markets 2015). All around, over 200 active ingredients of biopesticides are enrolled, and roughly 700 such products are accessible in the market. Considering the Indian market, 15 enlisted biopesticides were available during 2008 under the Insecticide Act (IA) (1968), and its market share is only about 4.2% of the total pesticide market. However, the biopesticide market is predicted to grow at a yearly growth rate of about 10% in the coming decade (Suresh 2012). Interestingly, the biopesticides enlisted have grown manifold during the previous years, NAAS (2013), and presently around 400 biopesticides are enlisted, and there are more than 1250 effectively enrolled biopesticide items available in Indian markets. This displays mindfulness among ranchers just like strategy backing of the administration to utilize the biologically sheltered items for bugging the executives. In absence of any particular guidelines, nearly 400 enrolled biopesticides are being marketed independently, and no MC is available (Sekar et al. 2016).

At the worldwide level, the administrative structures contrast broadly among various nations. In the USA, biopesticide creation is standardized beneath a different section as "Biopesticides and Pollution" inside the Environmental Protection Agency (EPA). Succeeding, in 1996 the Japanese Ministry of Agriculture, Forestry, and Fisheries (JMAFF) blended its framework with the rules of EPA. However, in Europe, biopesticides are assessed through the European Pesticide Regulation EC No. 1107/2009 which advances the creation of more safe substances by removing the unsafe products, and it has been advancing the enlistment of generally safe items through (2009/128/EC) basic and straightforward enrollment conventions

(Villaverde et al. 2014). Canada adopts just the security test, and the remainder of the nations need the information of both well-being and viability tests. The EC, JMAFF, and EPA guidelines toward biopesticides are created so that it requires less information when contrasted with synthetic items and decreased an opportunity to process the enrollment applications. In this specific situation, the International Organization for Biological Control of Noxious Animal and Plants (IOBC) completed a worldwide level audit on the utilization of biopesticides and administrative environment friendly measures to control pest and diseases. It focused on the requirement for smoothing out the enrollment procedure through orchestrating information necessities and conventions for hazard appraisals. In India, for any microorganisms utilized for bug and illness, the executives require enrollment for both creation and deal with the Central Insecticides Board (CIB) of the Ministry of Agriculture according to the Insecticides Act (IA), 1968, of the Government of India (GOI) and Insecticides Rules, 1971, which were as of late supplemented by the Pesticides Management Bill 2008. The biopesticides for the most part viewed as protected GRAS under this demonstration, and to advance its creation and use, give the advantage of enrollment the same just as temporary enlistment. In this manner, the makers can enlist the item either for customary enrollment under segment 9 (3) or for temporary enrollment under area 9 (3B) of the IA. While applying for enlistment, the information on item portrayal, well-being, toxicology, adequacy, and marking are fundamental. Notwithstanding the need and temporary enrollment for biopesticides in the Act, the enlistment conventions are made simpler and acknowledge nonexclusive information for many new items containing strains that are as of now enrolled. Such certifiable provisions are inbuilt in the Act which shows the enthusiasm of the administration in advancing the sheltered items for a bug, the board like different nations. So as to manage the business creation of these items, the Government of India has built four unique bodies to control the biopesticide creation. The Central Insecticides Board (CIB) is engaged with creating fitting arrangements, and the enrollment panel registration committee (RC) is capable to enlist the items for creation. While the Central Insecticides Laboratory (CIL) is in control to screen the nature of the items accessible in the market, the State Department of Agriculture (SDA) issues the assembling permit and performs the quality check. On the opposite side, according to the warning dated March 26, 1999, of the Central Insecticides Board, Ministry of Agriculture, biopesticide was put under the Insecticide Schedule Act 1968, and thus, the age of toxicological information turned into an essential for the enlistment of biopesticide.

There are severe guidelines and rules directing the usage and treatment of supplement constructed by the microbe. The main reason behind the call of this item is because definition and effective checking are enlistment in which item should meet explicit administrative necessities. Preceding this, the item must be built up in a transporter, for example, alginate (Bashan 2016) or biochar (Głodowska et al. 2016) via these sticking agents at the time of sowing seeds carrying microbial inoculants. On account of fluid microbe inoculum, these are poured and blend over the seeds preceding to planting or else trickled over seeds wrinkle during sowing time. Capacity besides the item's life expectancy is crucial to guarantee microbial

suitability, existence, and/or strain bioactions. There ought to likewise remain lucidity on intense against continuing biomolecule treatment. Much of the time, intense application happens only a few times in a developing period; this could likewise be on an objective phase of crop development, or in light of natural and abiotic situations (dry spell), though in constant treatment, the item can be treated at ordinary period splashes intermission or else a moderate delivery seed treatment (Backer et al. 2018). India stands likely the nation consisting of a maximum comprehensive lawful outline recognized for biofertilizers. Under the request for regulating fertilizers of 1985, the Indian Ministry of Agriculture gave a request in 2006 which was further corrected in 2009, that consists of biofertilizers below the Essential Commodities Act of 1955. Further, the demonstration explains the word biofertilizer as "the product containing transporter based (strong or fluid) living microorganisms which are horticulturally valuable as far as nitrogen fixation, phosphorus solubilization or supplement mobilization, to increment the profitability of the dirt or potentially crop." This word is additionally secured beneath the wide meaning of composts that "means any substance utilized or planned to be utilized as a fertilizer of the dirt and additionally crop" (Malusa and Vassilev 2014).

16.7 Conclusion and Future Prospects

The microbes in the consortium have the capability to provide the best opportunity to enhance crop growth and yield significantly that can be the only way to feed quality food to ever-growing population. The efforts must be taken to identify the compatibility between the microbial strains for the development of a cost-effective product, and this will positively regulate the plant physiology and transcription pattern. Nowadays, there are several bioformulations available in the market as the demand for organic food is increasing to avoid chemical uses in agriculture. The evaluation of microbial count in such formulations is a major problem. Thus, the development of rapid testing kits or methodologies for the evaluation of live microbial count in available products will enhance the quality and doses used during application. The assessment of MC in various field trials for their performance is needed before the preparation of bioformulations. The regulatory and commercialization policies must be strict to develop the microbial formulations in the consortium. Additionally, the metabolites produced from the microbial mixtures in the consortium could be used for enhanced crop production and the modification or upregulation in certain genes that can work in absence of live microbial inoculants. The biotechnological progress in research could be a good way for designing artificially developed MC by editing in the microbial genome.

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