Chapter 1 An Overall Insight Into the Attributes, Interactions, and Future Applications of "Microbial Consortium" for Plant Growth Promotion with Contemporary Approaches



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Abstract Plant-associated microorganisms in the form of microbial consortia play an important role in agricultural production. The use of single strain or individual microorganism-based bioformulation has limitations. Thus, having a microbial consortium, where two or more interacting microorganisms have additive, synergistic, or mutual complementarity in nature, results in the desired effects on plants and soil. In this review, we have discussed the insights of interactions and mechanisms through which an effective microbial consortium promotes plant growth, improves nutrient utilization efficiency, enhances yield, induces tolerance to abiotic stresses, may contribute toward pest and phytopathogen management., etc. within the rhizosphere under their efficient root colonization and biofilm formation. In addition, the activity of microbial consortia has also been highlighted, mainly as a species of plant growth- and health-promoting bacteria. Furthermore, there is a huge impact of microbial consortia on the rhizosphere, which is enhanced by the concept of microbiome engineering and strain improvement. Augmentation of soil with synthetic microbial communities (SynComs), which are extended versions of traditional consortia, is recently being realized as a tool to modulate the complete rhizosphere microbiome for beneficial effects. This article is aimed to explain the wide horizon of the use of microbial consortia that facilitates the sustainable development of agriculture and its applications for human welfare.

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1.1 Introduction

The soil-plant ecosystem's complexity often presents challenges for the single strain bioinoculants, when applied for plant growth promotion and disease control. By combining several microorganisms in the multi-species consortium, multiple beneficial activities are also added, and thus it is assumed that at least this group of microbes, i.e., the microbial consortium, will have more functional traits; hence, they perform better than respective individual microbial isolates (Woo and Pepe 2018; Nuti and Giovannetti 2015).

A microbial consortium is a group of different species of beneficial microorganisms, mainly plant growth-promoting bacteria, that act together as a microbial community. The co-culture of two or more microbial populations interacting synergistically forms a microbial consortium. It can perform diverse functions more efficiently which is difficult (not so efficient) or even impossible to carry out by a single organism. Since the division of labor is quite common in nature, it can be easily characterized in microorganisms established due to microbe-microbe interactions (Tshikantwa et al. 2018). Microbial consortia are "microbial cell factories" representing new synthetic biology approaches (Roell et al. 2019).

The microbial consortium proved effective in plant-microbe interactions, improvement of the soil profile, and soil nutrient status, which is supportive to induce plant growth, plant development in general, and enhancement of crop productivity. The microbial consortium helps in biofertilization, bioremediation, phytostimulation, and biological control of pests and pathogens (Sharma et al. 2018). They act in the solubilization of minerals in the soil, secreting phytohormones, producing enzymes (i.e., ACC deaminase) and chemical metabolites, and contributing to the bio-removal of soil pollutants and heavy metals (Arora et al. 2010; Pandey et al. 2012; Zhang et al. 2019a, b; Santoyo et al. 2021).

The synergism of microorganisms comprises the microbial consortium offering a new scope in agro-practices toward sustainable development. This may also avoid the agricultural requirements of microbial inoculants' trade-off in individual microbial populations.

1.2 Microbe-Microbe Interactions

The interactions between/among the species or strains play a major role in the beneficial effects of bacterial consortia (Singh et al. 2019). The consortium (bearing bacteria) can be classified into three types. It is based upon the effect on each other, for example, (a) positive or stimulatory, (b) negative or inhibitory, and (c) neutral. The positive interactions comprise generating a network supporting individual

members through cross-feeding wherein one bacterium utilizes the metabolic end products as nutrients for another member. Mutualism, protocooperation, and commensalism are some of the features. In mutualism, each or one of the members is benefitted in an obligatory association due to the exchange of required substances or mutual removal of toxins (Roell et al. 2019), while in the case of protocooperation, the interaction that occurs between species is beneficial to the growth rate of both populations but is not required either to persist. Similarly, commensalism is a positive one-way interaction in which one member benefits while the other remains unaffected (Dubey and Maheshwari 2022).

The suppression or inhibitory action of one another leads to the negative interactions that occur due to their growth inhibition of the structure and function. Such processes are (i) amensalism, (ii) predation, (iii) parasitism, and (iv) competition. When the growth of one of the members is affected due to the secretion of inhibitory substances (unidirectional), it is called amensalism, while predation and parasitism involved the growth of one species that depends upon the other species. Competition is mainly due to nutrition or space; therefore, the fast-growing organism dominated. In neutral interactions, members of the consortium do not influence each other. It occurs when two or more species consume different nutrients and neither produces any inhibitory compound to another consortium (Chaneton and Bonsall 2000).

1.3 Microbial Consortia

1.3.1 Definition and Design

A microbial consortium constitutes two or more compatible microorganisms of diverse/similar genera of different species in synergistic or additive interactions (Stockwell et al. 2011; Sarma et al. 2015). Long back, Higa and Parr (1994) advocated the use of effective microorganisms (EM) in the growth promotion of crops. The EM may also contain non-microbial biostimulants and stress-mediated/ stress-protective nutrients. Even less-defined microbial populations originating from the fermentation of various natural substrates, farmyard manure, or composting processes are recommended as inoculants. The microbial consortia contain a network of microorganisms and represent an elegant way to identify specific microbes that have a more central position in the network, often defined as "keystone" species or "hub." Such microorganisms generally co-occur with other taxa and likely exert a strong influence on the structure of microbial communities. The identified "hub" species may act on microbial communities and/or indirectly through (a) cascade modifications in the interconnected microbial network, (b) competition for space and nutrients, (c) alteration of the plant immunity, and (d) modification of the host physiology as identified (Kang et al. 2020). Microbial consortia consists either (i) a synthetic assembly by combining several isolated strains (Puentes-Téllez and Salles 2018) or (ii) complex microbial communities from nature (Skariyachan et al.

2017). In this scenario, the enrichment process is often used to get the desired microbial consortia.

1.3.2 Types, Process, and Development

Bashan and Prabhu (2020) highlighted the formation of advanced consortia with microbe-based products. Two types of consortia, i.e., simple and complex, are based on their differences in fermentation strategy (production of a large population of bacteria to be later formulated into an inoculant). For this, strains are grown individually or in combination including staggering into other species/strains in a suitable medium for all the plant growth-promoting (PGP) organisms. The consistency in results under field conditions is a benchmark of the success of bacterial consortia application which not only depends upon the type and function of strains but also includes their adaptation to adverse climate conditions, survival, and persistence in the soil after application (Verbruggen et al. 2013; Gosal and Kaur 2017).

1.4 Formulations: Difficulties and Success

Consortia formulation can be carried out by using selected PGP bacteria by combining a uniform bacterial cell concentration of all the participating strains. Later, after mixing, inoculant suspensions are prepared to achieve a final bacterial concentration of approximately 10^8 CFU/ml (~OD 600) as described by Gomez et al. (2021). To ensure different genera of PGP bacteria, for consortia formulations, the strain must be evaluated for some traits such as N fixation, P solubilization, siderophore production, IAA production, biofilm formation, ACC deaminase activity, etc. Thus, PGPR selected are recommended to design and construct microbialbased bioformulation for their application in a wide range of agro-ecosystem (Pandey et al. 2005, 2010). Santoyo et al. (2021) described plant growth stimulation by microbial consortia. Although many publications are appearing on plant-microbe interaction, it is significant to note that comparatively few appeared on the use of microbial consortium to perform plant growth and development enhancement to perform a variety of tasks in an ecosystem.

Consortium communication is governed by molecular signals. In this, quorum sensing plays a major role in the compatibility of bacterial communities comprising consortium formulations. Quorum sensing (QS) allows bacteria to switch between two gene expression programs: (i) at low density for individual and social behavior and (ii) at high cell density for social and group behaviors which are preferential for consortia (Ng and Bassler 2009). The QS enables bacterial cells in a formulation to function in unison, and they carry out as a collective, not allowing the desired effect of compatible consortia (Schikora et al. 2016).

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A proper description of the consortium, the taxonomic affiliation of the strains and identification protocols, the process of formulation, the effect of edaphic and other related parameters, and the population of consortia formulations can be carried out by using selected PGP bacteria. The PGP strain is to be evaluated for their nature not to inhibit the growth of each other by the "cross-streaking" method of Pierson and Weller (1994). This was further confirmed by the filter paper disk method as given by Sindhu et al. (1999). The strains are further listed for their consortiumforming abilities following the spectrophotometric method of Shanmugam et al. (2002). In one of the reports from our research group, we have designed different combinations of bacteria, viz., (i) Pseudomonas aeruginosa KRP1 + B. licheniformis KRB1, (ii) B. licheniformis KRB1 + Sinorhizobium meliloti RMP1, (iii) S. meliloti RMP1 + P. aeruginosa KRP1, and (iv) KRP1 + RMP1, a multi-species bacterial consortium of all the above strains (Maheshwari et al. 2010). The healthy seeds of Brassica campestris (Indian mustard) were bacterized with KRP1, KRB1, or RMP1 and by consortia as given above, and the maximum enhancement of vegetative growth parameters was observed in the consortium, in comparison to those that emerged due to individual treatment with KRP1, KRB1, or RMP1. This application of bacterial consortium proved to be most desirable for plant growth and development of *B. campestris* (Maheshwari et al. 2010).

According to Nuti and Giovannetti (2015), microbial consortia are based on multiple PGP microbial strains with complementary properties. Sometimes non-microbial biostimulants and stress-protective nutrients are added to reduce the product cost. Molina-Romero et al. (2021) observed the potential of a second-generation consortium formulated with *Azospirillum brasilense* SP7, *Pseudomonas putida* KT2440, *Acinetobacter* sp. EMM02, and *Sphingomonas* sp. OF-178A. The bacterial strains present in the consortium proved compatible and efficient for field applications and resistant to desiccation.

1.5 Root Colonization and Biofilm Formation

PGPR-plant interaction is an intricate and interdependent relationship that involved not only the microorganisms but also other abiotic and biotic factors of the rhizosphere region that also play a role in their successful partnership (Kshetri et al. 2015). Root colonization and biofilm formation by the microbial community and the underlying principles are also behind the success of these organisms to tide over unfavorable conditions as suggested (Dutta and Podile 2010). The nature of bacterial genera and their relationship with host plants are exhibited by aggressive root colonization due to adequate adhesiveness to its surface. The adhesion improved when the strains of *Azotobacter brasilence*, *Acinetobacter* spp., and *Sphingomonas* spp. were applied to *Zea mays* together in a consortium. The inoculation of the bacterial consortium also improves the root colonization capacity in comparison to that of individual treatments. De Oliveira et al. (2006) observed the root colonization of a consortium formulated with *Gluconacetobacter diazotrophicus*, *Herbaspirillum* *seropedicae*, *H. rubrisubalbicans*, *Azospirillum amazonense*, and *Burkholderia tropica*. Even the different isolates of *Burkholderia* sp. RHT8 and RHT12 led to synergism and root colonization in fenugreek's rhizosphere (Kumar et al. 2017). The combined effects of rhizo-competitive rhizosphere and non-rhizosphere *Bacillus* species enhanced the growth and yield in *Eleusine coracana* (Dheeman et al. 2020).

Root zone or "rhizosphere effect" is pronounced due to the successful establishment of bacterial consortia. This phenomenon is a crucial step to obtaining the beneficial effect of consortia on the host plant, which is further improved due to adequate adhesion and colonization (Shahzad et al. 2013). A significant difference was seen in maize when inoculated with A. brasilense, P. putida, Acinetobacter, and Sphingomonas spp. together. The plant taxa, variety, and other morphological also supportive of bacterial colonization formulated features are with Gluconacetobacter diazotrophicus, Herbaspirillum seropedicae. H. rubrisubalbicans, Azotobacter amazonense, and Burkholderia tropica in sugarcane (De Oliveira et al. 2006). The colonization capability of consortia of P. striata and Piriformospora indica is also dependent on corn varieties as observed by Singh et al. (2009). Earlier, Gusain and Bhandari (2019) studied the root colonization of Sinorhizobium meliloti, A. chroococcum, Serratia marcescens, and P. aeruginosa in different combinations of consortia which showed quite effective colonization in comparison to their counterparts. Santoyo et al. (2021) described plant growth stimulation and root colonization by microbial consortia.

The bacterial biofilm formation occurs quite commonly on the root surface and represents a hotspot for microbial interactions assisting them to form a consortium. It plays a significant role in the ecological network for shaping microbial communities for playing their role in sustainable agrobiological practices. The desired role of microbe-microbe interaction or mixed consortium involved in stimulating ecosystem functioning as well as in the enhancement of plant productivity (Pandit et al. 2020).

Currently, bioinformatics tools have been devised and used to investigate intermicrobial co-occurrence networks from community profiling or metagenomic data (Faust and Raes 2012); Layeghifard et al. 2017) study of the microbial networks. Plant interaction tends to indicate that positive correlation dominates among microbes from the same kingdom, whereas negative interaction primarily occurs through inter-kingdom microbe-microbe interaction (Agler et al. 2016). Thus, the role of microbial consortia is complex, and a more holistic understanding of microbial networks for holobiont fitness, is required (Hassani et al. 2018).

Aggressive bacterial genera in the root rhizosphere must have adequate adhesion and root colonization. Molina-Romero et al. (2021) highlighted that the adhesion improved when strains of *A. brasilense*, *Acinetobacter* sp., and *Sphingomonas* sp. were applied to maize together in a consortium. The inoculation of the bacterial consortium improved the bacterial colonization capacity in comparison to that of individual treatments. In another study, De Oliveira et al. (2006) observed the colonization of a consortium formulated with *Gluconacetobacter diazotrophicus*, *Herbaspirillum seropedicae*, *H. rubrisubalbicans*, *Azospirillum amazonense*, and *Burkholderia tropica*. In addition, the bacterial consortium offered an alternative allowing the efficient use of half of the recommended dose of nitrogen fertilizer. The use of the consortium allowed the lowering of a 50% mineral N application and generated beneficial agronomic practices along with the lower cost to the cultivars (Molina-Romero et al. 2021). Recently, a new approach is devised wherein the effect of microbial consortia is applied as fertilizer coating. For this, the use of illumine high-throughput sequencing (HTS) is involved to influence the bulk soil and rhizosphere microbial community applied to potato fields (Overbeek et al. 2021). On the other hand, bacterial consortium acts as a substitute to chemical fertilizers such as urea, DAP, etc. because decreased application of chemical fertilization along with bacterial consortium exhibited a similar effect on plant growth and yield as revealed while applying the recommended doses of chemical fertilizers (Kumar et al. 2010; Da Costa et al. 2013).

Other characteristics such as strain evaluation to salinity stress under drought resistance cannot be ruled out, to stimulate crop growth and improve tolerance to abiotic stresses, and prove more effective in extreme climate change conditions. Microbial inoculants may improve salt tolerance by altering hormonal root-shoot signaling that manages IAA production in plants by bacterial action, thus having the potential in enhancing salt tolerance (Etesami and Maheshwari 2018). Such an approach is beneficial for a realistic assessment of the potential of microbial consortia in a climate change world.

1.6 Abiotic Stress: Action and Mechanism

The application of microbial consortia can reduce the negative effects that arise due to abiotic stress conditions on crops. But for their effective application in the crops, novel approaches are required to explore bacteria-bacteria and plant-bacteria interactions or bacteria-fungi interactions. Isolating and identifying the stress-tolerant or stress-resistant microbes to recalcitrant agrochemicals and heavy metals is important (Xia et al. 2020; Katiyar et al. 2021).

Abiotic stresses inhibit plant growth and development due to oxidative damage attacking DNA and cellular membranes. The antioxidant enzymes neutralize the reactive molecules; thus, cells are protected. PGPB having catalase and peroxidase properties are proven more protective. The beneficial bacteria also produce trehalose which also benefits the plants to abiotic stress (Glick 2015; Kumar and Verma 2018). Microbial production of phytohormones also protects plants by the involvement of various physiological actions. PGPB induces the level of proline in plants. Proline scavenges reactive oxygen molecules and acts to stabilize proteins through molecular chaperons in stress conditions (Meena et al. 2019). The effect of the consortium of *Bacillus cereus* AR156, *B. subtilis* SM21, and *Serratia* sp. XY21 was reported to develop healthy cucumber plants, with much darker green leaves containing increased proline and chlorophyll contents, and induce superoxide dismutase activity (Wang et al. 2012). An increase in ethylene level is injurious to plants causing

senescence and other deleterious effects which occur due to the accumulation of a consortium of ACC deaminase-producing bacteria (*Ochrobactrum pseudogrignonense*, *Pseudomonas* sp., and *B. subtilis*) that significantly increased early vegetative growth plant parameters in *Vigna mungo* and *P. sativum*.

As human populations continue to increase, the disturbance of the soil ecosystem to enhance productivity may place greater demand on supplying soil essential nutrients. Therefore, it is essential to increase the understanding of the biological, physical, and chemical properties of soil along with the soil-microbe-plant relationship to enhance productivity with available nutrient pools (Millard and Singh 2010). The soil native ability to supply sufficient nutrients continuously decreases and emerges as a greater challenge for enhancing the productivity of crops and the quality of water, air, and fragile soil ecosystems. The relationship of soil-plant-microbes especially soil interaction influences plant compounds accurately, identifies the yield-limiting potential factors and growth and development, and minimizes the influence of those to manage the enhancement productivity (Metcalfe et al. 2011). Most of the research during the last decades was focused on the use of fertilizers and manures. Thus, information on the integrated approach of plant nutrition on the sustainability of soil fertility and crop productivity is necessary.

Soil fertility is the status or the inherent capacity of the soil to supply nutrients to plants in adequate amounts in suitable proportions. On the other hand, soil productivity is the capacity of the soil to produce crops with a specific system of management and is expressed in terms of yields (Van Ittersum et al. 2013). All productive soils are fertile, but all fertile soils are not necessarily productive. To produce crops of economic value and to maintain the health of the soil without deterioration is most important. Modern farming, driven by economic constraints, is forced to use artificial fertilizers, often to the detriment of the soil's natural fertility (Rana and Rana 2011).

1.7 Metagenomics and Biotechnological Approach to Increase Efficiency of Microbial Consortium for Plant Growth Promotion

When compared to single microorganisms, consortia are superior throughout many situations. The selection of consortium members in a way that maximizes performance is a significant obstacle. Microbial consortia have the advantage of being more adaptive to environmental changes due to their high stability, resilience, and multifunction. Human health, bioremediation and biodegradation, chemical and bioenergy generation, and food manufacturing are just a few of the areas where microbial consortia are playing crucial roles in the developing sector (Lee et al. 2013). Recent breakthroughs in synthetic biology have significantly enhanced both the synthesis of microbial consortia and the comprehension of microbial communication mechanisms (Song et al. 2014). Cell-cell interactions in relatively small

synthetic microbial consortia have recently been studied. Synthetic microbial consortia are typically less complex and easier to genetically modify than real microbial consortia, making the interaction and control processes easier to explore (Sanchez-Gorostiaga et al. 2019).

1.7.1 Microbiome Engineering

Microorganisms found on or within a plant have been shown to have beneficial effects, such as promoting growth or inhibiting pathogens (Ab Rahman et al. 2018). Altering the microbiome with plant growth-promoting rhizobacteria (PGPR) can improve plant development and reduce infections and abiotic stress (Kumar et al. 2018). Microbiome engineering can enhance agricultural yields and resilience by manipulating the plant holobiome. The plant's genotype is also very important for the formation and function of rhizospheric microbiomes and for getting the most out of PGPR (Arif et al. 2020). Beneficial interactions between plants and microbes have been studied to learn how to change plant genomes to attract and keep beneficial microbiomes. Different plant genotypes attract helpful and disease-suppressing microorganisms to varying degrees, reorganizing the microbiome assembly (Gao et al. 2021). The endophytic microbiome of plants also influences functional genes related to plant growth promotion (Singha et al. 2021).

The study of plant functional genomics during mutually beneficial plant-microbe interactions has allowed the manipulation of plant genomes to entice and sustain such microbiomes (Rosier et al. 2018; Vandana et al. 2021). This led to the idea of "designer plants." These genetically modified plants can release hormones or other substances that attract and keep good microbiomes (Stringlis et al. 2018). The targeted crop's yield can be dramatically increased through the application of a consortium that is compatible with the chosen plant and able to repair the rhizospheric microbiome (Tabacchioni et al. 2021). Several studies have also pointed out that wild-type relatives of domesticated crops can help us learn more about the role of genes in wild plants that are linked to microbiome assembly (Pérez-Jaramillo et al. 2018). The microbiome is often commonly referred to as the brain of a given environment because of the significant impact it has on the general health and well-being of that environment (Lavazza and Sironi 2019). Inoculated groups of microorganisms can rebuild the structure and function of the microbiome in plants and soil. Microbes create functional consortia in the rhizosphere; soil conditioning and important microbial strains can modify the rhizosphere microbiome's structure (Voges 2019). It is feasible to create artificial consortia with several functions for promoting plant development. This could fix some of the problems with traditional microbial biofertilizers, like not getting along with the host, not being able to compete well with native microbes, and not being able to adapt to the local environment (Hart et al. 2018). The development of the optimum artificial microbial consortium involves studying the microbes' origin, getting and cultivating the microorganisms, optimizing microbial interactions as per compatibility, and finally

investigating the consortia's performance. Microbiome breeding is another technique by which the microbiome can be altered for betterment. It requires allowing the host to filter which populations of bacteria are permitted to interact with it and will be passed straight to their progeny, thus indirectly affecting the microbiome (Mueller and Linksvayer 2022). This strategy involves spreading a microbiomeinfluenced phenotype of the host. For example, to study the microbial influence on the flowering pattern of Arabidopsis thaliana, the early and late flowering microcosms are studied over generations, and it was found that more phenotypic inflorescence was observed in the plants inoculated with microbiome from late flowering plants. The repeatability of flowering phenotypes shows that microbiomes can be regulated to influence plant characteristics and coordinate soil resource pools (Panke-Buisse et al. 2015, 2017). In the same study, an increase in total biomass and increased enzyme activity for the mineralization of nitrogen were observed in Brassica rapa when inoculated with the same (Panke-Buisse et al. 2015). Likewise, microbiome transformation is another technique where the beneficial microbiota from one species was inoculated in other species to promote plant growth (Arif et al. 2020). For example, Leptospermum scoparium is reported to release antibacterial agents to counter the growth of Pseudomonas pathogens. A similar biocontrol activity was observed in the kiwi plant when PGP bacterial microbiome from this species was inoculated in it (Wicaksono et al. 2018).

1.7.2 Molecular Tools to Increase Efficiency of Microbiome Engineering

Understanding the physiological and biochemical functionality of the consortium can be greatly aided by genetic engineering or the use of molecular tools in the microorganism involved with plant growth promotion. The extensive collection of genes that are engaged in the processes will be taken into consideration as potential targets to achieve an accurate comprehension of the function that each gene is carrying out (Kumar et al. 2020). The discovery of RNA interference (RNAi) and CRISPR is the most recent and commonly used biotechnological development in genetic tools in this regard (Boettcher and McManus 2015; Schultenkämper et al. 2020). RNAi relies on an endogenous process that regulates gene expression with short RNAs. Synthetic tiny RNAs (siRNAs or short hairpin RNAs) can be used to seize the indigenous RNAi mechanism. Either way, the inserted RNA is put into the RNA-induced silencing complex (RISC), which promotes target mRNA destruction (Carthew and Sontheimer 2009; Mohr et al. 2014). Reduced amounts of the target protein are the result achieved post-translationally by targeting the expression of the corresponding mRNA (Boettcher and McManus 2015).

CRISPR is a revolutionary way to change the genes of plants to improve specific traits, and thus it has become one of the most useful tools in the field of functional genomics (Pérez-Jaramillo et al. 2018). One important use of CRISPR-based genetic

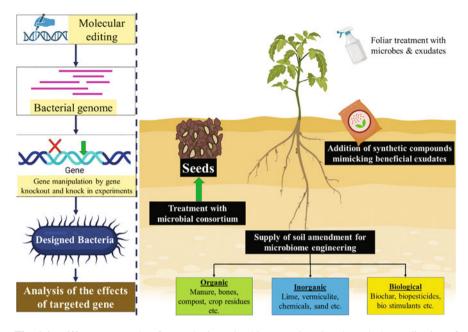


Fig. 1.1 Different approaches for employing microbiome engineering through the application of consortium

engineering tools is to alter the genes of plants or microbes to study how the genes work. One of the best things about the CRISPR tool is that it can completely shut down the target gene. To do this, designer plants could be genetically engineered using the CRISPR tool to make and release mass hormones or exudates that attract and keep beneficial microbial populations in the rhizosphere microbiome ecosystem (Bisht et al. 2019) (Fig. 1.1).

1.7.3 Next-Generation Microbial Synthetic Communities (SynComs) for Plant Yield Promotion

From basic natural or synthetic consortia to a more complex applied consortium of microbes, omics-based techniques can paint a comprehensive picture of a consortium's operation. Based on single omics approaches, metabolomics, metagenomics, transcriptomics, and metaproteomics have been developed to make it easier to study groups of microbes (Chandran et al. 2020). Quantifying the meta-proteome in a group of microorganisms is important for understanding how different protein functions work together and how they change over time which makes it the best of the different meta-omics approaches at showing how a microbial consortium's system works (Franzosa et al. 2015). Rapid developments in mass spectrometry

have led to the creation of many quantification strategies. Among these techniques, the isobaric leveling method and isobaric tags for relative and absolute quantitation (iTRAQ) are widely utilized for comparative proteomic research because of their high sensitivity and accuracy. The metabolomic analysis of the consortium dedicated to any process gives a prediction of how the combined effects the production of intermediate metabolites so that the best can be grouped to obtain the best results (Ma et al. 2019).

SynComs are microbial consortia designed to imitate the natural microbiome. The goal is to minimize the sophistication of the microbiome while keeping some of the natural interactions between bacteria and hosts, offering a spectrum of capabilities unattainable by a single bacterium. Additionally, synergistic interactions between members of SynComs may improve community stability (Kaminsky et al. 2019; McCarty and Ledesma-Amaro 2019). To unlock the potential of soil microbes and boost agricultural yields, microbial synthetic communities (SynComs) have been proposed as a useful technique that incorporates both microbial ecology and genetics in the construction of inoculants. The goal of this strategy is to identify and then recruit a group of microbes that can stimulate plant development in a variety of climates and the face of harsh events (York 2018). In recent times, the focus has been given to the development of microbially based goods due to the worldwide potential of these SynComs to boost agricultural production and sustainability (Singh et al. 2020).

Computational approaches, such as machine learning algorithms, will improve the screening and identification of beneficial bacteria, as well as the process of establishing the optimal microbe combination for a particular plant phenotype (Harfouche et al. 2019). The growing number of reference genomes and metagenomes in public databases helps to find bacteria with desirable features, and by using these genomic information and gene expression patterns, one can choose microorganisms with plant-beneficial functional features or metabolic capabilities (Vorholt et al. 2017). Genome surveys for several gene markers will be critical to finding relevant microorganisms because important properties like colonization efficiency and frequency of other attributes are likely to relate to multiple genes, and to solve this problem, genomics-based datasets filter microbiological candidates on a genomic markers' basis (Finkel et al. 2017). Thus, genome and metagenome sequencing, together with microbial characterization, could assist in building SynComs that bestow stable plant phenotypes and increase plant colonization and permanence (De Souza et al. 2020). A systematic flow to develop a successful SynCom is presented in Fig. 1.2.

In another way, SynComs help in understanding the physiology and function of microorganisms and the parameters regulating community assembly by manipulating a SynCom formulation by adding, removing, or replacing microorganisms (Vorholt et al. 2017). For example, removing a single strain of *Enterobacter cloacae* reduced the activity of microbial consortium which was related to reducing the severity of maize blight disease (Niu et al. 2017). Similarly, a SynCom with more microorganisms from the *indica* strain had a bigger effect on rice growth than a SynCom with more microorganisms from the *japonica* strain (Zhang et al. 2019a, b).

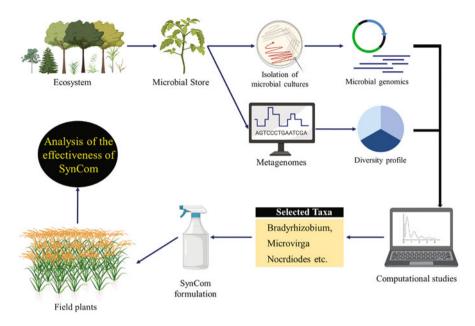


Fig. 1.2 A blueprint for the customization of stable and efficient synthetic microbial communities (SynComs), with the goal of increasing the resistance of crops to environmental challenges and yield production

Few obstacles or problems exist with the employment of SynComs. Most microbial species are likely to be uncultivable, making it difficult to assemble in a microbial consortium. Further, the cost of sequencing hundreds of thousands of samples is extremely expensive (Lewis et al. 2021). In addition, soil microbiomes are complicated, and the relationships between soil single taxa and environmental factors are inadequately documented, limiting our understanding of microbial candidates that might be employed to increase plant growth and productivity in the wild (Jayaraj et al. 2016). Therefore, most of the recently developed SynComs are comprised of bacteria only and a group of culturable microbes equipped with good plant growth-promoting dexterities.

1.8 Application: Microbial Inoculation and Soil Community

Microbial inoculation directly impacts the soil microbial community to increase the relative abundance of inoculated microbial genera. The rhizospheric microbial community composition differed substantially from the bulk soil microbial community composition (Overbeek et al. 2021). For example, in the case of potato roots, enrichment of the rhizosphere community over bulk soil was observed for

Proteobacteria and Eurotiomycetes. A similar difference in the microbial community was also observed by several workers (Berendsen et al. 2012; Xue et al. 2018). The external input of microorganisms closely associated with the rhizosphere contributed as core microorganisms and the alteration in the rhizospheric microbiome help in designing microbial inoculants beneficial to the plants growing under a variety of soil conditions (Sathya et al. 2017).

The biological management for the growth and development of plants is still at an early stage of development, while the approach appears to have tremendous potential, and many of the basic concepts necessary for the implementation are in place, and apparent obstacles such as information on biomass, formation of a product, site of application, and registration difficulties exist (Kumar et al. 2017). For increasing crop productivity and the maintenance and improvement of soil fertility for sustainable crop production, the multifunctional formulation may be promoted that involves microbial consortium utilizing the PGPR, which has been proven better and eco-friendly in comparison to that of formulation alone (Kshetri et al. 2017).

The microbial consortia are also used to control and optimize various industrial processes. Puentes-Téllez and Salles (2018) described the construction of effective minimal active microbial consortia for lignocellulose degradation. The simplification of the microbial community makes it easier to help and understand the individual roles of the strains in the consortia.

Skariyachan et al. (2018) worked on polymer degradation by novel thermophilic consortia of *Brevibacillus* spp. and *Aneurinibacillus* sp. associated with waste management landfills and sewage treatment plants. Earlier, the authors formulated bacterial consortia from plastic-contaminated cow dung. It is interesting to note that Subhashchandrabose et al. (2011) studied the biotechnology potential of consortia of cyanobacteria/microalgae and bacteria.

The PGP strain was evaluated for their nature to inhibit the growth of each other by the "cross-streaking" method of Pierson and Weller (1994). This was further confirmed by the filter paper disk method as given by Sindhu et al. (1999). The strains are further listed for their consortium-forming abilities following the spectrophotometric method of Shanmugam et al. (2002). Recently, Baliyan et al. (2022) reviewed the bacteriophage cocktails and antibacterial agents in crop protection.

1.9 Conclusions

The development of artificial consortiums developed with multifarious characteristics is a growing interest in using similar or diverse genera of beneficial bacteria in agriculture applications. The microbial consortia offer consistency and higher reproducibility of data under various environmental conditions and provide a broader array of the mechanism of action in comparison to that of individual beneficial bacteria applied alone for friendly crop production and protection system in agriculture. However, some critical challenges are yet to be resolved. Certain issues with the registration and marketing of formulations comprising mixed cultures limit their potential use in modern agriculture. The difficulties in understanding the specific role of each component of microbial consortium and their desirable effects may limit the predicted effect on the growth and development of crops. The molecular tools and biotechnological approach involving plant-microbe interaction, soil-microbe engineering, metagenomic soil profile, and next-generation synthetic microbial consortia are some of the most useful tools to make it easier to study microbe involved in the design and construction of microbial consortium systems. Further, bioinformatics and computational tools may improve the understanding of function of microbial consortia and their products for sustainable agriculture.

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