



## Soil Microbes as Bio fertilizer: Sustainable Agriculture and Environments

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### Abstract

The worldwide increase in human population raises a big threat to the food security of each people as the land for agriculture is limited and even getting reduced with time. Therefore, it is essential that agricultural productivity should be enhanced significantly within the next few decades to meet the large demand of food by emerging population. Not to mention, too much dependence on chemical fertilizers for more crop productions inevitably damages both environmental ecology and human health with great severity. Exploitation of microbes as bio fertilizers is considered to some extent an alternative to chemical fertilizers in agricultural sector due to their extensive potentiality in enhancing crop production and food safety. It has been observed that some microorganisms including plant growth promoting bacteria, fungi, Cyanobacteria, etc. have showed bio fertilizer-like activities in the agricultural sector.

Extensive works on bio fertilizers have revealed their capability of providing required nutrients to the crop in sufficient amounts that resulted in the enhancement of crop yield. Soil management strategies today are mainly dependent on inorganic chemical-based fertilizers, which cause a serious threat to human health and the environment. Bio-fertilizer has been identified as an alternative for increasing soil fertility and crop production in sustainable farming. The exploitation of beneficial microbes as bio-fertilizers has become of paramount importance in agricultural sector due to their potential role in food safety and sustainable crop production. Efficient plant growth-promoting microorganisms (PGPMs) solubilize the nutrients in soil and facilitate absorption by plants and consequently enhance the plant growth and yield. PGPMs also sustain the soil fertility, soil health, and nutrient mobilization efficiency under sustainable agriculture. Current soil management strategies are mainly dependent on inorganic chemical-based fertilizers, which caused a serious threat to human health and environment. The exploitation of beneficial microbes as a biofertilizer has become paramount importance in agriculture sector for their potential role in food safety and sustainable crop production. The eco-friendly approaches inspire a wide range of application of plant growth promoting rhizobacteria (PGPRs), endo- and ectomycorrhizal fungi, cyanobacteria and many other useful microscopic organisms led to improved nutrient uptake, plant growth and plant tolerance to abiotic and biotic stress. The knowledge gained from the literature appraised herein will help us to understand the physiological bases of biofertilizers towards sustainable agriculture in reducing problems associated with the use of chemicals fertilizers.

**Keywords:** Biofertilizer; Soil fertility; Crop productivity

### Introduction

A biofertilizer is a substance which contains living microorganisms which, when applied to seeds, plant surfaces, or soil, colonize the rhizosphere or the interior of the plant and promotes growth by increasing the supply or availability of primary nutrients to the host plant<sup>[1]</sup>. Biofertilizers add nutrients through the natural processes of nitrogen fixation,

solubilizing phosphorus, and stimulating plant growth through the synthesis of growth-promoting substances. Biofertilizers can be expected to reduce the use of synthetic fertilizers and pesticides.

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The microorganisms in biofertilizers restore the soil's natural nutrient cycle and build soil organic matter. Through the use of biofertilizers, healthy plants can be grown, while enhancing the sustainability and the health of the soil. Since they play several roles, a preferred scientific term for such beneficial bacteria is "plant-growth promoting rhizobacteria" (PGPR). Therefore, they are extremely advantageous in enriching soil fertility and fulfilling plant nutrient requirements by supplying the organic nutrients through microorganism and their by products. Hence, biofertilizers do not contain any chemicals which are harmful to the living soil.

Worldwide food demand is increasing rapidly and so more in developing nations where crop lands and resources hardly contribute to an efficient crop production needed to meet such an urgent demand for food. There is a need to intensify agricultural production in a sustainable manner through use of efficient agro-biosystems which consider the entire agroecosystem bio-chemical diversity and their potential to mitigate the adverse impacts of low soil fertility, abiotic stress, pathogens, and pests [2, 3]. In this context, global food security issue will foster reliance on innovation, development, and delivery of technologies that lead to increased food production while ensuring sustainable intensification of agriculture. A number of innovative and efficient technologies has been adopted such as smart irrigation systems, smart fertilizers [i.e., controlled release fertilizer and enhanced efficiency fertilizers (EEFs), etc.], integrated fertilization, and diseases biocontrol strategies as well as diverse imaging- and sensing-based technologies that provide highly valuable information for monitoring and securing crop productivity. Agricultural microbial biotechnology through the integration of beneficial plant-microbe and microbiome interactions may represent a promising sustainable solution to improve agricultural production [3]. For instance, advances in genomic, post-genomic, biochemistry, ecology, and symbiotic interactions of beneficial microbial strains have led to the development and commercialization of efficacious microbial products with proven success to improve crops' yield and adaptation to

environmental changes, and inputs of carbon and energy [4,5].

Today, microbial-based biofertilizers are considered to be among key agricultural components that improve crop productivity and contribute to sustainable agro-ecosystems. It is a component that aggregates a variety of microbial-based bio-products whose bioactivities are essential to stimulate and improve biological processes of the intricate plant-microbe-soil continuum [6]. Different kind of soil microorganisms that exhibit PGP traits can be used for the production of efficient biofertilizers [1, 7, 8,9].

Generally, beneficial rhizosphere microorganisms can boost plant growth via multiple regulatory biochemical pathways that include manipulating the plant hormonal signaling, preventing pathogenic microbial strains and increasing the bioavailability of soil-borne nutrients [10,11, 12, 13, 14]. Direct mechanisms generally facilitate resource (i.e., N, P, K, and essential micronutrients) acquisition, modulate plant hormone biosynthesis, and various molecules either extra-cellularly in the vicinity of rhizosphere (i.e., siderophores) or intra-cellularly such as aminocyclopropane-1-carboxylate deaminase which facilitate plant growth and development by decreasing ethylene levels, and alleviating osmotic (salinity and drought) stress in plants [15, 16]. Indirect mechanisms by which rhizosphere microorganisms could promote plant growth are mainly involved in decreasing the inhibitory effects of various phytopathogens through acting as biocontrol agents [12, 17] via antimicrobial metabolites biosynthesis (i.e., hydrogen cyanate, phenazines, pyrrolnitrin, 2,4-diacetylphloroglucinol, pyoluteorin, viscosinamide, and tensin, etc.), competition to nutrients and the elicitation of induced systemic resistance [18,19] which may occur due to a beneficial interaction of some rhizobacteria with plant roots resulting in plant resistance against some pathogenic microorganisms.

The positive impacts of microbial-based biofertilizers on growth and yield of staple crops may be limited to a single nutrient element such as N-fixing bacteria, but also to several nutrients, due to arbuscular mycorrhizal fungi [20]. Moreover, the development of microorganisms'

consortium which is a polymicrobial mixture that contains several microbial strains belonging to different functional groups may strongly promote plant growth, yields, and healthy agro ecosystems [21, 22]. Success in constructing effective polymicrobial formulations with multiple modes of action depends on how functional, complementary, and synergic the candidate strains are [22, 23]. For example, inoculation with mixed cultures of *Penicillium* spp. and AM fungi induced positive and synergistic effects (especially enhanced plant nutrition and growth) in cereals and legumes [24, 25, and 26]. Such positive impacts on legume crops have also been observed when co-inoculating with *Rhizobium* spp. and *Penicillium* [27, 28], rhizobia with AMF [29, 30], *Rhizobium* and P solubilizing-bacteria [31], or even with the tripartite inoculation with AMF-*Rhizobium*-P-solubilizing fungus [32, 33]. Multifunctional microbial consortia may also involve free-living NF bacteria as well as different PGP rhizobacteria with higher abilities to maximize plant growth, yield and efficient N uptake [34, 35, 36, 37, and 38].

Both basic and applied research on screening, designing, testing and validating potential microbial resources for their beneficial impacts on agriculture have gained global interest. Particularly, NF bacteria including both symbiotic and non-symbiotic and P solubilizing/mobilizing microorganisms have increasingly been used as biofertilizers, and now account for more than 75% of globally marketed microbial-based biostimulants. These segments are expected to grow by 20 and 13% for the P-solubilizers and N<sub>2</sub>-fixers segments, respectively [3, 39, 40, and 41]. Given their importance for promoting sustainable agriculture, these microbial-based bio stimulants need to be more deeply explored in combination with multiple nutrient resources such as mineral fertilizers and relevant agricultural practices in order to develop effective integrated strategies that sustain crop production and soil fertility. This is a highlight the importance of the latter nutrients- N- and P-supplementing microorganisms in a context of promoting sustainable agriculture owing to their specific metabolic functionalities to increase use of essential nutrients (P and N) by major crops such as cereals and legumes. Furthermore, recent

knowledge on the dual use of the microbial and mineral nutrient resources with peculiar emphasis on P fertilizers was presented as an example of positive IPNMS that may lead to a profitable “microbial/mineral” inputs marriage.

The intended to x-ray the role of bio fertilizers in sustainable agriculture thereby meeting the needs of agriculturists and plant biologists whose work focuses on creating clean and efficient means of to improving soil quality by nourishing and maintaining the useful and natural flora of microorganisms. Furthermore, it presents recent developments in the field of agricultural management that reveals the potentials of the application bio fertilizers in terms of increased nutrient profiles, plant growth and productivity and an improved tolerance to environmental stress.

#### **Nutrients necessary for plant growth**

Plants require a great number of elements (macronutrients: N, P, K, Ca, Mg, S, C, O, H and micronutrients: Fe, B, Cl, Mn, Zn, Cu, Mo, Ni) and their com-pounds for growth and development. Those elements are used to create and maintain the cells and the necessary life processes such as growth, reproduction, respiration and photosynthesis. As a part of photosynthesis, plants create various forms of polysaccharides, lipids, proteins and other organic molecules [42]. For normal functioning, the plant absorbs various chemical elements from the soil, which in combination with water and carbon-dioxide form compounds [43].

Numerous elements are widespread in the soil in forms that plants cannot assimilate. Because of that, the basic precondition for a plant to uptake certain element is its bioavailability [43]. Water and nitrogen are considered to be the most important factors for development of a plant [44]. Plants are only able to absorb nitrogen in the form of nitrate and ammonia, while molecular form from the air remains unavailable to them. When the level of nitrogen in the plants is low vegetation is limited, leading to reduced productivity [45]. Besides that, phosphorus usually originates from insoluble phosphate rock formations, and in spite of a large amount of phosphorus in the soil (400-1200 mg/kg), only a small part of it is available to the plant for plant metabolism processes. Phosphorus deficiency may cause slower growth of the plant

and reduced leaf biomass<sup>[46, 47]</sup>. Similarly, potassium regulates enzymatic reactions, salt stress resistance, stomata functions, photosynthesis and carbohydrate transport<sup>[48]</sup>. Lack of potassium in soil may cause plant functions disorder, resulting in poor crop quality<sup>[49, 50]</sup>. And furthermore, plant productivity is significantly influenced by the presence of other elements and plant hormones, thus, it is quite clear that every plant should be supplied with a sufficient amount of every nutrient. The long-term practice of enriching the soil through the use of chemical fertilizers has proven to be quite unfavorable for the environment and has led us to alternative solutions that will provide plants with the necessary compounds<sup>[51]</sup>. One of the more popular approaches is the use of microorganisms that promote plant growth, and their incorporation into microbial fertilizers which, when applied to seeds, plant itself or incorporated in the ground may provide all the nutrients plant need<sup>[52]</sup>.

#### **Plant growth promoting microorganisms (PGPMs)**

As previously stated, PGPMs naturally inhabit the rhizosphere, favorably affect the plant, improving its productivity and resistance to pathogens<sup>[52]</sup>. Some of the PGPMs, which in general can be divided into bacteria and fungi, include the following strains: *Azospirillum*, *Azotobacter*, *Rhizobium*, *Pseudomonas*, *Enterobacter*, *Bacillus*, *Paenibacillus*, *Klebsiella*, *Flavobacterium*, *Gluconobacter*, *Penicillium*, *Trichoderma* and *Streptomyces*<sup>[53]</sup>.

Microorganisms such as *Rhizobium*, *Klebsiella*, *Clostridium*, *Bacillus megaterium*, *Penicillium* sp., *Trichoderma viride* improve growth and crop yield, while *Pseudomonas aureofaciens*, *Trichoderma*, *Streptomyces* sp. may act as biocontrol agents against pests and plant disease<sup>[54]</sup>.

Based on their interaction with the plant, plant growth promoting bacteria (PGPB) can be divided into symbiotic or free-living bacteria<sup>[51]</sup>. They can further be divided into extracellular and intracellular. Extracellular PGPBs inhabit the space of the rhizosphere, the root surface or the intracellular space of the root cells, while the intracellular PGPBs inhabit the root cells, penetrate the cell wall and integrate with the plant, forming new organ on the plant tissue – nodule,

that provides optimal conditions for the bacteria<sup>[55,56]</sup>. On the other hand, plant growth promoting fungi include arbuscular mycorrhizas (AM), ectomycorrhizae (EcM) and root fungi such as *Penicillium*, *Trichoderma* and *Aspergillus*.

They produce organic acids and enzymes that inhibit pathogens or dissolve insoluble compounds<sup>[56]</sup>.

Knowing that every plant has a defense system against pathogens, it is interesting to discuss the way plants actually detect and distinguish beneficial microbes from a pathogenic kind. It is believed that every plant has a receptor with microbe-associated molecular patterns which are the key elements in a plant-microbe communication<sup>[57]</sup>. In that process, different signaling mechanisms are involved (chemoattraction, initiation of the nodulation process, release of volatile compounds etc.) and a variety of chemical compounds (organic acids, sugar, flavonoids, volatiles) are released. Presence of a certain compound is actually signal for starting off root colonization or nodule forming process. After colonizing the plants root, bacteria start to show their beneficial effects<sup>[58]</sup>.

There is a wealth of literature covering different possibilities of inoculation of wide range of plants, even in horticulture and fungiculture<sup>[59,60]</sup>. Rhizobia is used by great number of commercialized fertilizers designed for legume crops, although some studies show other choices might be successful as well<sup>[61]</sup>. For example, *Pseudomonas aeruginosa* was used to promote growth of faba beans and common beans at the same time preventing root rotting caused by *Fusarium culmorum*<sup>[62]</sup>. Numerous researches show that inoculation of maize or wheat provides excellent results. Different species have been tested out (*Burkholderia capacia*, *Bacillus subtilis*, *Azotobacter* sp., *Azospirillum* sp., *Pseudomonas* sp.) in a form of a single or mixed inoculum showing significant growth promotion in terms of plant's dry weight, root length and yield<sup>[63-70]</sup>. Accomplished results may differ based on the potting medium used. Earlier study<sup>[64]</sup> shows that sand-peat/manure mixture gave better results in terms of fresh plant weight, when compared to the soil which is original bacterial inhabitant.

### PGPM [Plant Growth-Promoting Microbes] mechanisms of action

PGPMs affect plants by increasing crop yield and plant resistance to stressful environmental conditions and pathogens<sup>[71]</sup>. These bacteria can directly affect the plant by producing substances that can regulate growth and improve the yield. Besides, they can increase water uptake, nutrient uptake and essential elements uptake, all of them having a beneficial effect on the plant<sup>[56, 68, 72, and 73]</sup>. Indirect mechanisms include the inhibition of pathogens through the production of antibiotics and enzymes. Among that, PGPMs increase the availability of micronutrients (uptake of Fe, Zn, and Se) through the processes of solubilization chelating and oxidation/reduction reactions in the soil<sup>[54]</sup>.

The main mechanisms PGPMs use to contribute to the increase of nutrients in the soil are nitrogen fixation and phosphate solubilization, along with solubilization of other minerals. After photosynthesis, nitrogen fixation is the most important biological process in nature, enabling the circulation of nitrogen in the biosphere<sup>[74]</sup>. Symbiotic bacteria from the group *Rhizobium* and *Frankia*, and non-symbiotic bacteria such as *Azospirillum* sp., *Azotobacter* sp. and *Acetobacter* sp. have the ability to assimilate N<sub>2</sub> from the atmosphere and convert it into NH<sub>3</sub><sup>-</sup> as part of a mechanism well-known as nitrogen fixation<sup>[75]</sup>. Nitrogen fixation is controlled through the amount of oxygen and the availability of nitrogen and is carried out with the help of the nitrogenase, enzyme produced by bacteria<sup>[76]</sup>. The transformation of nitrogen takes place through ammonification, nitrification, nitrogen fixation and denitrification<sup>[77]</sup>.

The conversion of insoluble forms of phosphorus into forms that are more available to the plant in the rhizosphere is achieved by means of bacteria called phosphate-solubilizers<sup>[75]</sup>. Some of the PGPMs, including *Pseudomonas* sp., *Bacillus* sp., *Burkholderia* sp., *Rhizobium* sp. and *Flavobacterium* sp., have the ability to solubilize some insoluble phosphate compounds. The usage of these bacteria as a part of bioinoculants may enhance the assimilation of phosphate and offers numerous advantages to the direct stimulation of plant growth<sup>[45]</sup>. In some cases, bacteria from the group

of *Bacillus*, *Pseudomonas*, *Serratia* and *Streptomyces* can take part in the solubilization and mineralization at the same time<sup>[78, 79, and 80]</sup> proved that *Sinorhizobium meliloti*, *Bacillus flexus* and *Bacillus megaterium* have the possibility to solubilize tricalcium phosphate and hydroxyapatite. During that process, some extracellular enzymes (various phosphatases) and important compounds (pH lowering organic acids, siderophores and hydroxyl ions) are released in order to dissolve the minerals. While substrate is being degraded, phosphorus is delivered into the soil<sup>[81, 82, and 83]</sup>.

Knowing the importance of other macro- and micronutrients, it is important to research strains that will enhance their absorption from the soil. It has been recently reported<sup>[70]</sup> that inoculation with Zn-solubilizing bacteria can help to enhance Zn nutrition by plants, therefore improving the growth of plant. In this particular study *Bacillus aryabhatai* and *B. subtilis* were used to inoculate maize, which resulted in better growth of the plant. On the other hand, *Azospirillum brasilense* is proved to be Fe-solubilizing bacteria increasing the Fe and biomass content in cucumber plants<sup>[84]</sup>, which is attributed to production of siderophores.

PGPMs are also able to make phytohormones which stimulate plant growth, thus the mechanism of their activity is known as biostimulation. Some of the most important phytohormones are auxins, cytokinins, gibberellins and abscisic acid<sup>[79]</sup>. Auxins are plant hormones with a cardinal role to modulate the development of a plant. As much as 80% of the PGPMs can synthesize the indole acetic acid (IAA), which has an important role in the stimulation of cellular division and differentiation<sup>[52]</sup>. IAA induces the occurrence of lateral roots among dicotyledons and adventive roots among monocotyledons, improves secondary thickening of the walls and an increase in xylem cells, which results in better minerals and water uptake<sup>[45, 76]</sup>. *Azospirillum* sp., fluorescent *Pseudomonas* sp., and several other PGPMs secrete IAA<sup>[45, 85]</sup>.

Gibberellins take part in cellular elongation and division, as well as the internodium elongation. The mechanisms which improve plant growth through gibberellins are as yet unknown<sup>[76]</sup>. Some authors believe that gibberellins increase the

density of the absorbent hairs on the root that soak up water and nutrients, which contributes to the formation of greater sized fruits, an increased number of buds, prevents the dormant stage of the bulb and stimulates parthenocarpy. The lack of gibberellins is responsible for the occurrence of dwarf plants<sup>[79]</sup>.

Cytokinins stimulate cellular division in some plants and in some cases the development of the root and absorbent hairs on the root<sup>[79]</sup>. In addition, they take part in the growth of plant callus and help the differentiation of the shoots<sup>[76]</sup>. Ninety percent of rhizosphere microorganisms have the ability to produce and release cytokinins, while approximately 30 compounds from the group of cytokinins that promote growth have a microbial origin. Existing data indicates that *Rhizobium* sp. produces cytokinins<sup>[79]</sup>.

Abscisic acid regulates the physiological processes in plant<sup>[76]</sup>. In part, it is synthesized in the chloroplasts, while its entire biosynthesis primarily takes place in the leaves, initiated by the stressful environmental conditions such as a lack of water and low temperatures<sup>[79]</sup>. It helps the germination of the seed, the closing of stomata and tolerance to environmental stress<sup>[76]</sup>.

Various pathogenic bacteria, fungi and nematodes may infect the plant and thus reduce crop yield to a great extent. As previously indicated, PGPMs significantly influence the induction of plant resistance to pathogens by synthesizing various antibiotics, siderophores, cyanides or lytic enzymes<sup>[77]</sup>.

One of the main mechanisms for the control of pathogens is the ability to synthesize one or more antibiotics. Many PGPMs with the ability to synthesize antibiotics also produce cyanide, which in most cases has a synergistic effect when combined with antibiotics<sup>[86]</sup>. Furthermore, with the aim to prevail over the restricted supply of iron in the soil, some PGPMs are able to produce siderophores. Siderophores are low molecular mass organic compounds with strong chelating affinity towards ions of iron ( $Fe^{+3}$ ). In presence of oxygen, most of the iron particles are only partly soluble and thus are not completely available to the living organisms<sup>[79]</sup>. Bacterial siderophores have a positive effect on the growth of plants, functioning as a source of iron that is readily usable to the plant<sup>[45]</sup>. Certain studies have

indicated that *Pseudomonas*, which produces siderophores, influences antifungal activity towards different pathogenic fungi<sup>[87]</sup>, while *Bacillus cereus* has a potential in biocontrol of rice fungi<sup>[88]</sup>. *Pseudomonas* strains have been studied way back in 1984 when it was proved that they inhibit growth of six fungi by virtue of siderophore production<sup>[89]</sup>. *Pseudomonas putida* efficiently controlled tomato foot and root rot caused by *Fusarium oxysporum* in laboratory experiments as well as at industrial level<sup>[90]</sup>.

In addition, PGPMs have a positive effect on the characteristics of the soil itself and their consortiums are successfully used in the processes of bioremediation. This is how nutrient poor and polluted soil becomes arable and available to agricultural production, since a transformation occurs in the hydrocarbons and other pollutants into less detrimental forms<sup>[91]</sup>. Microorganisms that effectively break down hydrocarbons and oil-based pollutants include *Nocardia* sp., *Pseudomonas* sp., *Acinetobacter* sp., *Flavobacterium* sp., *Micrococcus* sp., *Arthrobacter* sp., *Corynebacterium* sp., *Mycobacterium* sp., *Bacillus* sp., etc.<sup>[92]</sup>. AM fungi are also studied in the phytoremediation processes, indicating their role in improving soil conditions and enhancing plant tolerance to heavy metals<sup>[93]</sup>. Some studies have shown that commonly known PGPM (*Bacillus* sp., *Pseudomonas* sp., *Agrobacterium* sp., etc) not only improve the plant growth, but also reduce uptake of heavy metals by plants<sup>[94,95]</sup>. For example, *Microbacterium* sp. successfully prevented chromium toxic effect on pea by simply reducing its bioavailability in soil<sup>[96]</sup>. On the other hand, *Pseudomonas putida* is capable of simultaneously degrading naphthalene in soil, protecting the seed and the plant from possible lethal effect<sup>[97]</sup>.

#### Mass multiplication of microorganisms

Mass multiplication of microorganisms is achieved through the application of a batch, semi-continuous (fed-batch) or continuous cultivation in various growth media (submerge or solid-state). Continuous fermentation is considered an experimental procedure and is rarely used on an industrial scale<sup>[86]</sup>. Batch fermentations seem to be most commonly used, as in that case, it is easy to set up and control the bioreactor. In the case of the application of a semi-continuous fermentation

process, it is possible to perform: fed-batch fermentation with pulse feeding, exponential fed-batch fermentation and linear fed-batch fermentation<sup>[98]</sup>. The addition of nutrients during the semi-continuous processes extends the exponential and stationary phase, thus resulting in higher biomass concentration. Many experiments have proven that semi-continuous fermentation can lead to better results, giving higher yields of desired products<sup>[99, 100, and 101]</sup>. Still, this process is not used widely because it requires much more attention, nonstop control and monitoring, which can lead to higher production costs<sup>[86]</sup>. In order to achieve desired growth, it is necessary to provide required conditions of fermentation, that is, parameters such as medium composition, temperature, pH values, mixing speed and concentration of oxygen<sup>[102]</sup>.

#### ***Future Perspective and Challenges of Microbial-Based Agro-Inputs***

As global warming is becoming a reality endangering nutritional demand, there is a need for innovative agro-inputs that enable agriculture to adapt to worsening environmental situations and exploiting microbial resources is one of the most promising solutions to achieve such aim. Indeed, it is clear today that microbial inoculants, a sub category of the so-called biostimulants, have become one of the attractive agro-inputs for sustainable intensification of agriculture, especially for smallholders<sup>[103]</sup>. Biostimulants have gained substantial ground market wise, owing to the impressive know-how acquired during the last two decades, and most importantly to the involvement of low-cost technologies in their production process. However, despite all the aforementioned conveniences and numerous scientific and field evidences of their agronomic effectiveness, efforts are still required to make them full-fledged commodities that are used as standard by farmers.

There is a growing body of evidences about the large number of microbes that have been found to be highly beneficial for soil fertility and plant productivity in many major cropping systems. At the same time, many reports have demonstrated inconsistent and poorly repeatable results via controlled and field trials<sup>[104, 105]</sup>, which may indicate uncertainty in the efficacy of the microbial inoculants that should be aligned with

intricate biotic and abiotic factors including plant species, native microbial communities, environmental conditions, soil type and soil-related management practices such as fertilization, cropping systems, irrigation, and biocontrol strategies<sup>[106]</sup>. Progress in this area would ultimately depend on a clear understanding of the latter factors in order to guarantee a successful manipulation of agriculture microbes, their commercialization, and widespread use. This is in agreement with the saying “big potential in small packages” by Matt Kleinhenz (Third world congress on the use of biostimulants in agriculture 2017, Miami) who portrayed the current state of the microbial-based biostimulants whose development presumably rely on coping with several issues relatively to both technical and economic aspects. Another concern is arguably related to misconceptions and lacking objectives in terms of research programs development as most research works are driven by “substitution approaches” where microbial inoculants are labeled as direct competitors to well-established agro-inputs with proven efficacy such as fertilizers.

Next generation agriculture should henceforth make use of all available resources and designing novel agro-models that focus on how to achieve perfect alliance between biologicals, chemicals, and biocomputing technologies. In that regard, adopting multidisciplinary approaches in developing microbial-based solutions concurrently with mineral fertilizer resources is paramount as it could lead to creating market's opportunities and new agricultural paradigms based on new concept of sustainability, which is in tune with contemporary's conceptions of today's individuals. In this regards, scientists and manufactures interested in microbial-based biostimulants should focus on delivering stable formulations capable of withstanding harsh storage conditions and guaranteeing extended shelf life of active ingredients through limiting viability loss. Most importantly, microbial formulations must be compatible with conventional agro-equipment and other agro-inputs, especially mineral fertilizers, so their supply chains could be aligned. As a matter of fact, formulation is one of the most critical step in microbial inoculants manufacturing and several

carriers have been used with contrasted results depending on the microbial species and pretreatment methods. Those carriers mostly include organic materials (i.e., peat, lignite, and composts, etc.) and polymeric compounds (i.e., alginate, agar, pectin, and chitosan, etc.;<sup>[107]</sup>). Multi- and inter-disciplinary approaches are worth considering when designing innovative microbial formulations. This will open up new insights into an unexploited research area such as combining new-generation coating and microbial technologies that likely should arouse particular interests to innovative smart fertilizers. For instance, microbial biotechnologies would benefit from other emerging technologies such as those related to EEF and controlled release fertilizers, though not largely used for staple crops and costly to be applied for an intensive agriculture<sup>[108, 109, 110]</sup>. For example, recent advances in coating technology that have led to the development of new-generation fertilizers particularly aiming at improving N use efficiency (reducing leaching, volatilization, and denitrification) may be exploited to enhance P fertilizers efficiency and uptake. This would contribute overcoming common issues related to low P availability which is pH-dependent, readily bounded with divalent cations and belowground leached, thus precise release rate and efficient plant root P uptake may be achieved. That being said, to our knowledge little has been done regarding production of customized carriers able to respond to all required quality criteria. For instance combining new-generation coating and microbial technologies is an unexploited research area that should arouse more interests. Breakthrough in that department could be a true game changer, thus giving rise to innovative smart fertilizers, matching the few concepts that precision agriculture relies on (sensing technology, farming satellite, data analysis, and controlled release fertilizers, etc.) while providing possibilities to enhance specific microbial biological functions related to nutrient dynamic in soils.

Given altogether, developing strategies relying on understanding potential modes of actions that provide possibilities to enhance specific microbial functions related to nutrient dynamic in soils, strengthening scientific and industrial collaborative partnerships, meeting farmers'

requirements are considered paramount in conceiving targeted products and answering specific consumer needs. Fostering proximity to growers should be given a special consideration since farmers' acceptance has to be the utmost priority that can only be achieved through in-field demonstrations, producing reports and data specifically tailored for growers' specificities. In addition, needless to say that the triumph of the next generation of agro-inputs based on microbial inoculants is largely dependent on regulatory clearness and adopting collaborative mindset where progress is made through farmers, scientists from private and public research institutes, advisers and policy makers. This will help moving toward integrated and profitable ecosystems where all inputs are managed following wholesome principles and aiming at optimizing nutrient use efficiency in a context where climate variability is persistently threatening for food productivity.

### Conclusion

Microbial fertilizers have been in a focus of researches for quite a long time. They are considered to be ecologically acceptable alternative to chemical fertilizers and agrochemicals, which are overused and harmful to the environment. Although this idea is not brand new and has been subject of plenty of scientific papers for years now, many questions still remain unanswered and there is a lot of place for improvement. The production of microbial fertilizers does not depend solely on the detailed knowledge of the physiology of plants and microorganisms, but also on the large number of technological challenges such as the fermentation process, type of formulations, the population of microorganisms and their system of release. Thus, the development of a stable bioformulation is possible through combining knowledge from microbial and technical aspects. Additional research is necessary in order to enhance the production process and, what's most important, to improve the products reliability and practical usage.

Environmental stresses are becoming a major problem and productivity is declining at an unprecedented rate. Biofertilizers can help solve the problem of feeding an increasing global population at a time when agriculture is facing

various environmental stresses. It is important to realise the useful aspects of biofertilizers and implement its application to modern agricultural practices. The new technology developed using the powerful tool of molecular biotechnology can enhance the biological pathways of production of phytohormones. If identified and transferred to the useful PGPRs, these technologies can help provide relief from environmental stresses. However, the lack of awareness regarding improved protocols of biofertiliser applications to the field is one of the few reasons why many useful PGPRs are still beyond the knowledge of ecologists and agriculturists. Nevertheless, the recent progresses in technologies related to microbial science, plant-pathogen interactions and genomics will help to optimize the required protocols. The success of the science related to biofertilizers depends on inventions of innovative strategies related to the functions of PGPRs and their proper application to the field of agriculture. The major challenge in this area of research lies in the fact that along with the identification of various strains of PGPRs and its properties it is essential to dissect the actual mechanism of functioning of PGPRs for their efficacy toward exploitation in sustainable agriculture.

Fertilizers play an important role in enhancing crop productivity. However, chemical fertilizers are expensive, non-eco-friendly, cause eutrophication, reduce organic matter and microbiotic activity in soil and are hazardous to health. Therefore, the use of biofertilizers is desirable as they are natural, biodegradable, organic and more cost-effective than chemical fertilizers. Biofertilizers consist of plant remains, organic matter and some special class of micro-organisms. Biofertilizers help to increase quality of the soil by providing nutrients and natural environment in the rhizosphere. The micro-organisms present in biofertilizers are important because they produce nitrogen, potassium, phosphorus and other nutrients required for benefit of the plants. Most biofertilizers also secrete hormones like auxins, cytokinins, biotins and vitamins which are essential for plant growth. Biofertilizers give protection to plant by secreting antibiotics which are effective against many plant pathogens. Biofertilizers also protect plant from salinity and drought stress. Biofertilizers are

inexpensive and safe inputs which provide a wide scope for research in the areas of organic farming and development of stress-free environment. Overall, the significant role of biofertilizers in plant growth productivity and protection against some stresses makes them a vital and powerful tool for organic and sustainable agriculture.

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