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REVIEW PAPER

Biofertilizer – A Key Player in Enhancing Soil Fertility and Agricultural Sustainability

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ABSTRACT

The overuse of chemical fertilisers has devastating effects on agricultural systems and the environment. Thus, environmentally friendly substitutes for chemical fertilisers are needed. The potential contribution of biofertilizer to food security and environmentally sound practises has increased its profile in the agricultural sector. Soil fertility may be improved by the use of biofertilizers, which contain microorganisms like fungus, bacteria, and protozoa that can fix nitrogen, dissolve phosphorus, and sequester iron. Biofertilizers are made up of microorganisms that are both living and dormant and supply nutrients for plant development. Nitrogen-fixing microbes, phosphorus-mobilising and phosphorus-solubilizing microbes, potassium-solubilizing microbes, blue-green algae, and azolla were just some of the biofertilizers discussed in this review, along with their applications in crop production, production processes, and examples of helpful microbes used in biofertilizer industries.

HIGHLIGHTS

- Biofertilizers are eco-friendly contain living microorganisms are nitrogen fixer, phosphate solubilizers and mobilizes, plant growth promoting bacteria, etc.
- Biofertilizers play an image role in agricultural sustainability.

Keywords: Biofertilizer, Sustainable agriculture, Rhizobium, PGPR, Nanotechnology

As the world's population rises, so does the urgency with which it must secure a reliable food supply. Because it is likely that the world's population will reach 8 billion in 2030 and 9 billion in 2050, the amount of food that is produced will need to grow by between 60 and 70 percent (Lee *et al*. 2014; Sosa *et al.* 2017). In industrialised nations where arable land is in short supply, people turn to artificial fertilisers, which boost crop output, to improve the land's potential to produce. Fertilisers are organic or inorganic compounds that, when applied to plants, soil, or by fertigation, can replenish the soil's natural nutrients and improve soil fertility as well as crop growth (Edgerton, 2009).

Crops require macronutrient (nitrogen, calcium, magnesium, phosphorus, etc.) and micronutrient (zinc, iron, boron, etc.) supplements for their growth. The use of synthetic chemical fertilisers for nitrogen and phosphate has been largely responsible for the development of an extremely productive and intense agricultural system (Schultz *et al*. 1995). Nevertheless, there are consequences to consider when utilising chemical fertilisers over an extended period. There is a possibility that eutrophication will occur as a result of the concentration of these substances in the water. These chemicals not only lessen the fertility of the soil, but they also increase the salinity of the soil and worsen nutrient inequality. Modern agriculture's excessive, unbalanced, and constant synthetic input of chemical fertilisers has

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degraded soil quality (by making it biologically inert and extremely salty), surface and ground water, biodiversity, and ecosystem functioning (Socolow, 1999). Because of this, biofertilizers might be considered a target of sorts. Without microbes, the potential for the existence of higher species does not exist. As evidenced by several studies (Kuhad, 2012; Kumar *et al*. 2021; Kumar *et al*. 2017), the claim that all life originates from bacteria and other microorganisms is true. Microbes can be thought of in all conceivable places, beyond the range of the imagination (Khati, 2018). Microbes are omnipresent and versatile, making them vital to the earth. They frequently assist and control biogeochemical cycles. Microbes play an important role on the committee of stewards of the planet's health and property (Timmy *et al*. 2017).

To increase the supply or availability of primary nutrients to the intended crops, biofertilizers contain living microorganisms that colonise the rhizosphere or the interior of the plants and promote growth (Mazid *et al*. 2011). When applied to soil, seeds, or plant surfaces, biofertilizers can be defined as "substances that contain living microorganisms that colonise the rhizosphere or the interior of the plants to promote growth by increasing the supply or availability of primary nutrients to the target crops (Raja, 2013). Traditions in soil and crop management, such as crop rotation, organic adjustments, tillage maintenance, recycling of crop residue, soil fertility renovation, and the biocontrol of pathogens and insect pests, can be greatly aided by the use of biofertilizers (Sahoo *et al*. 2013). They are ready-to-use, live formulations of beneficial microorganisms that can increase soil and/or crop productivity by fixing nitrogen (N), solubilizing phosphorus (P), and mobilising nutrients through their biological metabolism when applied to seed, roots, or soil treatment (Mahanty *et al*. 2016). Only 10–40% of applied nutrients are used by the plant; the other 60–90% are lost through immobilisation, leaching, volatilization, and other mechanisms. Biofertilizers include organisms that fix atmospheric nitrogen in the soil and root nodules of legume crops, so making the nitrogen accessible to the plants. They convert the phosphates, tricalcium, and other insoluble forms into soluble forms. These biofertilizers can be applied to soil or seeds to boost the availability of nutrients and increase production by 10% to 20% without harming the soil or the environment. In addition to the aforementioned information, long-term usage of biofertilizers is more affordable, environmentally benign, productive, and accessible to marginal and small farmers than chemical fertilisers (Ghumare *et al*. 2014).

Nitrogen fixation, phosphorus solubilization, phosphorus mobilisation, potassium solubilization, micronutrient solubilization, plant growth promotion, and preventing the depletion of soil organic matter (Jeyabal and Kupuswamy, 2001) are just some of the mechanisms by which biofertilizers increase yields and improve plant health (Figure1). Biofertilizers provide renewable plant nutrients for agricultural ecosystems. Biofertilizers provide live latent bacteria from the plant's rhizosphere to seed, nursery soil, or the plant canopy. Biological activities, including solubilizing inaccessible forms of phosphorus, mobilising phosphorus, and solubilizing other crop nutrients, boost agriculture productivity (Kour *et al*. 2020; Thomas and Singh, 2019). Like other synthetic chemicals, bio-fertilisers contain beneficial bacteria, growth agents, and additives that may promote microbial growth. Bio-fertiliser increases food content, protein, amino acids, vitamins, nitrogen, and other necessary elements by 10%–40% (Bhardwaj *et al*. 2014). The inhibition of pathogens and other unwanted microbes through parasitism, the competition for resources and habitats within the rhizosphere, the production of antagonistic chemicals (such as antibiotics, hydrogen cyanide, and siderophores), and enzymes all have indirect impacts on the development of plants (Basu *et al*. 2021).

Plant growth-promoting microorganisms are bacteria that thrive in the rhizosphere, which is the region around the roots. As a result of their presence in the rhizosphere, these bacteria are able to stimulate the growth of plants. These kinds of bacteria are referred to as plant growth-promoting bacteria (PGPB). There are several PGPBs that are now being sold commercially as biofertilizers (Calvo *et al*. 2014). Plant growth promoting bacteria (PGPBs) provide critical components such as nitrogen via nitrogen fixation, phosphorous by the procedure of phosphorous solubilization, potassium intake, and hormones like auxins, cytokinins, and gibberellins, all of which

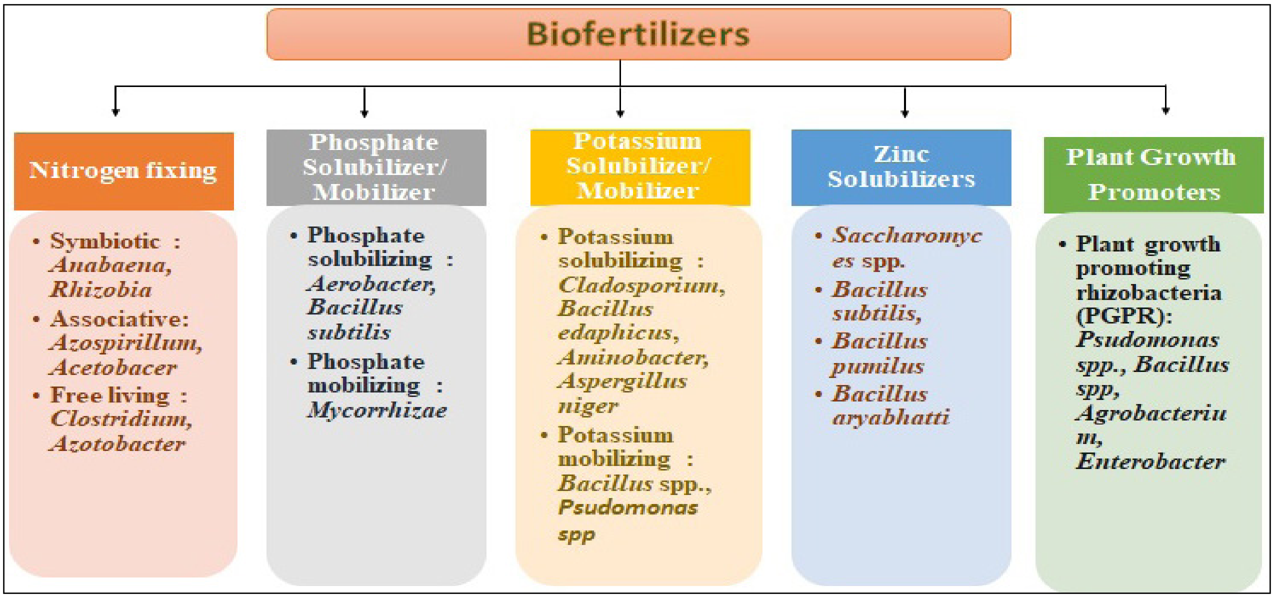

Fig. 1: Types of Biofertilizer with living microorganisms

have a direct role in plant growth. *Azotobacter*, *Azospirillum*, *Rhizobium*, *Cyanobacteria*, phosphate and potassium solubilizing microorganisms, and mycorrhizae are PGPRs that flourish in no-tillage or minimum-tillage soil. Crop yields are primarily constrained by biotic and abiotic stresses. PGPRs serve a significant function as bioprotectants in the context of modern scientific methodologies that have been widely employed to benefit crops under stress. Biofertilizers help directly fix atmospheric nitrogen and are inexpensive and environmentally beneficial. They boost root development, and the use of PGPR aids in the prevention of plant diseases. Several microorganisms, such as nitrogen-fixing soil bacteria (like *Azotobacter* and *Rhizobium*), nitrogen-fixing cyanobacteria (such as *Anabaena*), solubilizing phosphate bacteria (e.g., *Pseudomonas*), and arbuscular mycorrhicical fungi, are frequently utilised as biofertilizers (Table 1).

TYPES OF BIOFERTILIZERS

Nitrogen fixing Microbes

The inert nitrogen that makes up about 78 percent of the air is useless to plants. Ammonium and nitrate are two forms of nitrogen that plants may take up. However, the lighting industry does create some ammonia, and industrial ammonia can be made at high pressures and temperatures using the Haber-Bosch process and a catalyst based on iron. In the process of biological nitrogen fixation (BNF), microorganisms employ nitrogenase enzymes to transform atmospheric nitrogen into ammonia, the usable form of N. Nitrogen is "fixed" into proteins via a transamination pathway after being converted to ammonia by the nitrogenase enzyme (Islam *et al*. 2016; Laguerre *et al*. 2007; Raklami *et al*. 2019). Free-living bacteria, such as *Azotobacter*, associative bacteria, such as *Azospirillium*, cyanobacteria, and symbiotic species, such as *Rhizobium*, *Frankia*, and *Azolla*, are some examples of nitrogen-fixing bacteria. Different sub group of species are available for nitrogen fixations are mentioned below:

(a) Azotobacter

They are free-living bacteria that are photoautotrophic, aerobic, and non-symbiotic. They belong to the family *Azotobacteriaceae* and play a very essential role in the nitrogen cycle of plants. *Azotobacter* is a rod-shaped, Gram-negative aerobic organism with cysts with thick walls and no endospores present. They may be found in natural as well as alkaline soil; however, they are found in arable soils more frequently. These cysts resist desiccation and harmful chemical and physical agents. During the stage in which they are found as cysts, they do not fix nitrogen and exhibit optical refraction. Its mobility depends on whether or not it has peritrichous flagella. It may produce water-soluble pigments that have a bright yellow-green, red-violet, or brownish-black colour. It thrives between temperature 20–30°C and pH 7.0–7.5. *Azotobacter*

Table 1: Biofertilizers and their mechanisms

Biofertilizer	Microorganism(s)	Plant growth promoting activity	Effect on plant productivity parameters	References
Nitrogen- fixing bacteria	***Azotobacter*** sp. strain *Avi2* (MCC 3432)	Nitrogen fixation, production of IAA, siderophore	Improved vegetative and reproductive growth in rice	(Banik and Dangar, 2019)
	Azotobacter chroococcum, A. vinelandii	Nitrogen fixation, P solubilization, production of NH_3, HCN, IAA	Increased shoot and root length, leaf and root number, chlorophyll content of maize	(Jain *et al*. 2021)
	Rhizobium meliloti	Nitrogen fixation, production of siderophore and chitinase	Increased growth and yield, pods quality and better use of nitrogen in peanut	(Mondal *et al*. 2020)
	Enterobacter cloacae (PGLO9)	Nitrogen fixation, phosphorous solubilization, siderophore production, ACC deaminase	Enhanced potato growth and yield, significant increase in root and shoot length, root as well as shoot biomass	(Verma *et al*. 2018)
	Sinorhizohium sp.	Nitrogen fixation, production of IAA	Increased seed yield in mug bean	(Verma, 2011)
Phosphate-solubilizing bacteria (PSB)	*R. leguminosarum* ***Pseudomonas moraviensis, Bacillus halotolerans,***	P solubilisation P solubilization, production of IAA,	Improved growth and yield, and better resistance against drought in soybean improved wheat seed germination,	(Igiehon *et al*. 2019)
	Bacillus subtilis LK14	P solubilization, production of IAA	Enhanced host plant's nutrient uptake and amelioration of stress	(Khan *et al*. 2016)
	R. leguminosarum	Phosphate solubilization	Better resistance and improved growth	(Fahsi *et al*. 2021)
Potassium-solubilizing bacteria (KSB)	***Bacillus mucilognosus***	Potassium solubilization	Increased the plant biomass, yield and uptake of K in Sudan grass	(Basak and Biswas, 2008)
	Acidothiobacillus ferooxidans	Potassium solubilization	Increased growth and yield	(Ansari *et al*. 2017)
	Bacillus edaphicus strain NBT	Potassium solubilization	Increased root and shoot Growth, and K content in cotton and rape	(Sheng, 2005)
	Pseudomonas acidothiobacillus	Potassium solubilization	Enhance crop yield	(Parmar and Sindhu, 2019)
	Bacillus mergaterium	Potassium solubilization	Promoted soyabean seed germination seedling growth	(Bakhshandeh *et al*. 2020)
Zinc solubilizing bacteria	***Bacillus aryabhatti***	Zinc solubilization	Plant growth is increased in maize	(Mumtaz *et al*. 2017)
	Pseudomonas pseudoalcaligenes, Bacillus pumilus	Zinc solubilization	Boosted rice plant height and dry weight	(Jha, 2019)

Plant Growth Promoting Rhizobacteria	***Arthrobacter, Agrobacterium, Rhizobium***	Promoting plant growth	Hormones are produced that promotes development of root, increase availability of nutrient	(Meena *et al.* 2014)
	Bacillus megatertum	Promoting plant growth	Increased potential of roots to absorb water in salinity condition	(Olivera *et al.* 2009)
Co-inoculation of important bacteria	*Enterobacter* species, *Psudomonas aeruginosa*	Production of IAA, siderophore, Nitrogen fixation, Phosphate solubilization	Increased yield, Straw yield	(Latef *et al*. 2020)

nigricans, *Azotobacter chroococcum*, *Azotobacter vinelandii*, *Azotobacter beijerinckii*, and *Azotobacter paspali* are a few examples. The other members are mostly soil-borne and rhizospheric, apart from a few rhizoplane bacteria. *Azotobacter chroococcum* and *Azotobacter paspali* are widely known for their ability to function as biofertilizers for a number of non-legume crops. (Thomas and Singh, 2019; Noar *et al.* 2018). Gibberellins and naphthalene acetic acid, hormones produced by these bacteria, reduce root infections and promote root development by helping plants absorb minerals and nutrients. These bacteria produce B-complex vitamins. *Azotobactor* bacteria produce thiamine, riboflavin, and other vitamins in rice, maize, sugarcane, and vegetables (Wani *et al.* 2013). Some *Azotobacter* strains have been identified as a possible biocontrol agent and have been shown to excrete bioactive chemicals, including phytohormones that increase mineral intake by boosting root development (Mahanty *et al.* 2016; Noar *et al.* 2018). In the presence of a lack of iron, *Azotobacter vinelendii* produces the siderophore azotobactin (Noar *et al.* 2018).

(b) Rhizobium

Rhizobia are soil bacteria that colonise the roots of legumes and symbiotically fix atmospheric nitrogen. Rhizobia in their bacteroid form in nodules will look and behave differently than those in free-living settings. In terms of the amount of nitrogen that can be fixed, they are the most effective form of biological fertiliser that can be used on legumes (Jehangir *et al.* 2017). Bacteria belonging to the phylum *Rhizobacea* possess the ability to fix nitrogen in the environment and include the genera *Bradyrhizobium*, *Sinorhizobium*, *Azorhizobium*, and *Mesorhizobium*, in addition to the genus *Rhizobium*. Mutualists are organisms that live in symbiotic relationships with the roots of legume plants, whereas diazotrophs are free-living organisms that fix nitrogen in plants that are not legumes (Verma, 2011). The nod, nif, and fix genes are responsible for regulating the bacterium's ability to nodulate and fix nitrogen. Complex enzymes like Nitrogenase, which contains Di Nitrogenase Reductase, which requires iron (Fe) and molybdenum (Mo) as cofactors, are needed for this process (Hoffman *et al*. 2014). It uses electrons to turn N_2 into NH_3, and oxygen is also necessary for *Rhizobium* species (Santi *et al*. 2013). Depending on changes in the cofactor of dinitrogenase, there are three main types of nitrogenase complexes. Mo, V, and Fe nitrogenases are all types of nitrogenases (Yang *et al*. 2011). Some genes, referred to as "Nif" genes, are involved in fixing nitrogen dioxide in free-living and nitrogen-fixing organisms (Black *et al*. 2012). The Nif genes have a role in the production of cofactors, activation of the Fe/Mo cofactor, regulation of enzyme activity, and electron donation (Ahemad, 2015).

(c) Azospirillum

The genus *Azospirillum* belongs to the family *Spirillaceae* and consists of gram-negative, non-nodule-forming, aerobic, nitrogen-fixing bacteria. *Azospirillum* makes granules of poly-hydroxybutyrate. High motility, large polar flagellums, and peritrichous flagella all contribute to the swarming behaviour of the *Azospirillum* genus. *Azospirillum* grows best in anaerobic conditions (NO_3 as the electron acceptor, denitrification) but can also thrive in microaerophilic (N_2 or NH_3 as nitrogen sources) and fully aerobic (NH_3, NO_3, amino acids) environments. *Azospirillum* creates unique, scarlet-red, wrinkled colonies on Rojo-Congo red substrate (Mosa *et al*. 2016; Alexandre, 2017; Lin *et al*. 2016). *Azospirillum* is used alone as a seed treatment before planting. It produces phytohormones and siderophores, solubilizes phosphate (Puente *et al*. 2004), has biocontrol action (Bashan and De-Bashan, 2010), and protects plants from soil salinity and harmful chemicals (Creus *et al*. 1997). Species of Azospirillum that can help in nitrogen fixation are *Azospirillum lipoferum, Azospirillum brasilense, Azospirillum halopraeferens, and Azospirillum amazonense* (Ayyaz *et al*. 2016). Wheat seeds treated with lectins from two different strains of *Azospirillum brasilense*, Sp7 (epiphyte) and Sp245 (endophyte), were tested by Alen'kina and Nikitina (2021) in order to determine how abiotic stress influenced the growth and germination characteristics of the host wheat plant.

(d) Blue Green Algae (BGA)

Cyanobacteria are a promising new class of microorganisms for greener farming in the long run (Singh *et al*. 2016). Diazotrophes are a kind of cyanobacteria that may be used to produce low-cost and environmentally acceptable biofertilizers. In addition to preventing plant nitrogen deficits, they also increase soil aeration, water retention, and vitamin B_{12} content (Hall *et al*. 1995). There are many different species of free-living cyanobacteria or blue-green algae that have the potential to fix nitrogen, some of which are *Anabaena*, *Nostoc*, *Aulosira*, *Totypothrix*, *Cylindrospermum*, and *Stigonema*. *Cyanobacteria* are self-sustaining, meaning they can develop, expand, and produce their own organic byproducts without the help of host organisms. The relationship between *Azolla* and *Anabaena* exemplifies symbiosis for nitrogen fixation and nutrient enrichment in the rice paddy area.

They show that a high level of sporulation in the organism is attributable to the breakdown of lignin in the cell wall and the subsequent release of phenolic compounds (Malliga *et al*. 1996). Blue-green algae produce sticky chemicals, make soil more porous, and excrete vitamins, amino acids, and phytohormones such as auxin and gibberellins (Roger and Reynaud, 1982; Rodríguez, 2006). They decomposed, increased soil biomass, and also lowered soil salinity (Rodríguez, 2006). BGA suppresses undesirable vegetation and efficiently absorbs metals, performing bioremediation (Ibraheem, 2007). All heterocystous forms of BGA are capable of biological nitrogen fixation, and they all have a special structure known as a heterocyst that allows them to do so. However, some BGA can fix ambient nitrogen without heterocysts due to newly discovered specific circumstances in low oxygen settings. The BGA provided 5 to 20 tonnes of fresh-weight cyanobacteria and fixed 25 to 40 kg of biological nitrogen per acre per season in paddy fields. This is because there is a lot of readily available organic matter, which is very beneficial to succeeding crops (Dhar *et al*. 2007; Prasanna *et al*. 2003). Barley, oats, tomato, radish, cotton, sugarcane, maize, chilli peppers, and lettuce are only some of the crops that have been grown with the help of these biofertilizers (Thajuddin and Subramanian, 2005).

Phosphate solubilizing/ mobilizing microbes

According to Bamagoos *et al*. (2021), phosphorous is an essential macronutrient that is necessary for the expansion and maturation of a plant. Although soils contain a lot of phosphorus, plants can only use a tiny quantity of it. Soil phosphorus concentrations can vary widely, from around 400 mg to about 1200 mg per kg. However, there is a relatively modest amount of orthophosphate (soluble or inorganic bioavailable phosphorus), which reduces agricultural yields due to low phosphorus availability in the soil (Miller *et al*. 2010; Wang *et al*. 2017). Phosphorus that is unavailable to plants can be solubilized by microorganisms in the soil and made available to them. They are known as phosphate-solubilizing microorganisms (PSM). Microorganisms that are capable of solubilizing phosphate, such as bacteria, fungi, and actinomycetes, are essential to the process of transforming insoluble inorganic phosphate into more straightforward and water-soluble forms. Chen *et al*. (2006) found that the P-solubilizing microbial population in soil is made up of bacteria (PSB) that form 1 to 50% of the population, whereas fungi (PSF) only make up 0.1 to 0.5% of the population's P solubilisation capability.

Phosphate-solubilizing bacteria are responsible for the processes of solubilisation and mineralization that occur in soil. These processes include the conversion of the organic form of phosphate into its inorganic form (Tandon *et al*. 2020). In order to break down the organic phosphate stores, PSB secretes organic acids, including citric acid and gluconic acid. PSB also secretes the enzymes phytases and nucleases, which are essential for the mineralization of organic phosphate reserves (Ku *et al*. 2018). Many bacterial species like *Pseudomonas*, *Achromobacter*, *Bacillus*, *Brevibacterium*, *Erwinia* sp., *Corynebacterium*, *Xanthomonas* sp., *Nostoc*, *Rhodococcus* sp., *Burkholderia*, *Flavobacterium* sp., *Micrococcus* sp., *Sarcina* sp., and *Scytonema* have been reported to solubilize phosphorous in soil (Oteino *et al*. 2015; Santoyo *et al*. 2021).

As noted by Khan *et al*. (2014), a fungus that colonises higher plant roots is responsible for transporting phosphorus from the soil into the plant's system. *Mycorrhizae* are fungal interactions with plant roots that benefit both parties. *Mycorrhizae* occur naturally in most soil types, particularly mining wastes, agricultural soils, and field crop soils. 95% of plants have mycorrhizal connections. In exchange for carbon, fungi return nutrients to the soil, improving the capacity of the host to absorb nutrients. *Endomycorrhiza*, *ectomycorrhiza*, and *ectoendomycorrhiza* are morphologically different *mycorrhizae* based on fungal penetration of root cells. The most efficient biological fertilisers are the three types of endomycorrhizal fungus (Berruti *et al*. 2016; Hodge and Storer, 2015). Fungi belonging to the *Basidiomycetes*, *Ascomycetes*, and *Zygomycetes* phyla all take part in endomycorrhizal associations of varying complexity. *Mycorrhizae* mobilise phosphorus, cycle nutrients, and boost microbial biomass. Indigenous arbuscular mycorrhizae (AM) in soil colonise plant roots and promote growth by increasing phosphorous availability. *Mycorrhizal* fungi boost soil phosphorus levels by either solubilizing inorganic phosphorus sources or mineralizing organic phosphorus. In contrast to AM fungi or non-*mycorrhizal* roots, *mycorrhizal* fungi create ectoenzymes that enable host plants to acquire organic nitrogen and phosphorus (Bargaz *et al*. 2018; Kumar *et al*. 2018; Hodge and Storer, 2015; Ritika and Utpal, 2014; Wahbi *et al*. 2016). Many different genera, including *Scutellospora*, *Glomus*, *Acaulospora*, and *Gigaspora*, are used as biofertilizers. *Pseudomonas*, *Bacillus*, *Enterobacter*, *Azospirillum*, *and Rhizobium* are examples of *rhizobacteria*, which can also solubilize phosphorus due to the fact that they colonise the roots of plants and stimulate plant growth. (Adesemoye and Kloepper, 2009).

Potassium solubilizing/ mobilizing microbes

Potassium is the third-most important plant nutrient after nitrogen and phosphorus. Potassium is abundant in soil, but only 1–2% is accessible to plants. Therefore, crop plants require a continual potassium replenishment system in the soil solution. Bacteria, fungi, and actinomycetes solubilize soil potassium by means of producing inorganic and organic acids and by chemical processes like acidolysis, polysaccharides, complexolysis, chelation, etc. (Meena *et al*. 2015). Potassium can influence significant physiological processes such as the generation of starch, the development of roots, and the movement of stomata, resulting in small-sized seeds and plants that are more susceptible to disease, which will lead to a drop in crop yield (Troufflard *et al*. 2010). Some examples of potassium solubilizer microbes (KSMs) are *Pseudomonas, Acidothiobacillus*, *Paenibacillus* sp., *Aminobacter*, *Sphingomonas*, *Aminobacter*, *Bacillus circulans*, *Burkholderia*, *Enterobacter hormaechei, Acidothiobacillus ferrooxidans,* etc. (Parmar and Sindhu, 2019), and their application can enhance crop yield (Sindhu *et al*. 2016). *Bacillus cereus* (B1), *Bacillus megaterium* (B2), *Trichoderma longibrachiatum* (F1), and *Trichoderma simmonsii* (F2), individually as well as in combination, promoted soybean seed germination and seedling growth in laboratory as well as in pot experiments (Bakhshandeh *et al*. 2020).

Zinc Solubilizers

A very prevalent micronutrient deficit is zinc deficiency (Hafeez *et al*. 2013). Chlorophyll, enzymes, proteins, and metabolic processes all require zinc in order to be prepared functionally (Ali *et al*. 2008). Chlorosis, reduced membrane integrity and leaf size, slowed shoot growth, decreased grain production, impaired pollen formation, stunted root development, impaired water uptake and transport, and increased susceptibility to heat, light, and fungal diseases are all indications of zinc deficiency in plants. Zinc—the most important element—makes up 0.008% of the crust. However, more than 50% of Indian soils are zinc deficient, requiring 1.5 ppm of bioavailable zinc (Katyal and Rattan, 1993). External application of soluble zinc sulphate ($ZnSO_4$) assists plants in overcoming the challenges they face while attempting to absorb zinc from the soil. The crop only consumes 1–4% of the total zinc available, and 75% of the applied zinc is transformed into mineral components (Zn-fixation) that plants cannot assimilate; hence, externally applied formulations are not fully used. Microorganisms like *Pseudomonas* sp., *Rhizobium* spp., *Bacillus aryabhattai*, *Thiobacillus thioxidans*, and *Azospirillum* sp. are particularly notable for their ability to solubilize zinc. In soils with higher levels of native zinc or in combination with insoluble zinc compounds like zinc oxide (ZnO), zinc carbonate ($ZnCO_3$), and zinc sulphide (ZnS), the results show that *Bacillus* sp. having Zn solubilizing properties can be used as a biofertilizer for zinc. To wit: Mahdi *et al*. (2010) A growth-enhancing effect was seen in maize after inoculation with the zinc-solubilizing bacterium *Bacillus aryabhattai* (Mumtaz *et al*. 2017). Jha (2019) found that zinc-solubilizing *Pseudomonas pseudoalcaligenes* and *Bacillus pumilus* boosted rice plant height and dry weight.

Plant Growth Promoting Rhizobacteria (PGPRs)

Plant growth-promoting rhizobacteria (PGPR) colonise roots or rhizosphere soil and benefit crops. PGPRs as biological agents are one of the alternatives to chemical agents that have been demonstrated to impart resistance against a variety of disease occurrences (Murphy *et al.* 2000). Microorganisms existing in the rhizosphere can stimulate plant development in two ways: directly and indirectly. Direct and indirect processes of PGPR that contribute to increased plant development include nitrogen fixation, phosphate solubilization, exopolysaccharide synthesis, phytohormone production, hydrogen cyanide creation, lytic enzyme production, antibiotic production, induced system resistance, and siderophore production. In addition, it has been demonstrated that IAA, gibberellins (GA), and cytokinin production and excretion can increase root surface area, which in turn enables plants to more efficiently absorb nutrients from the soil and has also been found to affect seed germination, stem elongation, root hair creation, flowering, fruit setting, and other developmental processes. Rhizobacteria can either cause resistance by means of a salicylic acid-dependent process known as SAR or even perceive plant responses to jasmonic acid and ethylene. *Rhizobacteria* that can induce resistance and function as antagonists (e.g., *Pseudomonas* sp. and *Bacillus* sp.) might be used to develop novel inoculants with synergistic effects, allowing for more effective biocontrol in agricultural settings (Olivera *et al*. 2009).

In addition to *Pseudomonas* and *Bacillus*, other bacterial species like *Enterobacter*, *Klebsiella*, *Azobacter*, *Variovorax Azosprillum*, and *Serratia* have been researched and commercialised for their PGPR, but only a small percentage of them have been used in agricultural practises across the world (Glick *et al*. 2012). Certain symbiotic bacteria can get inside host plant cells and create mutually beneficial relationships and specialised structures. Rhizobium is a mutualistic symbiotic bacterium that fixes atmospheric nitrogen for leguminous agricultural plants in nodules. *Bacillus spp*. and *Pseudomonas spp*. are two examples of PGPR that are capable of producing endospores and hence survive in a broad range of environments. This makes it easier to effectively formulate biofertilizer using these organisms. Higher biomass yields have been observed following inoculation with *Bacillus edaphicus* (in wheat), *Paenibacillus glucanolyticus* (in black pepper), or *Bacillus mucilaginosus* (in eggplant, pepper, and cucumber) in combination with the phosphate-solubilizing *Bacillus megaterium* (Meena *et al.* 2014; Etesami *et al*. 2017). Soil-dwelling microorganisms can be used as biofertilizers to supply minerals like iron and copper, in addition to nitrogen, phosphate, and potassium. Bioremediation methods employing PGPRs for pesticide contamination have attracted attention because of their eco-friendliness, cost-effectiveness, and possible environmental elimination (Nawaz *et al*. 2011). Sharma *et al*. (2017) have also reported degradation of the pesticide chlorpyrifos by *Pseudomonas* bacterial strains that were isolated from the rhizosphere of apple plants. PGPR has been studied for its importance in agriculture, forestry, horticulture, and environmental protection.

MASS PRODUCTION OF BIOFERTILIZERS

A wide variety of microorganisms falling into the categories of PGPB, PGPR, or AM can be incorporated singly or as a mixture into the biofertilizer formulation for a wide array of soil conditions. Though various bioinoculants available on the market are not sufficiently beneficial due

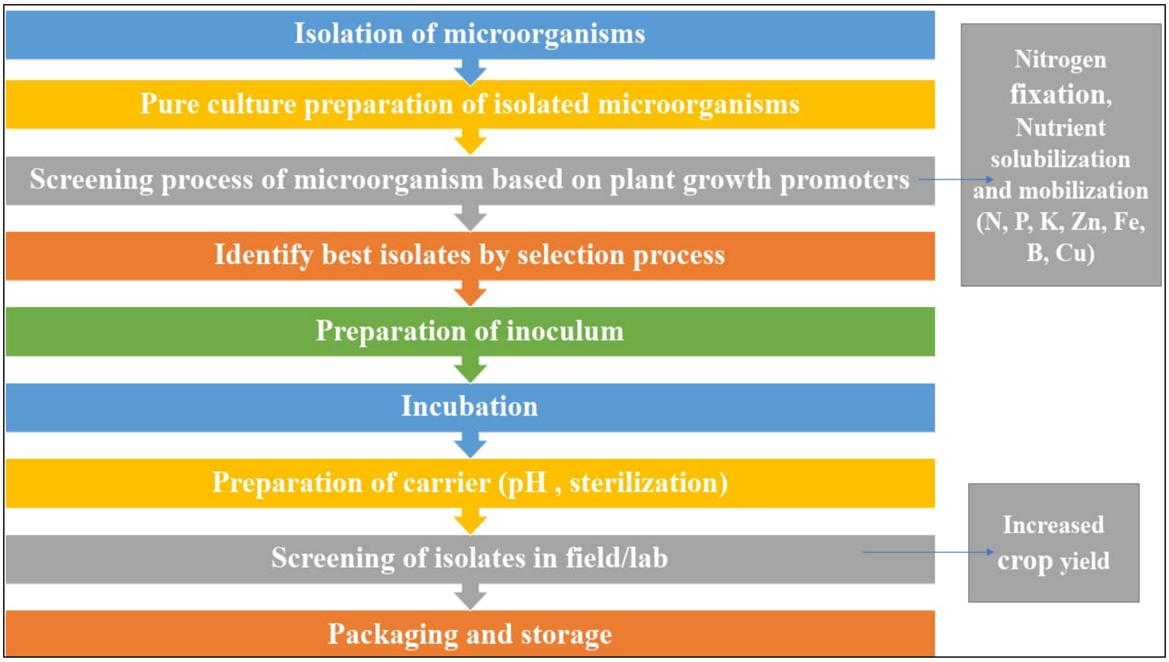

Fig. 2: Common steps in mass production of bio fertilizer

to contamination, poor quality, or being unable to produce the same stimulatory effect in different field conditions, Hence, the formulation of the bioinoculant is the most critical phase and should be prepared in such a way that the microorganism can survive from storage to application (Amenaghawon *et al*. 2021). The formulation of bioinoculant comprises various steps like strain selection, carrier selection, seed pelleting, packaging, and storage (Fig. 2). For strain selection, the isolated microorganisms screen for plant growth-promoting traits and select strains containing good plant growth-promoting (PGP) traits. The formulation of bioinoculant can be either in liquid or solid form. Solid formulations contain carriers, additives, and protectants. The carriers should be selected on the basis of their properties, like high water holding capacity, being non-toxic, simple to process, chemically stable, less expensive, readily available, and being able to carry a large number of bacteria and withstand their survival for an extended period of time. Vermiculite, charcoal, press mud, biogas sludge, farmyard manure, peat, and soil mixture are some of the common carrier materials used in the production of high-quality biofertilizers (Zhou *et al*. 2017).

Though these might come with disadvantages such as a shorter shelf life, susceptibility to temperature fluctuations, vulnerability to contamination, and decreased efficiency because of low cell counts, hence, liquid formulations are created for a variety of microorganisms like *Azotobacter, Rhizobium, and Azospirillum*. For the preparation of liquid inoculants, some protective agents like synthetic polymers (polivinyl alcohol), natural polymers (alginate, gelatin), glycerol, etc. are added to the microbial culture for long-term survival by limiting heat transfer and maintaining water activity (Bernabeu *et al*. 2018). Despite being more expensive, liquid formulations offer benefits including simpler manufacturing, higher cell counts, a longer shelf life, no contaminating factors, storage up to 45°C, and increased soil competency (Ngampimol and Kunathigan, 2008). After the preparation of the bioinoculant, storage for a longer period is the most important step. Different agents like glycerol, oils from horticulture, lactose, pero-dexin, etc. are used as protecting agents during storage, which impart osmotic protection to cells. High water activity protects the cells from desiccation, maintains nutrients, and, with a high water binding capacity, protects the cell during cold storage by preventing the formation of ice crystals (Zárate *et al*. 2005).

FUTURISTIC BIO FERTILIZERS FOR SUSTAINABLE AGRICULTURE

For the purpose of achieving advantages that are both immediate and long-term, it is critical to conduct research on biofertilizers and incorporate the programmes' findings into agricultural practises. Nature's greatest blessing on agriculture is biofertilizers, which may be used in place of potentially hazardous pesticides, weedicides, and commercial chemical fertilisers without reducing harvest success and yet contributing to sustainable agriculture (Kumar *et al*. 2022). There is a growing trend towards chemical-free products as consumers try to alleviate their concerns about pesticide residues in food, global warming, and animal welfare. By 2025, the biofertilizer market is projected to expand from its present valuation of $2.3 billion, an increase of 11.6% (Kumar *et al*. 2022). The most significant obstacles to the widespread use of biofertilization in India are a lack of a committed policy to efficiently use biofertilizers, poor inoculant quality, an absence of expertise in inoculation technology among extension workers and farmers, and an effective system for delivering and supplying inoculants (Bodake *et al*. 2009; Jangid *et al*. 2012).

Biofertilizers benefit soil and sustainable agriculture. Long-term usage of biofertilizers increases soil fertility significantly while remaining economically and environmentally beneficial (Stewart and Roberts, 2012). Seed treatment is the most common, efficient, and cost-effective approach for all inoculants (Sethi *et al*. 2014). For best results, treat seeds with two or more microorganisms. *Rhizobium*, *Azotobacter*, and *Azospirillum*, nitrogen-fixing bacteria can be administered alongside phosphorus-solubilizing microorganisms (Chen, 2006). For agricultural sustainability to continue in the face of climate change and other abiotic pressures, it is crucial to have a better knowledge of rhizosphere biology (Duby *et al*. 2016). Even though biofertilizers are becoming increasingly widespread, the technology is still in its infant phases of application. Currently available biofertilizers tend to focus on boosting the soil's levels of macronutrients like nitrogen, potassium, and phosphorus, while there are also zinc-specific options on the market. In order to provide comprehensive crop and soil health management, ongoing research can aid in the development of biofertilizers with multifunctional nutrient profiles and enhanced nutrient density. Research on eco-friendly carrier materials that can encapsulate active cultures for targeted and sustained action will help biofertilizers improve their effectiveness and commercial acceptance while conquering their shorter shelf life and susceptibility to temperature compared to liquid-based ones. Research and development can lower liquid fertiliser prices, giving farmers more options.

Biofertilizers provide a realistic alternative and potential solution to the myriad issues afflicting the current production system because of their minimal environmental impact and potential beneficial impacts on agricultural sustainability. Increasing the use of nanotechnology in agriculture has the potential to reduce environmental impacts and satisfy rising global food demands. Nanotechnology involves microorganisms beneficial to plant growth and hence requires extensive research and development. The use of nanofertilizers in farming is on the rise. Nanofertilizers are non-toxic, efficient in using nutrients, and can lower production costs. The use of nanoparticles in agriculture aims to reduce the amount of chemicals used to protect plants, reduce the amount of nutrients lost when fertilising, increase crop output, and adapt to changes in the climate. Utilising target-specific nanoparticles is one way to facilitate environmentally responsible breeding, which involves minimising the quantity of chemicals that are discharged into the environment and shielding plant tissue that is not the intended target. These are two of the most important components of environmentally responsible breeding (Sahin *et al*. 2023). Encapsulating nanobiofertilizers with a conjugation of gold, aluminium, and silver nanoparticles will slow the release of PGPR into the target cell (Gupta *et al*. 2021).

CONCLUSION

In integrated nutrient management, biofertilizers can replace chemical fertilisers in sustainable agriculture. Biofertilizers, which are inexpensive and gentle on ecosystems, are crucial to organic farming, ecosystem preservation, and environmental protection. To improve plant growth and soil health, we must understand how microorganisms and crops interact directly. As a consequence of the evolving agricultural environment, which puts an emphasis on sustainability as well as on managing soil and crops in a holistic manner, biological fertilisers now have a lot more opportunity to flourish. This is an exciting development. Field testing is necessary, in addition to on-going research and development, in order to ensure that agricultural practises are effective in their application.

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