**Bioinoculants: An Ecofriendly Approach towards Artificial Fertilizers in Sustainable Agriculture"**

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**Abstract:**

 Bioinoculants, also known as microbial biostimulants, are gaining widespread attention in modern agriculture due to their potential to enhance crop productivity and improve soil health while reducing the reliance on synthetic chemicals. The different types of bioinoculants, includes nitrogen-fixing bacteria, phosphate-solubilizing microorganisms, plant growth-promoting rhizobacteria (PGPR), mycorrhizal fungi, and other beneficial microbes. The role of bioinoculants in fostering plant-microbe interactions, particularly within the rhizosphere, shows how these interactions enhance nutrient uptake, promote root system development, and contribute to the overall health and resilience of crops. Moreover, the potential synergistic effects of combining multiple bioinoculant strains to optimize their benefits are explored.

**Keywords**: bioinoculant, plant growth promoting bacteria, rhizobium

**Introduction:**

Bacteria that colonise the rhizosphere and plant roots and enhance plant growth by any mechanism are referred to as plant growth-promoting rhizobacteria (PGPR). Another major benefit of PGPR is the ability to produce antibacterial compounds that are effective against certain plant pathogens and pests (Dey et al., 2004; Herman et al., 2008; Minorsky, 2008). Bioinoculants are environmentally friendly microorganisms with a variety of products that are frequently used to boost the soil's potential and supply the host plant with the necessary nutrients it needs. Chemical fertilisers should not be used because they significantly alter the number of microorganisms in the soil. The use of PGPR in bioinoculant form results in successfully sustained agricultural yield development, and such formulated goods contain living microbial cells of bioinoculants, which also aid in seed treatment and augment the mobilisation process of nutrients by the low-cost approach (Chaudhary et al., 2020).

Concerns have been raised with regard to the environment's depletion, the safety of animals, and human health as a result of the unrestricted use of chemical fertilisers and pesticides to improve crop yields. Soil degradation has accelerated concurrently as a result of poor land management, various environmental causes, drought, flooding, high temperatures, and soil salinity. The use of biofertilizers like plant growth-promoting rhizobacteria (PGPR) is an eco-friendly solution to these problems (Basu et al., 2021). By colonising roots, PGPRs—free-living bacteria—can promote plant growth and/or offer defence against biotic and abiotic stressors (Kloepper and Schroth 1978). Although these microbes have long been regarded as promising tools, researchers are still trying to understand how they operate and how well they perform in the field. Even under challenging environmental conditions like drought, salinity, and high temperatures, the use of plant growth-promoting microorganisms has proven to be a more environmentally friendly method ideal for improving plant culture in a variety of species.

**Types of bioinuculent**

The introduction of bio inoculants, which contain advantageous microorganisms, to seeds, seedlings, or soil can increase plant growth and productivity Rhizobium bioinoculants are used to improve nitrogen availability in the soil for leguminous crops, thereby reducing or eliminating the need for synthetic nitrogen fertilizers. (Fig. 1.) (Turner, 2013; Lebeis, 2014; Smith, 2015).

Microbes possess the ability to assimilate and acquire vital nutrients from the soil, facilitating their availability to plants. They contribute to the improvement of soil physicochemical properties and have the capacity to modulate various aspects of plant biology, including the production of antibiotics, plant hormones, secondary metabolites, and other signalling molecules. In addition, a variety of biostimulants released by microorganisms have a profound effect on plants' physiological and metabolic processes. (Nelson,2015 Leach,2017,  Massalha,2017)

**Rhizobium:**

Rhizobium is a genus of bacteria that forms a symbiotic relationship with leguminous plants, such as peas, beans, and clover. These bacteria infect the roots of these plants and form specialized structures called nodules, where they convert atmospheric nitrogen into a form that the plant can utilize. This process is known as biological nitrogen fixation. Performance of microbial inoculants as Rhizobium inoculants under field conditions is the principal criterion of selection as a commercial biofertilizer.

According to Lucy et al. (2004), soil is a highly heterogeneous and unpredictable environment, making it frequently challenging to attain desired results. Because of this, regular application of biofertilizers encourages soil microbes to persist and multiply, which helps to maintain soil fertility and promote sustainable agriculture (Choudhury and Kennedy 2004).

 Rhizobium use in field and pot trials consistently led to statistically significant advances in a number of crop production metrics, including grain yield, root length, leaf length, and plant weight (Naher et al. 2009; Mehboob et al. 2011; Yanni and Dazzo 2010). Rhizobium sp. (SB16) colonisation of Mayang Segumpal rice resulted in a 36% increase in plant biomass over the non-inoculated control and a 4.47% increase in tissue nitrogen content (Naher et al. 2009). Abera et al. (2016) showed considerably increased mean grain yields of maize in Toke Kutaye (western Ethiopia) following the application of half the prescribed nitrogen fertiliser after faba bean precursor crop with rhizobium inoculation. Similar to this, Saini et al. (2004) proposed that only 50% of the necessary fertiliser might be administered along with bioinoculants (Rhizobium, or A. brasilense, Bacillus megaterium, and Glomus fasciculatum) for maximum crop output of sorghum (Sorghum bicolor L.) and chickpea (Cicer arietinum L.).

Three of the nine bacteria strains assessed by Canto et al. (2023), which studied indigenous drought-tolerant rhizobium strains as possible biostimulants for common bean in Northern Spain, were found to be highly effective under drought (specifically 353, A12, and A13). These strains outperformed plants that had been inoculated with the CIAT899 reference strain and plants that had been chemically fertilised with N while still maintaining high levels of infectiveness (nodulation capacity) and efficacy (shoot biomass output). A novel and sustainable production method for pulses has emerged: inoculating them with biostimulants such rhizobium strains with excellent nitrogen fixation efficiency and drought resistance.

**Azospirillum**

 Azospirillum is a genus of bacteria that is commonly used as a bioinoculant in agriculture. These bacteria are beneficial plant growth-promoting rhizobacteria (PGPR) that can establish a symbiotic relationship with a wide range of plants, including cereals, vegetables, and grasses. While Azospirillum bacteria are not capable of fixing atmospheric nitrogen like Rhizobium, they can promote nitrogen availability in the soil through other means. Azospirillum can solubilize and mineralize organic nitrogen in the soil, making it more accessible to plants. Additionally, they can enhance the uptake of nitrogen from synthetic fertilizers or organic sources.

According to Salantur et al. (2006), the inoculation of plants with Azospirillum causes a considerable change in a number of growth parameters in different cereals, including an increase in plant biomass, nutrient uptake, tissue N-content, plant height, leaf size, tiller numbers, root length, and volume. Azospirillum inoculation resulted in an increase in yield along with an increase in nitrogen levels, that was attributed to higher levels of nitrogen fixation or assimilation by vegetation (Wani and Lee, 1991). According to Rafi and Charyulu (2016), Setaria italic showed an increase in N content, dry shoot, root, and plant weight. The uptake of mineral ions was greatly improved (30%-50% over controls) when Z. mays and Sorghum bicolor were injected with A. brasilense strain Cd or Sp-7 (Lin et al., 1983). Additionally, a 20%–30% increase in shoot dry weight was noted.

**Mycorhizae:**

 Mycorrhizae, specifically arbuscular mycorrhizal fungi (AMF), are often used as bioinoculants in agriculture and horticulture. Mycorrhizal fungi have a remarkable ability to extend their fine hyphae into the soil, greatly increasing the effective root surface area for nutrient absorption. They can efficiently extract nutrients, such as phosphorus, nitrogen, potassium, and micronutrients, from the soil and deliver them to the host plant. This improves nutrient uptake, especially in soils with low nutrient availability. Mycorrhizal fungi form a symbiotic relationship with the roots of most plant species, including trees, shrubs, and agricultural crops. This symbiosis benefits both the fungi and the plants involved.

Baisru et al (2021) examined 68 mycorrhizal products from 28 manufacturers across Europe, America, and Asia. The products were evaluated for a variety of characteristics, including physical forms, arbuscular mycorrhizal fungal composition, quantity of active components, claims of purpose served, mode of administration, and recommendation. They claim that all goods are made from the Glomeraceae, of which three species predominate overall: Rhizophagus irregularis (39%), Funneliformis mosseae (21%), and Claroideoglomus etunicatum (16%). The benchmark products' least frequent species is Rhizophagus clarus. Just 19 percent of the items include extra beneficial bacteria, and one-third of them are single species AMF. 44% of the examined items exclusively contain AMF, while the remainder also contain other active components. The vast majority of the items (84%) made claims about the advantages of plant nutrients.

Faye et al. (2013) conducted a two-step experiment using maize to evaluate 12 arbuscular mycorrhizal fungus (AMF) inoculants in a greenhouse setting. Seven inoculants boosted root colonisation levels in comparison to control soil six weeks after planting, however only three inoculants modestly increased the shoot biomass of maize plants.

**Plant growth promoting bacteria:**

**Phosphate-solubilizing bacteria (PSB):**

These bacteria convert insoluble forms of phosphorus in the soil into soluble forms that plants can absorb. PSB produce organic acids, such as citric acid and gluconic acid, which can chelate phosphorus, releasing it from minerals like rock phosphate. Inoculating seeds with P-solubilizing microorganisms is a potential method that might assist with phosphorus deficit status. Examples of PSB include species of Bacillus, Pseudomonas, and Enterobacter.

The seed cotton production and plant height were greatly improved by phosphate-solubilizing Bacillus sp.Bacillus inoculation at 90 kg P ha-1 resulted in the highest seed cotton yield (1733.3 kg ha-1). At all P values, bacterial inoculation increased seed cotton yield compared to the corresponding control. At 30, 60, and 90 kg P ha-1, the inoculation-induced increases in seed cotton yield were 8.08, 7.93, and 7.57%, respectively. According to Ponmurugan and Gopi (2006), PGPR, which has the ability to solubilize phosphate, increased the production of growth hormones, the availability of phosphorus, and the rate of nitrogen fixation.

 According to Panhwar et al. (2014), PSB populations were found to be greater in rhizosphere soil than in non-rhizospheric soil, with the highest populations being discovered in PS and Pikovskaya and the lowest in PA media plates. The PSB9 strain produced on an NBRIP plate has the highest P solubilizing activity (69.58%). PSB that has been isolated could create a variety of organic acids and growth hormones like IAA. Even though the majority of the identified strains are capable of growing in nitrogen-free semi-solid medium and are able to produce siderophore, a number of PSB isolates from the Bacillus sp. have been demonstrated to have an antagonistic impact against R. solani (sheath blight).

Liu et al. (2015) The NBRIP liquid medium culture revealed that four PSB strains reduced the medium pH (to 4.3) after three days of incubation and released WS-P up to 523.69 mg/l, while the Krome3 strain dissolved 95.3% of the added tricalcium phosphate after 35 days of incubation. After being incubated in sand, PSB raised WS-P but not Mehlich-3 P.

**Silicate-solubilizing bacteria:**

Silicate-solubilizing bacteria solubilize insoluble forms of silica in the soil, releasing plant-available silicon. Silicon plays a role in plant defense mechanisms, enhancing disease resistance and stress tolerance. The transformation of silicates into soluble silica is carried out by a species of bacteria known as silicate-solubilizing bacteria (SSB). These bacterial communities produce the enzyme silicase, which breaks down silicates into soluble silica so that plants can absorb silicon.

Sulizah et al (2018) isolated five silicate solubilizing bacteria OS4, OS5, OS7, OS12 and OS13. In Bunt and Rovira broth, OS12 solubilized 1,053 ppm of silicate, while OS7 achieved the greatest Solubilizing Index (1,10). It has been discovered that Bacillus, Pseudomonas, and Paenibacillus bacteria may dissolve silicate minerals.

**Sulfur-solubilizing bacteria (SSB):**

Sulphur is a crucial ingredient for plant growth, and SSB aid in saturating insoluble sulphur molecules in the soil so that plants can access them. These bacteria generate enzymes that change sulphur from an insoluble form to one that is soluble. Thiobacillus and Rhodococcus species are examples of SSB.

Malviya et al (2022) Out of the thirteen sulfur-oxidizing bacteria (SOB) that Out of the thirteen sulfur-oxidizing bacteria (SOB) that) isolated from coal mines, only two isolates—Sulfur. maltophilia DRC-18-7A and Sulphur. pavanii DRC-18-7B—performed well as microbial inoculants. These plants prompt the pigeonpea to develop secondary and tertiary roots earlier than the untreated control.

**Potassium-solubilizing bacteria (KSB):**

KSB are capable of solubilizing insoluble potassium minerals in the soil, releasing potassium ions that can be taken up by plants. These bacteria produce organic acids that aid in the solubilization process. Bacillus, Pseudomonas, and Azotobacter are examples of KSB.

Supanjani *et al.* (2006) reported adding P- and K-containing rock materials to the soil and inoculating Capsicum annuum with phosphorus- and potassium-solubilizing bacteria increased P availability from 12% to 21% and K availability from 13% to 15% in the soil as compared to control, which improved N, P, and K uptake in the crop.

According to Ghumare et al. (2014), microphos biofertilizers can provide 30 kg P2O5/ha when injected into seeds or seedlings. According to Kumar and Kumawat (2014), summer mung bean cv. T-1 that has been planted with a combination of chemical fertiliser (10 kg N and 20 kg P2O5/ha), 50% N as vermicompost (10 kg/ha), and biofertilizers (20 g PSB/kg seed) improves crop productivity, economics, and soil fertility.

**Zinc-solubilizing bacteria (ZSB) :**

ZSB are a group of beneficial microorganisms that have the ability to solubilize insoluble forms of zinc in the soil, making it available for plant uptake. These bacteria play an important role in improving zinc availability, which is essential for plant growth and development.

In a field experiment, Ramarethinam and Chandra (2005) found that the inoculation of the potash-solubilizing bacteria *Frateuria aurantia* greatly boosted egg plant output, plant height, and K absorption compared to control. According to Mikhailouskaya and Tchernysh (2005), the effect of inoculating K-mobilizing bacteria on numerously eroded soils was comparable to yields on moderately eroded soil without bacterial inoculation, which led to an increased wheat yield up to 1.04 t/ha.

 The important crops such as maize (,Goteti et al 2013, Hussain et al., 2015, Biari et al 2008, Omari et al 2016), rice (Vaid et al., 2014, Tariq et al., 2007, Zeb et al., 2018, Gontia-Mishra et al., 2017, . Idayu et al., 2017) and wheat (Ramesh et al., 2014, Kumar et al., 2017 Singh et al., 2017, Rana et al., 2012, Kalinowski et al., 2000, Khande et al., 2017) Considering that the grain components from these crops provide the most significant staple foods on a global scale, they have been thoroughly examined for Zn biofortification in response to ZSB inoculants. The Zn translocation (%) was increased by a putative ZSB microbial strain called Bacillus sp. in two different Basmati rice types, ranging from 22-49% (for Basmati-385) to 38% (for Basmati-381) and 18–47% (for Super-Basmati Rice) (Shakeel et al. 2015). The study by Wang *et al*. (2014) illustrated the role of “*Enterobacter* sp. SaCS20” and “Sphingomonas sp. SaMR12” in improving the Zn content in polished rice by 11.2% and 13.7%. Bacterium “Rahnella sp. JN6” improved the plant growth and increased Zn accumulation in Brassica napus (oilseed rape) in pot experiments (2013).

*Thiobacillus thioxidans, Saccharomyces sp., and B. subtilis* are only a few examples of the various bacteria that may solubilize zinc from soil. According to Raj (2007), these microbial inoculants can be utilised as biofertilizers to solubilize zinc in soil. When soil zinc is present in higher concentrations in other insoluble forms, such as zinc oxide (ZnO), zinc carbonate (ZnCO3), and zinc sulphide (ZnS), instead of the more expensive zinc sulphate, such Zn-solubilizing bacteria (*Bacillus* sp.) may be utilised as biofertilizers.

**Conclusion:**

In addition to others, the phylum Ascomycota, Actinobacteria, Basidiomycota, Bacteroidetes, Chlorobi, Cyanobacteria, Chlorophyta, Euryarchaeota, Firmicutes, Gemmatimonadetes, Mucoromycota, Proteobacteria, and Tenericutes contain mineral-solubilizing bacteria. Mineral solubilizing microbes (MSMs) produce a variety of substances that either indirectly or directly promote plant growth and development, which includes plant growth regulators, phosphorus, potassium, zinc, selenium, and silicon dissolution, biological nitrogen fixation, siderophores, ammonia, hydrogen cyanide, hydrolytic enzymes, and bioactive substances/secondary metabolites.

In an era when agriculture has to deal with a multitude of environmental concerns, biofertilizers can help address the task of feeding the growing world population. Adopting biofertilizers into conventional farming adheres to and comprehending their positive impacts are vital.

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Figure 1: Types of bioinoculant

