



# A REVIEW ON MICROBIAL ASSOCIATION : ITS POTENTIAL AND FUTURE PROSPECTS IN FRUIT CROPS

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## Abstract

Biofertilizers and bio-formulations have emerged as supplements to mineral fertilizers and hold a promise to improve yield as well as quality of fruit crop. Biofertilizers and bio-formulations offer a more economical as well as eco-friendly option. Recently, the importance and management of beneficial microorganisms has increased and led to the establishment of commercial trends around the world. The production of biofertilizers and their commercialization is focused on the creation and support of sustainable production system. They occupy an important place as they help in making important plant nutrients thus, providing a scope for reduction in use of costly chemical fertilizers, which can pollute soil in long term use. Moreover, other properties such as auxin production have been attributed to biofertilizers. The increasing concern about the environment and socio- economic impact of chemical agriculture has led many farmers and consumers to seek alternative practices for agricultural sustainability and marketability.

**Key words :** Bio-inoculants, biofertilizers, mycorrhiza, microbial, fruits.

## Introduction

Soil is one of the last great scientific frontiers and rhizosphere being the most active portion of that frontier in which various bio-geochemical processes influence a host of landscape and global scale phenomenon's. A better understanding of these processes is critical for maintaining the health of the plant and feeding the organisms that live on soil and prolong soil productivity and biodiversity of the environment (Morrissey *et al.*, 2004). There is a small but concerted effort under way to harness the root system of plants in an attempt to increase yield potentials of staple food crops in order to meet the projected doubling in global food demand in the next 50 years (Zhang *et al.*, 2010 and Giles *et al.*, 2008). These efforts are being done in the face of a changing global climate and increasing global population, which will inevitably require more productively grown food, feed and fibre on less optimal and often infertile lands; which already prevails in many developing countries (Tilman *et al.*, 2002). Meeting the global challenges of climate change and population growth with a better understanding and control of rhizosphere

processes will be one of the most important science frontiers of the next decade for which a diverse, interdisciplinary trained workforce will be required. The rhizosphere concept was first introduced by Hiltner to describe the narrow zone of soil surrounding the roots where microbial population are stimulated by root activities (Hiltner, 1904).

Thin layer of soil immediately surrounding the plant roots is extremely important and active area for root activity and metabolism is known as 'rhizosphere'. Since bacteria are the most abundant microorganisms in the rhizosphere, it is highly probable that they influence the plants physiology to a greater extent, especially considering their competitiveness in root colonization (Berg and Smalla, 2009). Microorganisms that colonize the rhizosphere can be classified according to their effects on plants and the way they interact with roots, some being pathogens whereas other trigger beneficial effects. Arbuscular mycorrhizal fungi's (AMF) are obligate biotrophs, which can form mutualistic symbiosis with the roots of around 80% of plant species (Abbott and Robson, 1982). On the basis of this symbiosis they can establish

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extra-radicle mycelia, which disperse outside the roots to have access to a greater quantity of water and soil minerals for the host plants. In return, the symbiont receives plant carbohydrates for the completion of its life cycle. The mycorrhizal symbiont almost occurs in all fruit tree species grown in nursery or field (Calvet *et al.*, 2004). It is well documented that AM symbiosis can increase plant growth and nutrient uptake, improve fruit quality and enhance several abiotic stresses such as low temperature stress, drought, salt stress, etc. (Mena-Violante *et al.*, 2006 and Miransari, 2010). Inducing better root colonization & enhances mycorrhizal beneficial roles on fruit trees.

However, the beneficial roles depend on both fungal and fruit crop species. Rhizobacteria's inhabit plant roots and exert a positive effect ranging from direct influence mechanisms to an indirect effect. So, the bacteria inhabiting the rhizosphere and beneficial to plants are termed Plant Growth Promoting Rhizobacteria (PGPR). There are several PGPR inoculants currently commercialized that promote growth through at least one mechanism; suppression of plant disease (Bioprotectants), improved nutrient acquisition (Biofertilizers), or phytohormone production (Biostimulants).

The microorganisms which improve nutrient acquisition are known as biofertilizers. The biofertilizers are microbial preparations containing living cells of different microorganisms, which have the ability to mobilize plant nutrients in soil from unusable to usable form through biological process. They are eco-friendly and play a significant role in crop production. These were mainly used for field crops but now-a-days their potential can be seen to be fully exploited for fruit crops also. Biofertilizers play a very significant role in improving soil fertility by fixing atmospheric nitrogen both in association with plant roots and without it. They help in solubilizing insoluble soil phosphate and produce plant growth substances in the soil. Biofertilizers fix 20-200 kg N/ha/year, solubilize phosphorus in the range of 30-50 kg P/ha/year and mobilize P, Zn, Fe, Mo to varying extent. They also help host plants to impart disease resistance and withstand stress conditions by different mechanisms which vary depending upon the type of biofertilizer agent involved.

Biofertilizers are used in live formulation of beneficial microorganisms which on application to seed, root or soil, mobilize the availability of nutrients particularly by their biological activity and help to build up the lost micro-flora and in turn improve the soil health in general. Thus, the use of biofertilizer is increasing day by day due to increase

in the price of chemical fertilizers, its beneficial effect on soil health and increase in production of crop. The global mandate of Dorrel and Besson (1996) today is to use organic source of plant nutrients to restore the soil health. The fertilizers are not only short in supply, but their high cost and produced at the cost of irreparable loss of non-renewable energy.

Nitrogen fixing bacteria and phosphate solubilizer are the main biofertilizers for horticultural crops. These microorganisms are either free living in soil or symbiotic with plants and contribute directly or indirectly towards nitrogen and phosphorus nutrition of the plants. According to SubbaRao (1998), biofertilizers are otherwise called microbial inoculants, are the carrier based preparation containing beneficial microorganisms designed to improve the soil fertility and help the plant growth by their increased number and biological activity in the rhizosphere. Motsara (1995) reported that inoculation of *Azospirillum* and *Azotobacter* exerted beneficial effect on yield with varying physiological activities, including synthesis of plant growth promoting substances. Biofertilizer is a cost effective renewable energy source and plays a crucial role in reducing the inorganic fertilizer application and at the same time increasing the crop yield besides maintaining soil fertility. In other words, biofertilizers are based on renewable energy sources and are eco-friendly as compared to commercial fertilizers (Verma and Bhattacharyya, 1994). In the recent years, there is an urgent need to supplement the fossil fuel based inorganic fertilizers not only due to the hike in prices of chemical fertilizers, but also a need is felt to maintain long term soil productivity and ecological sustainability. Biological routes of improving soil fertility and health for optimum crop production form a vital component of integrated nutrient management. These routes are operated through the use of FYM and biofertilizer along with chemical fertilizers. Use of biofertilizers for crop production is gaining momentum as they are environmentally safe when compared to chemical fertilizers (Soorianathasundaram *et al.*, 2000).

### **Classes of biofertilizers**

Biofertilizers add nutrients through the natural processes of fixing atmospheric nitrogen, solubilizing phosphorus and stimulating plant growth through the synthesis of growth promoting substances. They can be grouped in different ways based on their nature and function.

### **Occurrence and distribution**

Among the biofertilizers, vasicular-arbuscular mycorrhizae fungi, *Azotobacter*, *Azospirillum* and *Pseudomonas* are of great significance to the fruit crops.

i.e., VAM fungi: The fungus grows in the cortex and develops lipid rich ovoid bodies (vesicles) and highly branched, haustorium like structures within the host cells (arbuscules). The fungus is a member of the family Endogonaceae, the major genus is *Glomus*, considered to be the most abundant of all soil fungi. It is an obligate symbiotic fungus and is not very host specific.

**1. *Azotobacter*** : A free living heterotrophic nitrogen fixing bacteria encounters in neutral to alkaline soil conditions not only provides nitrogen, but produce a variety of growth promoting substances. *Azotobacter* cells are not usually present on the root surface, but are present in rhizosphere. *A. chroococcum* and *A. vinilandii* are deemed to be the most commonly occurring species *Azotobacter* is capable of converting nitrogen to ammonia, which in turn is taken up by the plants (Kamil, *et al.*, 2008). *Azotobacter* sp. can also produce antifungal compounds to fight against many plant pathogens (Jen-Hshuan, 2006).

## 2. *Azospirillum*

*Azospirillum* an associative micro-aerophilic nitrogen fixer commonly found in association with the roots of higher plants is of interest. Low energy requirement, high nitrogen fixation capacity, abundant establishment in roots of higher plants and tolerance to high soil temperature (30-40°C) are responsible for its suitability under tropical conditions. The most important *Azospirillum* spp. is *A. brasilense*, which has a wide range of tolerance against abiotic stresses. The bacteria stimulate plant-growth even in the presence of several stresses such as drought (Noshin *et al.*, 2008). The beneficial effects of biofertilizers have been widely reported in banana (Tiwary *et al.*, 1998 and Mohandas, 1996).

## 3. *Azolla*

*Azolla* is a free floating fern grows naturally in stagnant water of drains, canals, ponds, rivers and potential source of nutrient mainly in field crops. It moderates the soil pH conditions and enhances the availability of nitrogen, phosphorus and certain micronutrients. *Azolla* being green manure can substitute 40-50 kg nitrogen/ha. It is a source of nutrients to poultry, fish and water animals.

## 4. *Cyanobacteria*

*Cyanobacteria*, also known as blue green algae, are free-living N<sub>2</sub>-fixing complex, popularly used in rice crop. *Nostoc*, *Anabaena*, *Aulosira* etc. are well known examples of cyanobacteria. *Cyanobacteria* play an important role in maintenance and build-up of soil fertility,

and yield as a natural biofertilizer. They reduce molecular atmospheric nitrogen to ammonium, which can then be utilized for amino acid and protein biosynthesis.

## 5. Phosphate solubilizing microorganism

This group covers bacteria, fungi and some actinomycetes. These organisms solubilize the unavailable forms of inorganic-P like tricalcium, iron, aluminum and rockphosphates into soluble forms by release of a variety of organic acids like succinic, citric, malic, fumaric, glyoxalic and gluconic acids (Venkateswarlu *et al.*, 2007). Some heterotrophic bacteria and fungi are known to have the ability to solubilize inorganic phosphorous from insoluble sources. Important phosphate solubilizing organisms are *Pseudomonas striata*, *Bacillus polymxa*, *Aspergillus* and *Penicillium digitatum*. These micro-organisms can grow in insoluble phosphatic sources. It is reported that PSB culture increased yield up to 200-500 kg/ha and thus 30 to 50 kg of superphosphate can be saved (Jen-Hshuan, 2006).

## 6. Potassium Solubilizing Bacteria

Potassium solubilizing bacteria (KSB) are heterotrophic bacteria such as *Bacillus mucilaginosus* and *Bacillus edaphicus* are example of microorganisms that used in biofertilizer. These are able to solubilize potassium rock through production and secretion of organic acids.

## Biofertilizer in fruit crops

The use of biofertilizer even though not spread up on a wide scale for all crops, however, there is a growing awareness among the farmers that production can be boosted by the use of biofertilizers in case of cereals, pulses, oil seed and some of the cash crop like vegetable and sugarcane (Verma and Bhattacharyya, 1994). Biofertilizer is a recent concept being used in horticultural crops. Generally, fruit crops have now received more attention than vegetables and ornamental crops. *Glomus fasciculatum*, *Glomus mosseae*, *Azospirillum*, *Azotobacter* and PSB (phosphorous solubilizing bacteria) are found useful for different horticultural crops. Use of biofertilizers particularly inoculating with *Azotobacter* could substantiate 50% nitrogen requirement of banana and produce higher yield over full doses of nitrogen application. The absorption of mobile nutrients like nitrogen also increases in association with VAM fungi.

Beneficial response of *Azotobacter* and *Azospirillum* in enhancing the productivity of banana was also reported by Mohandas (1996). VAM fungi are responsible for more than two fold increased acquisition of the less mobile nutrient elements like P, Ca, S, Zn, Mg

and Cu from the rhizosphere. The high efficiency of *Azospirillum* for fixing nitrogen and better mobilization of fixed phosphorus by VAM even at high temperature can make these highly suited for Mosambi (Manjunath *et al.*, 1983). The per cent of wilting in VAM treated trees of guava was recorded to be lower as compared to untreated trees (Srivastava *et al.*, 2001). The root colonization per cent was higher in *Glomus mosseae* inoculated papaya plants. Nutrient content of N, P, K and also of Fe, Mn, Zn and Cu increased due to VAM inoculation. The improvement in yield parameters in the presence of *Azospirillum* might be due to its dual nature in nitrogen fixation and production of phytohormone substances and increased uptake of nutrients such as nitrogen (Govindan and Purushothaman, 1984).

Shen *et al.* (2013) examined that the compost and BIO treatment more effectively controlled fusarium wilt disease banana. The treatment resulted in higher total soluble sugars (TSS) to titratable acidity (TSS : TA) ratios, yield, culturable and total soil bacteria and culturable actinobacteria population. All these results confirmed that application of bio-organic fertilizer could more effectively control *fusarium* wilt disease in field conditions by improving soil microbial communities. Baset *et al.* (2010) suggested that inoculations of plant growth promoting rhizobacteria (PGPR) have been shown to produce beneficial effects through growth stimulation in legumes and cereals and an attempt has been made to use rhizobacteria in bananas. Studies on biofertilizers along with chemical fertilizers were undertaken for assessing their effect on growth, yield and quality in Mosambi. Effects of floral and foliar application of plant growth promoting rhizobacteria (PGPR) on yield, growth and nutrition of sweet cherry was studied by Esitkena (2006). Co-inoculation of BA-8 + OSU-142 increased Fe and Zn contents of leaves up to 50.5 and 35.5% compared with the control, respectively. Manganese content of leaves significantly also increased by BA-8 (26.6%) and OSU-142 (27.0%) applications compared with the control. The results of the present study suggested that *Pseudomonas* BA-8 and *Bacillus* OSU-142 alone or in combination have a great potential to increase the yield, growth and nutrition of sweet cherry plant. Role of biofertilizers in fruit crops are discussed under following headings:

### 1. Effect of biofertilizer on vegetative character

Nripendra *et al.* (2012) conducted an experiment on pomegranate and observed that mycorrhizal treatments were superior over non-mycorrhizal treatment in pomegranate. Increase in height, root length, number of leaves, dry weight, of shoot and roots and mycorrhizal

dependency percentage in pomegranate was observed when *Glomus epigaeum* (GE) + *G. mosseae* + *Gigaspora calospora* were used. VAM significantly increase growth of plants compared to non-mycorrhizal control and was also effective in increasing nutrient uptake by the plants. VAM influenced growth attributing character and yield attributing component. About 50% saving of phosphorus was achieved through the use of VAM.

Khade and Rodriguez (2009) reported that VAM fungi (*Glomus fasciculatum*) were found to be effective in papaya in increasing the plant height, stem girth, petiole length and number of leaves. Sharma and Bhutani (1998) investigated on the response of VAM on apple seedlings in combination with VAM, *Azotobacter* and inorganic fertilizers. They reported that dual inoculation with *Glomus fasciculatum* and *Azotobacter chroococcum* produce larger plants, which had a more leaf area. In Egypt, Wu *et al.* (2011) conducted an experiment on peach (*Prunus persica* L. Batsch) seedlings inoculated with *Glomus mosseae*, *G. versiforme* and *Paraglomus occultum*, respectively. After 100 days of mycorrhizal inoculations, mycorrhizal colonization of one-year-old seedlings ranged from 23.4% to 54.9%. Generally, the formulated mycorrhizal symbiosis significantly improved plant growth performance, such as plant height, stem diameter, shoot, root or total dry weight. Kerni and Gupta (1986), found greatest percentage increase in seedling height of mango, seedling diameter and number of leaves with treatment 49 g N, *Azotobacter* + 49 g N, or *Azotobacter* alone as compared to control. Kumar and Shanmugavelu (1988) reported that both soil and foliar application of nitrogen and in combination with *Azotobacter* increase the plant height, plant girth, number of hands bunch and number of finger/ hands significantly in banana cv. Robusta. Mohandas (2012) studied effect on rootstock cv. Totapuri inoculated with AM fungi on scions of mango hybrids Arka Puneet and Arka Aruna and found that nursery seedling produces shoot earlier compared non mycorrhizal fungi. Patil and Shinde (2013) was reported that 50% RDF + FYM + *Azotobacter* (50g) + PSB (50g) + VAM *Glomus fasciculatum* (250 g) found beneficial for growth and yield of Banana cv. Ardhapuri. The result indicates the application of 50% (compost, rock phosphate and feldspar) + 50% of the NPK mineral recommended fertilizers N (157 g/vine), P O (87 g/vine) and K O (112 g/vine) + biofertilizer was the best management system for ensuring the best vegetative growth parameters as shoots length, number of leaves/shoot and leaf area and leaf mineral content in the "Superior Seedless" grape vine. Therefore, these organic

and natural rocks fertilizers in combination with NPK biofertilizers can reduce the need for about 50% of NPK mineral fertilizers. In pomegranate the study was revealed that the Microbial inoculum (*Azotobacter* spp. *Azospirillum* spp. and *Bacillus megatherium* which found in Biogein, Nitrobine, phosphorein; in addition, Arbuscular Mycorrhizal (AM) fungi were used to reduce the recommended dose of nitrogenous and phosphoric fertilizers of the trees. The growth parameters *i.e.* shoot length, leaf area, leaf dry weight and leaf chlorophyll of treated trees with microbial fertilization were significantly increased compared with uninoculated control trees (Eman *et al.*, 2006). The papaya seedling inoculated with (Arbuscular Mycorrhizal Fungi) *Glomus intraradices*, seedlings inoculated with *Glomus mosseae* and with mixed inoculums [*Glomus intraradices* + *Glomus mosseae*]. In the inoculated seedling with fungi significantly increase in growth parameters *viz.*, plant height, stem girth, leaf area and root length and also increase in fresh and dry weights of shoot and root systems was observed (Khade and Rodrigues, 2009). In a study on apple, Sharma and Sharma (2006) reported that AM spore population with leaf area, shoot extension growth and yield was found to be positive and significant. Similarly, per cent root colonization also had positive and significant correlation with leaf area and shoot extension growth. They attributed the positive and significant correlation of AM spore population and intensity of root colonization with root and shoot growth to enhanced nutrient uptake by plant root from soil. Nursery and field experiments were carried out Aseri *et al.* (2008) to assess the effectiveness of selected N<sub>2</sub>-fixing bacteria and AM fungi alone or in combination, on the growth and biomass production of *Punica granatum*. In both experiments, the combined treatment of *Azotobacter chroococcum* and *Glomus mosseae* was found to be the most effective. Biofertilizer inoculation had enhanced shoot dry weight by 16–36%. Thus a vast set of studies have proof their efficiency as useful in the crop based enhancement of vegetative growth.

## 2. Effect on yield characters

The beneficial effect of *Azotobacter* inoculation in fruit and vegetable crops was well discussed by various workers. To study the effect of biofertilizers on growth, yield and fruit quality in low chill pear cv. Gola after subjecting it to different doses of biofertilizers (*Azotobacter*, VAM and PSB) revealed that *Azotobacter* @ 30 g incorporated was the most effective for enhancing vegetative growth of tree, fruit yield and physical quality of fruit. Chemical qualities of fruit significantly improved by the treatment of 90 g VAM incorporated. Treatment

of 60 g *Azotobacter* enhanced the leaf phosphorus content more effectively (Manoj *et al.*, 2013). Plant growth promoting effects of *Bacillus* M3, *Bacillus* OSU-142 and *Microbacterium* FS01 were tested alone or in combination on apple (*Malus domestica* L.) cv. Granny Smith in terms of yield and growth results shows that root inoculation of PGPR strains significantly increased cumulative yield (26.0-88.0 %) in apple cv. Granny Smith compared with the control (Karlidag *et al.*, 2007). In a field experiment, three-year-old Kinnow trees budded on JattiKatti (*Citrus jambhiri*) rootstock were inoculated by three different Arbuscular mycorrhiza (AM), *Glomus manihotis* (T<sub>1</sub>), *Glomus mosseae* (T<sub>2</sub>), and *Gigaspora gigantia* (T<sub>3</sub>), separately or in combination (T<sub>4</sub>). AM application improved growth attributes of existing and newly developing shoots. Flowering and fruiting were also bettered by AM inoculation (Shamshiri *et al.*, 2011).

Keeping in view the importance of biofertilizers, as alternative environment-safe fertilizers Pesakovic *et al.* (2013) conducted a study on the effect of biofertilizers *viz.*, diazotrophic nitrogen-fixing bacteria *Klebsiella planticola* and nitrogen fixing and phospho-mineralizing bacteria (*Azotobacter chroococcum*, *A. vinelandi*, *Dexia* sp., *Bacillus megatherium*, *B. lichenformis* and *B. subtilis*) on strawberry cv. 'Senga Sengana' under green house conditions. They noticed significantly higher yield attributes with respect to the application of liquid inoculum of diazotrophic bacteria *Klebsiella planticola*.

## 3. Effect on soil characteristics

The plants inoculated with *Azotobacter* and *Azospirillum* derive positive benefit in terms of enhancement in uptake of NO<sub>3</sub><sup>-</sup>, NH<sub>4</sub><sup>+</sup>, H<sub>2</sub>PO<sub>4</sub>, K<sup>+</sup> and Fe<sup>2+</sup> increased nitrate reductase activity in plants and production of antibacterial and antifungal compounds (Wani, 1990). Combined application of inorganic fertilizer and biofertilizers in banana cv. Barjahaji significantly increased the available NPK status, organic C and microbial biomass and dehydrogenase activity in soil after harvest. Shirsath *et al.* (1998) reported that VAM inoculation either singly or in combination significantly increased root and shoot dry weight as well as P uptake over non-mycorrhizal treatments. Combined inoculation of *Acaulospora calospora* + *G. mosseae* + *G. margarita* and single inoculation of *G. mosseae* were superior in increasing dry weight of ber seedlings as compared to rest of the inoculation treatments. Experiment conducted at TNAU, by Aneesa Rani and Sathiamoorthy (1997) on effect of organic and biofertilizers on root enzyme activity of papaya cv. Co-6 revealed that highest dehydrogenase enzyme activity in treatment substituted

with 50% organic N and 50-70% organic P along with biofertilizer *Azospirillum*, *Phospobacteria* and VAM. Ruiz (1992) from Cuba, observed that the quantities of beneficial microorganisms in the soil increased considerably due to the use of *Azotobacter*, mycorrhiza and phosphorins in banana. The commercial yield is also increased by 25-30% and save 50% of inorganic fertilizers. In Banana, the plants of cv. Elakki Bale were studied for their response to inoculation with biofertilizer by (Mohandas, 1996) *viz.* VAM, phosphate solubilizing bacteria and *Azospirillum brasillense* alone or in combination. VAM colonization was found up to 70-80% while that of PSB and *Azospirillum* was found up to 70%. The available P in the soil increased in VAM and PSB treatments and available soil N increased in *Azospirillum* treatment. Sharma *et al.* (2014) found out that out of eleven phosphate solubilizing *Pseudomonas* species isolates, four were more promising for the management of replant problem of apple. Plant growth is frequently limited by an insufficiency of phosphates, which were considered one of the most important growth-limiting environmental factors also one of the major factors in replant disease.

#### 4. Effect on quality characters

Singh *et al.* (2000) reported that the treatment combination of P + VAM + N was the best treatment for producing better growth and yield of high quality fruit in Mosambi. This treatment also influences plant height, trunk diameter, canopy volume, root growth and biomass production as compared to control. The use of phosphorene was found to improve fruit set and yield as well as physical and chemical properties of fruits than control. Suresh and Hasan (2001) in West Bengal evaluated the response of inoculation with *Azospirillum* and phospho-bacteria on fruit quality of banana (*Musa MA*) cv. Giant Governor by manipulating the doses of nitrogen and potassic fertilizers. The results revealed that inoculation of biofertilizers along with the application of recommended dose of fertilizer proved most effective in improving fruit quality of Dwarf Cavendish banana cv. Giant Governor. Rana and Chandal (1999) reported that the plant growth, yield and fruit quality of strawberry were significantly increased with the application of biofertilizer and nitrogenous fertilizers. Maximum TSS content was observed with *Azotobacter* inoculation along with 80 kg N/ha. Sharma (2002) observed that application of *Azotobacter* + 75% inorganic N definitely improve the quality of banana *viz.*, total sugar (16.88%), starch (2.28%) and protein (1.50%) and were recorded significantly higher the over recommended fertilizer dose. Abd El-Migeed (2007) conducted an experiment on

Washington Navel Orange by using organic and biofertilizer and obtained result showed that treatments included biofertilizers improved average fruit weight, vitamin C content and peel thickness than that without adding biofertilizers. Singh and Banik (2011) reported that Application of 500:250:250 g NPK/tree + 50 kg FYM + 250 g *Azospirillum* of INM system is best for achieving better yield and quality in mango cv. Himsagar. The combined biofertilizer application of *Azotobacter* + *Azospirillum* + AM + PSM in mango cv. Himsagar was most effective in improving the soil and fruit and size of fruits and the availability of maximum organic carbon (0.97%), available N (294.11 kg ha<sup>-1</sup>), P (37.44 kg ha<sup>-1</sup>) and K (231.00 kg ha<sup>-1</sup>). Leaf nutrients were also improved with the application of biofertilizers (Datta and Kundu, 2012). The fruit quality of Strawberry Cv. Chandler *viz.* total soluble solids, total sugars, ascorbic acid and anthocyanin content was highest in fruits obtained from plants supplied with 25 per cent nitrogen through FYM + 75% nitrogen in the form of urea + *Azotobacter* recording 6.81 °Brix, 4.73 per cent, 73.71 mg/100g fresh berries and 0.191 OD respectively (Umar *et al.*, 2009). Dutta *et al.* (2014) was found that among different treatments *Azospirillum* + *Azotobacter* + VAM was most effective in improving the fruit quality in guava followed by *Azotobacter* + VAM. This treatment also resulted in maximum content of leaf minerals (N, P and K). The efficiency of inorganic fertilizer at three levels was more when supplemented with both *Azotobacter* and VAM in mango cv. Amrapali. Higher fruit yield was obtained when the plants were treated with 100% NPK + *Azotobacter* + VAM (98.1 kg/plant) or 75%NPK + *Azotobacter* + VAM (93.5 kg/plant) as compared to much lesser yield (60 kg/plant) with 100% NPK. It was concluded that the treatments 100%NPK + *Azotobacter* + VAM and 75% NPK + *Azotobacter* + VAM were effective and may be adopted to improve the vegetative growth and productivity with quality fruits (Kundu *et al.*, 2011). Leaf petiole nutrient element contents of N, P and K were increased in a significant manner in Flame seedless vine and clusters weight and berries quality, yield was also significantly increased as microbial biofertilization were applied (Hoda, 2012). Sharma *et al.* (2013) reported that the chemical attributes *viz.* TSS (12.92 & 12.97 OB), total sugars (8.56 & 8.65 per cent) and the minimum physiological loss in weight (14.29 per cent) after 10 days under ambient conditions were found to be maximum with the application of *Azotobacter* + 50% of N tree<sup>-1</sup> through FYM + 50% of N tree<sup>-1</sup> through inorganic fertilizer. Rao *et al.* (2007) reported that VAM and BBF along with 50% reduction in both N and P fertilizers (when recommended 300 : 120

: 120 kg/ha/y), saving of 50% cost of chemical fertilizers and improvement in soil fertility, leaf quality and cocoon parameters, thus the technology can be recommended to sericulture farmers of semi-arid conditions.

Aseri *et al.* (2008) investigated that inoculation dual inoculation treatment of *Azotobacter chroococcum* and *Glomus mosseae* had resulted in a significantly higher total chlorophyll content as well as accumulation of reducing sugars, total phenols and amino nitrogen in four months old inoculated plants. Total chlorophyll was observed highest in dual inoculated seedlings followed by *G. mosseae* and *A. brasilense* alone. A similar trend was found in reducing sugars and amino nitrogen contents, whereas total phenols were found to be maximum with *A. brasilense* followed by dual inoculation treatment.

### Advantages of biofertilizers

The utilization of microbial products (biofertilizers) has several advantages over conventional chemicals for agricultural purposes :

- ä Biofertilizers can add 20-200 kg N ha (by fixation), liberate growth-promoting substances and increase crop yield by 10-50%.
- ä N-biofertilizers can provide 25-30% of chemical fertilizer equivalent N
- ä PSB biofertilizer can provide 12-20 kg P<sub>2</sub>O<sub>5</sub>/ha
- ä Mycorrhiza can provide adequate P and other micro nutrients
- ä Help in increasing water absorption
- ä Mixed biofertilizer give better impact
- ä Keep soils biologically active and help in soil health maintenance
- ä target organisms seldom develop resistance as is the case when chemical agents are used to eliminate the pests harmful to plant growth
- ä The self-replication of microbes circumvents the need for repeated application.
- ä The proper developed biocontrol agents are not considered harmful to ecological processes or the environment (Wua *et al.*, 2004).
- ä They are neither toxic substances nor microbes themselves will be accumulated in the food chain.

### Mode of action of biofertilizers

The mechanism involved in the plant growth promotion by bio inoculants or biofertilizers can be understood by the following subheads in brief. They have been seen to influence various biological phenomenon in plant. The detail roles with the brief on the parameters

they influence are discussed below.

### Increased availability and uptake of nutrients

Through biological nitrogen fixation, solubilization of insoluble phosphates and mobilization of plant nutrients in more quantities are made available for crop plants by the root associated organisms. Increased nitrogen, phosphorous and potassium content of inoculated plants at different stages of crop growth have been found resulting in significant increase in grain yield. The PGPR inoculation changes many root and shoot parameters. These changes are directly attributed to positive bacterial effects on mineral uptake by the plant. Shamsuddin *et al.* (2000) found increased amounts of P and K uptake in banana plants inoculated with PGPR.

### Production of plant growth promoting substances

Many root colonizing bacteria including the nitrogen fixing *Azospirillum* and phosphorus solubilizing *Pseudomonas* spp. is known to produce growth hormones, which often leads to increased root and shoot growth. Plants differs in the levels and ratio of the hormones required to maintain normal growth and development. Therefore, it might be expected that at different stage of plants respond differently on invasion of hormone producing bacteria. Singh *et al.* (2013) observed significantly greater vegetative growth and leaf nitrogen (N), phosphorus (P) and potassium (K) contents when recommended doses of chemical fertilizer (RDF) along with locally isolated strains of *Azotobacter*, *Azospirillum* and arbuscular mycorrhizal fungi (AMF) were inoculated either solely or in combination with seedlings of Red Delicious and Lal Ambri cultivars. Dobbelarere *et al.* (1999) too suggested that secretions of plant growth promoting substances such as auxins, gibberellins and cytokinins by the bacteria seem to be responsible for these beneficial effects in plants inoculated with consortium of biofertilizers. The increased growth of PGPR inoculated plants might also be due to the higher N accumulation by bacterial N<sub>2</sub> fixation and better root growth, which in turn would have promoted the greater uptake of wate and nutrients (Mia *et al.*, 2005).

### Suppression of growth of phyto-pathogenic microorganisms

Production of antibiotics and bacteriocins by the introduced organisms has been suggested as a possible mechanism by which pathogen are inhibited. Among the various PGPRs identified, *Pseudomonas fluorescens* is one of the most extensively studied rhizobacteria, because of its antagonistic action against several plant pathogens. Banana bunchy top virus (BBTV) is one of the deadly virus which severely affects the yield of banana (*Musa*

**Tables 1 :** Grouping of biofertilizers on the basis of the nature and function.

S. No.	Groups	Examples
<b>N<sub>2</sub> fixing Biofertilizers</b>		
1.	Free-living	<i>Azotobacter, Clostridium, Anabaena, Nostoc</i>
2.	Symbiotic	<i>Rhizobium, Frankia, Anabaena azollae</i>
3.	Associative Symbiotic	<i>Azospirillum</i>
<b>P Solubilizing Biofertilizers</b>		
4.	Bacteria	<i>Bacillus megaterium</i> var. <i>phosphaticum</i> , <i>Bacillus circulans</i> , <i>Pseudomonas striata</i>
5.	Fungi	<i>Penicillium</i> sp., <i>Aspergillus awamori</i>
<b>P Mobilizing Biofertilizers</b>		
6.	Arbuscular mycorrhizal	<i>Glomus</i> sp., <i>Gigaspora</i> sp., <i>Acaulospora</i> sp., <i>Scutellospora</i> sp. & <i>Sclerocystis</i> sp.
7.	Ectomycorrhiza	<i>Laccaria</i> sp., <i>Pisolithus</i> sp., <i>Boletus</i> sp., <i>Amanita</i> sp.
<b>Plant Growth Promoting Rhizobacteria</b>		
8.	Iron uptake	<i>Pseudomonas fluorescens</i>
9.	Potassium uptake	<i>Bacillus</i>
10.	Nitrogen uptake	<i>Azospirillum</i>
11.	Sulfur uptake	<i>Thiobacillus</i>
12.	Phosphorus uptake	<i>Bacillus, Pseudomonas</i>

spp.) crop in Western Ghats, Tamil Nadu, India. It has been demonstrated that application of *P. fluorescens* strain significantly reduced the BBTV incidence in hill banana under greenhouse and field conditions. The investigation made by Raman (2012) on the efficacy of biofertilizers (*Azotobacter chroococcum*, *Pseudomonas striata* and *Trichoderma viride*) considering different aspects of seedling growth under apple nursery showed that highest reduction of the pests attack was observed on inoculation with *Pseudomonas striata* and *Trichoderma viride*, while the infestation minimized in seedling due to antifungal fungistatic organic compounds and toxins on inoculation with combined application of *Azotobacter chroococcum*, *Pseudomonas striata*, *Trichoderma viride*.

There is reduction in the inoculum density of plant pathogens due to the introduction of certain inoculants. They can suppress a broad spectrum of bacterial, fungal and nematode diseases and can also provide protection against viral diseases. Some bacteria support plant growth indirectly, by improving growth restricting conditions either via production of antagonistic substances or by inducing resistance against plant pathogens (Tilak *et al.*, 2005).

### Conclusion

The effect of biofertilizers generally on plant and yield are not as striking as that of chemical fertilizers. Since, it is a living system, thus the influence is subject to environmental, biological and nutritional stresses. Moreover, the performance of the microbial inoculants depends on the quality of the inoculant added as well as

**Table 2 :** List of growth regulators produced by different plant growth promoting rizobacteria.

Phytohormones	PGPRs
Indole-3-acetic acid (IAA.)	<i>Acetobacter diazotrophicus</i> and <i>Herbaspirillum seropedicae</i>
Zeatin and ethylene	<i>Azospirillum</i> sp
Gibberellic acid (GA3 )	<i>Azospirillum lipoferum</i>
Absciscic acid (ABA	<i>Azospirillum brasilense</i>

accurate specification is required to avoid poor performance of the inoculants. To become successful venture this biofertilizer technology must reach to the hands of the farmers. Therefore some points to be pondered *i.e.*, more efforts are put to fully exploit the role of biofertilizers in the farmers' field by means of extension activities like field demonstration, farmers' fair and training programme. To enhance the efficiency of biofertilizers application measure like multiple cultures containing biofertilizers like. *Azotobacter* + PSB + *Azospirillum*, *Azospirillum* + *Azotobacter* etc. can be exploited. The efforts are also desired in the direction of improvement of shelf life of bioinoculant in the biofertilizers during storage. Improvement of carrier material or isolation of strains, which is having more shelf life can help in this respect. Efforts are also desired in the direction of development of simple, low cost technologies so that farmers could be able to produce their biofertilizers at their own place economically.



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