

Review Article

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Role of Microbial Biofertilizers in Vegetable Production- A Review

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ABSTRACT

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Biofertilizers comprises living microorganisms symbiotically associated with plants, when applied to soil or a propagule intends to increase the soil fertility, seed germination and plant growth by encouraging the efficient supply of nutrients to the plants. Since past 50-60 years, soil management practices are mostly reliant on inorganic fertilizers, which has invited serious fortune to the environment (ruining soil fertility and increased pollution) and human health problems (disease risk). Heeding to it researchers have found an eco-friendly alternative by the way of incorporating microbial biofertilizers to supersede chemical fertilizers. Microbial biofertilizers has been analysed as substitute in procuring soil fertility and expanding vegetable production. Microbial fertilizers are promising enough to outstand the chemical fertilizers ensuring sustainable agriculture without disrupting the environment.

Introduction

Biofertilizer term refers to substances containing effective strains of living microorganisms such as fungi, algae, bacteria that can expedite soil microbial activities to enhance the active supply of nutrients in a way that plants can easily incorporate. Inorganic fertilizers indeed played a significant role in enhancing agricultural productivity, but they overstretched the use of renewable sources. Although, farmers are still applying over dosage of chemical fertilizers in lieu of high production nevertheless their excess has cost us soil contamination, soil

toxicity, water table contamination etc. causing environmental pollution (Mahdi *et al.*, 2010) as well as causing several types of cancers and cardiovascular disease in humans (Engel *et al.*, 2000). Their quick action or contribution towards high yield and low price had brought them promptly into the prime focus of the marginal farmers. Among agricultural crops, vegetable crops being short-duration, flexible provided with high productivity accompanying health benefits plays a significant role in food trade especially in India. Vegetables are important for human nutrition in terms of bioactive nutrient molecules such as dietary fibre,

vitamins and minerals, and non-nutritive phytochemicals (phenolic compounds, flavonoids, bioactive peptides, etc.). They are packed with vitamins (C, A, B1, B6, B9, E) and anti-oxidants which can help in growth, repairing of body cells and reduce risk of dreadful diseases like cancers. These nutrient and non-nutrient molecules reduce the risk of chronic diseases such as cardiovascular diseases, diabetes, certain cancers, and obesity (Pennington *et al.*, 2009; Malaterreb *et al.*, 2018).

A high vegetable diet has been associated with reduced risk of human cardiovascular disease (Mullie and Clarys, 2011). Owing to health benefits and high productivity, area under vegetable production is constantly rising, and so is the chemical fertilizers application. Marginal farmers need to improvise their farming skills and incorporate organic farming or at least organic fertilizers and microbial biofertilizers. Organic farming contributes to quality vegetables therefore adoption and application of the microbial biofertilizer is mandatory for modern agriculture to flourish sustainably.

Biofertilizers are the essential component of organic farming as they help in maintaining soil fertility for longer time period. The microbes present in these fertilizers provide nutrient to plants by using different mechanism and also encourage immunity of plants to protect their selves from the attack of diseases and pests as well as abiotic stresses. Biofertilizers add nutrients through the natural processes of nitrogen fixation, solubilizing phosphorus and stimulating plant growth through the synthesis of growth promoting substances. Some biofertilizers are viz. symbiotic nitrogen fixing biofertilizers, free-living biofertilizers, associative symbiotic nitrogen fixing biofertilizers etc. Biofertilizers can be applied directly to the crop or also with the combination of chemical

fertilizers and have different mode of action. If the microbial inoculant is not applied properly, the benefits from the biofertilizer may not be obtained. The biofertilizer can be synthesized in solid or in liquid form for spraying on the plants. Bio-fertilizers are usually amended with carrier material to increase effectiveness of the bio-fertilizers and also enhance the water retention capacity.

The incorporation of microorganisms into carrier materials enables easy handling, long term storage, and effectiveness of the bio-fertilizer. Carrier material such as saw dust, talcum dust, manure, earthworm cast can be used. There is lot of work done by many researchers to know the effects of biofertilizers and they have achieved many successful results. Keeping in mind the above key points, narrating the potential key role biological fertilizers could play if incorporated towards vegetable productivity and sustainable agriculture, we are presenting hereby a review of all researches done in this field exaggerating the fact how microbial biofertilizers could help in safeguarding the environment and prove as an eco-friendly and cost effective input for the farmers.

Nitrogen fixing microbes

Nitrogen fixing microbes comprises of symbiotic nitrogen fixing biofertilizers (including *Rhizobium*, *Azolla* etc.), free living nitrogen fixing biofertilizers [*Azotobacter*, *Cyanobacteria* (blue green algae) etc.] and associative symbiotic nitrogen fixing biofertilizers (*Azospirillum*). Along with these there are microbes which fix phosphorus or solubilize the phosphorus like Phosphorus Solubilizing Bacteria (PSB). Various studies done regarding the application of microbial fertilizers among vegetables and their beneficial effect towards yield and quality parameters have been provided in Table 1.

Symbiotic nitrogen fixing biofertilizers

Rhizobium

These are the widely recognized symbiotic nitrogen fixers that belong to the Rhizobiaceae family and typically consist of various genera, such as *Mesorhizobium*, *Sinorhizobium*, *Allorhizobium*, *Azorhizobium*, *Bradyrhizobium*, and *Rhizobium*. *Rhizobium* are motile, gram-negative, non-sporulating rod type which tend to symbiotically fix atmospheric nitrogen. *Rhizobium* helps reduce the molecular N₂ to NH₃ in the root nodules, which is then readily absorbed by the plant roots.

The N-fixation is carried out by a complex enzyme nitrogenase consisting of dinitrogenase reductase with iron as its cofactor and dinitrogenase with molybdenum and iron as its cofactor (Mahanty *et al.*, 2016). *Rhizobium* can fix 50-200 kg N ha⁻¹ which helps to meet up to 80 to 90% nitrogen need of the crop as their natural presence in nodulating legume crops makes them less dependent on inorganic nitrogen (Kour *et al.*, 2020).

Azolla

It is a symbiotic diazotroph which has the capacity to fix nitrogen in the atmosphere found in temperate and tropical environments. There is a symbiotic relationship between *Azolla* and *Anabaena* cyanobacteria. *Azolla* helps to provide the *Anabaena* with a carbon source and its nitrogen requirement is met by cyanobacteria's atmospheric nitrogen fixation. The benefit of growing *Azolla* as a biofertilizer helps provide N and K requirements to the plant. *Anabaena azollae* is considered to be the most dominant biofertilizers and commonly used for the wetland rice in South-east Asia and estimated to fix around 40-60 Kg N/ha in rice crop (Kannaiyan, 1993).

Free living nitrogen fixing biofertilizers

Azotobacter

Azotobacters are free living nitrogen fixing bacteria which belongs to azotobacteriaceae family and mostly found in alkaline and neutral soils. It does not require any host and fixes the atmospheric nitrogen especially in non-leguminous plants without any symbiotic relationship (Jaga and Singh, 2010). Application of *Azotobacter* bio-inoculants may increase 10-12% crop productivity leading to synthesis of ample amount of biologically active substance like nicotinic acid, biotin, heteroauxins, vitamin B and gibberellins etc, which increase root growth and uptake of the minerals (Jaga and Singh, 2010). *Azotobacter* sp. has the ability to produce antifungal antibiotics and fungi static compounds against pathogens like *Fusarium* sp., *Alternaria* sp., *Trichoderma* sp. etc.

Cyanobacteria

Cyanobacteria referred as "blue-green algae" or BGA, are free living, aquatic, small, unicellular bacteria and possess photosynthetic property i.e. they can manufacture their own food. They are one of the largest bacterial species and the dominant nitrogen fixers among them are *Calothrix*, *Nostoc*, *Anabaena* and *Aulosira* (Sahu *et al.*, 2012). By building up soil fertility, they help to increase yield along with excretion of various substances that promote growth, e.g. amino acids, phytohormones, vitamins (Rodríguez *et al.*, 2006), soil salinity reduction, weed growth prevention, soil P content increase (Wilson, 2006) etc. When inoculated with cyanobacteria, vegetables such as chilli, spinach, radish, tomato have shown the beneficial effects (Thajuddin and Subramanian, 2005). *Nostoc* and *Anabaena* are have been found to fix about 20–25 Kg of N/ha (Kour *et al.*, 2020).

Table.1 Application of microbial biofertilizers among various vegetables and their effects on growth and production

S. no.	Biofertilizer	Plant	Description	Inorganic fertilizer used	References
1.	PSB	Asparagus	All growth characters in asparagus were maximum when PSB was applied with organic fertilizers.	Applied	Palande <i>et al.</i> , (2017)
2.	<i>Azotobacter</i> , PSB	Bottlegourd	Application of 2.5kg dose of <i>Azotobacter</i> and PSB each found highly profitable resulting high C:B ratio.	Applied	Patle <i>et al.</i> , (2018)
3.	<i>Azospirillum</i> , <i>Phosphobacteria</i>	Brinjal	Combined application of microbial fertilizers and chemical fertilizers enhanced growth and yield.	Applied	Latha <i>et al.</i> , (2014)
4.	<i>Azotobacter</i> , <i>Azospirillum</i> , PSB	Brinjal	Growth and yield attributes increased with <i>Azotobacter</i> + <i>Azospirillum</i> +PSB application.	Applied	Solanki <i>et al.</i> , (2010)
5.	<i>Azotobacter</i> and PSB	Brinjal	Morphological and yield characters of the plant were maximum by the application of <i>Azotobacter</i> and PSB alone provided with biotic stress resistance.	Applied	Doifode <i>et al.</i> , (2014)
6.	<i>Azospirillum</i> , PSB	Brinjal	Growth and yield components were maximum with root dipping treatment of 125g <i>Azospirillum</i> and PSB.	Applied	Kiran (2006)
7.	<i>Azospirillum</i> , PSB, VAM, <i>Azotobacter</i>	Broccoli	Curd size, yield, protein, lipid, sulphate contents of broccoli curd was maximized after applying 50% <i>Azospirillum</i> and <i>Azotobacter</i> .	Applied	Singh <i>et al.</i> , (2014)
8.	<i>Azotobacter</i> , <i>Azospirillum</i>	Cabbage	<i>Azotobacter</i> and <i>Azospirillum</i> were used as the organic source of nitrogen.	Applied	Gupta <i>et al.</i> , (2004)
9.	<i>Azospirillum</i>	Carrot	Morpho-physiological, yield, biochemical components increased with <i>Azospirillum</i> application.	Applied	Mog,B. (2007)
10.	<i>Azospirillum</i> , PSB, VAM, <i>Azotobacter</i>	Cauliflower	The application of <i>Azotobacter</i> , <i>Azospirillum</i> , VAM and PSB significantly increased growth parameters when PSB was followed by <i>Azospirillum</i> . VAM followed by PSB gave better leaf width. PSB significantly increased curd size and curd weight.	Applied	Kachari <i>et al.</i> , (2009)
11.	<i>Azotobacter</i> and <i>Azospirillum</i>	Chilli	<i>Azotobacter</i> along with RDF gave the high yield. <i>Azospirillum</i> treatment gave high growth parameters. <i>Azospirillum</i> was found better over <i>Azotobacter</i> .	Applied	Khan & Pariari (2012)
12.	PSB, <i>Rhizobium</i>	Frenchbean	Pod yield/ha maximized with <i>Rhizobium</i> + PSB+ Organic matter.	Applied	Thakur <i>et al.</i> , (2018)
13.	VAM and PSB	Frenchbean	Growth and yield characters of French bean were enhanced due to application of 2kg VAM and 2.5kg PSB per ha.	Applied	Ramana <i>et al.</i> , (2010)
14.	<i>Rhizobium</i>	French bean	Seeds inoculated with synthetic <i>Rhizobium</i> gave the maximum growth, yield and seed quality components.	Not applied	Ahmed <i>et al.</i> , (2016)

15.	<i>Azospirillum</i> , <i>Azotobacter</i> , PSB	Knol-khol	PSB, <i>Azotobacter</i> and <i>Azospirillum</i> inoculation lead to yield and biochemical parameters.	Applied	Choudhary <i>et al.</i> , (2017)
16.	<i>Azotobacter</i> , <i>Azospirillum</i> , <i>Pseudomonas</i>	Lettuce	Plant height, no. of leaves, leaf area index and yield was maximum in seed inoculation with <i>Azospirillum</i> .	Not applied	Chamangasht <i>et al.</i> , (2012)
17.	<i>Azospirillum</i>	Lettuce	<i>Azospirillum</i> inoculated lettuce seeds yield a higher number of transplanted plants with superior quality than non-inoculated ones.	Not applied	Fasciglione <i>et al.</i> , (2012)
18.	<i>Azotobacter</i> , PSB	Okra	The highest yield parameters were obtained with the application of combination of organic manures together with <i>Azotobacter</i> and PSB in okra crop.	Applied	Bairwa <i>et al.</i> , (2009)
19.	<i>Azospirillum</i>	Onion	The application of <i>Azospirillum</i> , increased the yield of onion and also enhanced the nitrogen level in soil.	Applied	Yadav <i>et al.</i> , (2004)
20.	<i>Azospirillum</i> , VAM, PSB	Onion	Seed yield of onion was maximum when treated with GA ₃ along with <i>Azospirillum</i> +PSB+ VAM	Not applied	Waghmode <i>et al.</i> , (2010)
21.	<i>Microbein</i>	Onion	Highest yield of total bulbs and increase in N, P, K, Fe, Mn, Zn, Cu, Pb, NO ₃ and NH ₄ was obtained.	Organic manure applied	Shaheen <i>et al.</i> , (2007)
22.	<i>Azotobacter</i> , PSB	Potato	Application of <i>Azotobacter</i> along with combination of PSB and organic manure enhanced the high yield of potato.	Organic manure applied	Kumar <i>et al.</i> , (2013)
23.	<i>Azotobacter</i>	Potato	<i>Azotobacter</i> alongwith the 75% RDF of nitrogen and phosphorus resulted in higher yield per hectare.	Applied	Kumar <i>et al.</i> , (2006)
24.	<i>Azotobacter</i>	Potato	<i>Azotobacter</i> increased the yield of tuber by 4-24% along with nitrogen.	Applied	Singh,K.(2001)
25.	<i>Rhizobacterin</i> , <i>Microbein</i> and <i>Phosphorein</i>	Potato	Cultivar diamond gave the high yield when treated with phosphorein whereas rhizobacterin treatment gave the highest tuber weight.	Not applied	Farag <i>et al.</i> (2013)
26.	<i>Nitroxin</i> , PSB	Pumpkin	Application of biofertilizer alongwith 50% chemical fertilizers gave the maximum seed yield, fruit yield, photosynthesis rate and chlorophyll content.	Applied	Habibi <i>et al.</i> , (2013)
27.	<i>Azospirillum</i> , PSB, <i>Azotobacter</i>	Radish	Growth, yield, and nutritional quality of radish maximized when one fourth quantity of <i>Azotobacter</i> , <i>Azospirillum</i> , PSB alongwith RDF were applied.	Applied	Shani <i>et al.</i> , (2017)
28.	<i>Azotobacter</i>	Tomato	Application of <i>Azotobacter</i> with RDF (150kg N + 60kg P + 60 Kg/ha) increased the growth and yield parameters of the plant.	Applied	Gabhiye <i>et al.</i> , (2003)
29.	<i>Azotobacter</i> , <i>Azospirillum</i> and PSB	Tomato	Fruit yield maximized when treated with RDF + PSB whereas, fruit quality increased with RDF+ <i>azospirillum</i> +PSB.	Applied	Kadlag <i>et al.</i> , (2007)
30.	Nitrobin	Tomato	Nitrobin biofertilizer enhanced tomato growth and yield characters.	Applied	Gmaa.(2015)

Associative symbiotic nitrogen fixing biofertilizers

Azospirillum

Azospirillum is a gram negative motile bacteria belonging to order Rhodospirillales, with currently 17 species in use as biofertilizers, *Azospirillum brasilense* and *Azospirillum lipoferum* are most widely used species (Rodrigues *et al.*, 2015). It promotes plant growth enhancing IAA, gibberellins and cytokinins production and found to fix 20-40kg/N/year when applied in non-leguminous plants. They can easily be isolated from the soil and from the aerial part of the plant. *Azospirillum*'s key effects consist of modifications in root morphology that eventually stimulates plant growth (Fibach-Paldi *et al.*, 2011). It was determined that by triggering cell wall modifications and osmotic adjustments, it can assist in plant survival under stressful conditions (Groppa *et al.*, 2012; Richardson *et al.*, 2009). The strains of *Azospirillum* are widely applied as biofertilizers in various vegetables (Hungria *et al.*, 2010; Mehnaz, 2015).

Phosphorus Solubilizing Bacteria (PSB)

Phosphorus is a major nutrient that plays a crucial role in fostering crop growth and development (Soetan *et al.*, 2010). Its bioavailability is very poor and therefore not accessible to plants. It is available in two forms in the soil, i.e. organic and inorganic. Inorganic P is supplied in precipitated form by chemical fertilisers and plants cannot take up this form of Phosphorus. Phosphobacteria have the ability of converting the insoluble form of phosphorus to a soluble form and make it available to plant by releasing various organic acids (succinic acid, oxalic acid, glutamic acid, citric acid, malic acid and fumaric acid). Taking into account the exchange reaction, chelation and acidification

these bacteria solubilizes the insoluble phosphorus for plants. From soil, different species of *Pseudomonas* and *Bacillus* have been isolated which exhibit the P-solubilising attributes (Mishra *et al.*, 2014). PSB can applied in all vegetables through seed treatment, soil application or seedling dip. Plants with limited root systems would be the most benefitted by PSB application (Abd El-Lattief, 2016).

Vesicular Arbuscular Mycorrhiza (VAM)

VAM fungi are inter-cellular and obligatory endosymbionts that have a beneficial relationship with plant roots since it extends and contaminates within the root zone. The root system transports nutrients to fungi and instead fungi tend to sustain plant roots with water and nutrients. Root length can expand through fungal hyphae and hyphae extend around 100 times in soils and enables plants to accumulate several nutrients. VAM fungi improve seedling tolerance to high temperature, drought and insect pest attack.

Factors limiting the use of biofertilizers

Lack of awareness among farmers.

Biofertilizers are plant specific i.e. one biofertilizer which works on one crop does not help in another crop.

They have short shelf-life as compared to chemical fertilizer so the major problem is storage for long term.

Unavailability of carrier material for specific biofertilizer.

Biofertilizers requirement is more to fulfil the need of nutrient required by the plant.

Future prospects

Realizing the importance of biofertilizers and their implementation in modern agriculture is a must. Biofertilizers help in improving the productivity and the fertility of soil so more

food will produce to feed burgeoning population. Biofertilizers will gradually help soil to regain its fertility for long term health. Biofertilizers application will reduce the use of chemical fertilizers and thus it reduces the additional cost of farmers. They are ecofriendly in nature and reduce the environmental pollution. Biofertilizers are only at the starting phase still need more efforts to bring changes in modern agriculture. Microbial extraction, their colonization, production, marketing, application, good knowledge among farmers etc. are necessary for more and more utilization of biofertilizers in modern agriculture aiming at the reduction of chemical fertilizer application in the field for high productivity. More studies in the field of plant and microbes interaction are required so that more efficient technology is used to get more production without disturbing the environment. Biofertilizers like *Azotobacter*, *Azospirillum*, *Phosphobacter*, *Rhodobacter* etc. can help plant to survive in stress conditions and to perform well. The application of biofertilizers not only will benefit the agricultural ecosystem but it also contributing to a holistic and sustainable environment.

In conclusion the modern agriculture, the excessive use of chemical fertilizers and pesticides is disturbing the sustainability of our agricultural land. These chemicals are becoming threat to human health because of consumption of chemically produced food by humans resulting dreadful diseases. These chemicals also have atrocious impacts on air, water and soil, thus disturbing the ecological balance. Use of biofertilizers is becoming a big challenge to ensure the food safety and environment protection. Now attention is shifting towards organic production of food because of the harmful effects of the chemical fertilizers. The application of bio-fertilizers having beneficial microbes is gaining

importance in promoting the crop productivity to a large extent and can help to solve the food need problem of increasing population of world. Soil erosion, water logging, accumulation of toxic elements are the main reasons which diminished the soil fertility in India. Biofertilizers are helpful in solving such kinds of problems and make the soil more productive as they are eco-friendly in nature.

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