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Role of bio-fertilizers towards sustainable agricultural development: A review

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Abstract

The worldwide increase in human population every year raises a major threat to the food security of the people as the land for agriculture is restricted and even drastic reduction with time. Therefore, it is essential that agricultural productivity should be enhanced significantly within the next few decades to meet the large demand of food by emerging population. Not to point out too much dependence on chemical fertilizers for more crop productions certainly damages both ecosystem and human health with great severity. Bio-fertilizers are one of the greatest nature gifts of our agricultural science as a replacement to chemical fertilizers. Bio-fertilizer contains microorganisms which encourage the adequate supply of nutrients to the host plants and ensure their proper development of growth, development and regulation in their physiology. Living microorganisms are used in the preparation of bio-fertilizers which have specific functions to augment plant growth and reproduction. Bio-fertilizers being essential components of sustainable farming play vital role in maintaining long term soil fertility and sustainability of crop production.

Keywords: Biofertlizers, Microorganisms soil fertility, crop production

1. Introduction

The World population is now 7.6 Billion and India alone contains 1.35 Billion people, which is increasing day by day. This imparts pressure on the agricultural lands and other resources which are needed for food of this huge population. According to 15th Census of India in 2011, the population decadal growth of 17.64% was observed of which around 68.84% is rural population. This growing human population demands conventional agriculture to meet its needs of food which makes farmers to depend on usage of chemical fertilizers and pesticides for increased productivity (Santos *et al*. 2012) [49] The chemical fertilizers are made in industries with known composition of various nutrients like nitrogen, phosphorus, potassium, sulfur *etc*. Harmful effects of usage of such fertilizers include weakening of roots of plants, increase of disease incidence, soil acidification (Chun-Li *et al*. 2014) [11] and eutrophication of ground water and other water bodies (Youssef and Eissa, 2014) [64] Nutrients like nitrates leach to ground water and cause Blue Baby Syndrome also called 'acquired methemoglobinemia' (Knobeloch, *et al*. 2000). Such chemicals will have a great impact on the future generations. In this regard, eco-friendly approaches are gaining popularity with a view of bio-safety of which Biofertilizers play a major role in sustainable agriculture. The global market for biofertilizers is expected to exceed a market worth of USD 10.2 billion by 2018. European and Latin American countries are the leading consumers of biofertilizers, due to the stringent regulations imposed on the usage of chemical fertilizers, eventually getting replaced bybiofertilizers (Raja, 2013) [45].

Biofertilizers may be defined as "substances which contain living microorganisms that colonize the rhizosphere or the interior of the plants and promote growth by increasing the supply or availability of primary nutrients to the target crops, when applied to soils, seeds or plant surfaces". (Mazid *et al*. 2011) [35] They colonize in rhizosphere accompanying interior of the plant and stimulates growth by increasing the accessibility and uptake of mineral nutrients to the host plant (Malusa *et al*. 2012) [32] They are the preparations containing cells of microorganisms which may be N fixers, P solubilizers, S-oxidisers or organic matter decomposers. They are "eco-friendly" agro-input of organic origin and work on conversion of unavailable essential elements to available form through their routine metabolic activities. (Vessey, 2003) [61] Plant utilizes the only 10% - 40% of applied nutrients, the rest of 60-90% is lost in the form of immobilization, leaching, volatilization *etc*. Biofertilizers help in slow and continuous release of nutrients by their metabolism and thus form an important component of Integrated Nutrient. Management (INM) system to sustaining agricultural productivity and a healthy environment (Adesemoye and Kloepper, 2009) [1]

Table 1: Different groups of biofertilizers **(**Barman *et al*. 2017) [5]

Sl. No.	Groups	Examples
1. Nitrogen (N_2) fixing Bio fertilizers		
i.	Free living	Azotobacter, Clostridium, Anabaena, Nostoc
ii.	Bio Symbiotic	*Rhizobium, Frankia, Anabaena azollae*
iii.	Associative-Symbiotic	*Azospirillum*
2. P-Solubilizing Bio fertilizers		
i.	Bacteria	*Bacillus subtilis* *Pseudomonas striata*
ii.	Fungi	*Penicillium* sp., *Aspergillus awamori*
3. P-Mobilizing Bio fertilizers		
i.	Arbuscular Mycorrhizae	*Glomus*sp., *Gigaspora*sp.
ii.	Ectomycorrhiza	*Laccarai sp., Pisolithus sp., Boletus sp., Amanita sp.*
iii.	Ericoid Mycorrizha	*Pezizellaericae*
4. K- solubilizers		
i.	Bacteria	*Frateuria aurantia*
5. Micronutrient solubilizers (Silicate and Zinc soloubilzers)		
i.	Bacteria	*Bacillus* sp.
6. Plant Growth Promoting Rhizobacteria		
i.	Bacteria	*Pseudomonas fluorescens*

1.1 Role of Bio fertilizers

Biofertilizers show plant growth promoting qualities and increase the yield by various mechanisms like nitrogen fixation, P-solubilization, P-mobilization, K solubilization, micronutrient solubilization, plant growth promotion, preventing the depletion of the soil organic matter (Jeyabal and Kupuswamy, 2001) [24] and maintenance of the natural habitat of the soil.

Table 2: Recommendations of different Bio-fertilizer for various crops

Biofertlizer	Recommended Crops
Rizobium	Pluses, Oilseeds, Fodders
Azospirillum	Rice, Wheat, millets, maize, sorghum, sugarcane
Azotobactor	Rice, Wheat, millets, other cereals, cotton, vegetables, sunflower, mustered, flowers
Azolla	Submerged rice with maximum temperature
Blue Green algae	Submerged rice
PSM	All crops

***Source*:** FAI, 2006-07

1.1.1 Nitrogen fixation

Certain bacteria and algae elemental, atmospheric nitrogen into plant usable forms like ammonical and nitrate forms. This process is called Biological nitrogen fixation (BNF) (Gothwal *et al*. 2007) [18] This mechanism is exploited by utilizing such microorganisms in the form of potent biofertilizers which act as a substitute for chemical fertilizers, help in maintenance of soil nitrogen reserves and give large crop yields (Peoples and Craswell, 1992) [41] They can be grouped into free-living bacteria like *Azotobacter*, associative like *Azospirillium*, cyanobacteria and symbiotic organisms like *Rhizobium*, *Frankia*and *Azolla* (Gupta, 2004) [20] Symbiotic associations are the maximum contributors for BNF in nature (Fig. 1). Apart from Azotobacter, many other genera fix nitrogen. Latt and others (2018) isolated and identified six free living N-fixing bacteria belonging to the genera *Azotobacter, Alcaligenes* and *Azomonas* by16s rRNA sequencing.
Certain genera are discussed here.

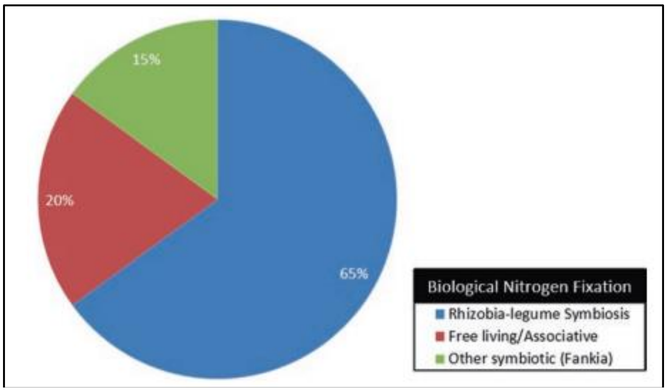

Fig 1: Biological nitrogen fixation (Modified from Bouizgarne *et al*. 2015) [8].

1.1.1.1 *Azotobacter*

Azotobacter is an aerobic, gram-negative, heterotrophic, free living nitrogen fixing bacteria. With the capacity of independent BNF, it has been found that some *Azotobacter* species are in association with some crops like cereals (Martyniuk and Martyniuk, 2003). [34] They are generally present in natural and alkaline soil occurring more in arable soils. The genus Azotobacter comprises of different species: *A. chroococcum, A. vinelandii, A. beijerinckii, A. paspali, A. armeniacus, A. nigricans and A. salinestri* (Gothandapani *et al*. 2017) [17]. Azotobacter spp. are motile and mesophilic with capability of fixing on an average 20 kg N/ha/per year (Rawia *et al*. 2009). *Azotobacter* spp. as biofertilizer has other roles to play like synthesis of antibiotics, plant growth hormones (Pandey *et al*. 1989), [40] vitamins, exopolysaccharides and pigment (Jimenez *et al*. 2011) [25] and antifungal activity (Sudhir *et al*. 1983). Table 3. Shown that the per cent increase in yield due to application of *Azotobacter* over chemical fertilizers in different crops.

Table 3: Effect of *azotobacter*on crop yield (Dudeja *et al*. 1981) [14]

Crop	Increase in yield over yields obtained with chemical fertilizers (%)	Crop	Increase in yield over yields obtained with bio fertilizers (%)
Wheat	8-10	Potato	16
Rice	5	Carrot	40
Maize	15-20	Cauliflower	2-24
Sorghum	15-20	Tomato	7-27
Other	13	Cotton	9-24

1.1.1.2 *Azospirillum*

Azospirillum are gram-negative, non-nodule-forming, aerobic, nitrogen-fixing bacteria belonging to the family Spirillaceae. They are associated with roots of mainly C_4 crops having dicarboxylic pathway (Hatch-Slack pathway) of photosynthesis because they grow and fix nitrogen on the organic salts of malic and aspartic acid (Mishra and Dash, 2014). [38] *Azospirillum* greatly affects root development and exudation (Trabelsi and Mhamdi, 2013). [59] It is capable of fixing 20–40 kg nitrogen under micro-aerobic conditions. Important species of *Azospirillum* are *A. lipoferum*, *A. brasilense*, *A. amazonense*, *A. halopreferans* and *A. trakense*. Steen houdt and Vanderleyden (2000) [60] reported that when *A. brasilense sp.* produced different phytohormones resulting in increase in maize growth and yield. Currently, *Azospirillum* is used as seed treatment just before sowing without the application in slurry with other products (i.e. micronutrients, fungicides, insecticides, polymers *etc*.). Apart from BNF, it has also been proved of phytohormone and/or siderophore production, phosphate solubilization (Puente *et al*. 2004), () biocontrol activity (Bashan and Bashan, 2010) [6] and protection of plants against stress like soil salinity or toxic compounds (Creus *et al*. 1997) [12].

1.1.1.3 *Rhizobium*

Rhizobium is an example of symbiotic association colonizing legume roots and fixes the atmospheric nitrogen. Bacteria obtain products of photosynthesis as energy source from plant and fix nitrogen from the air for their host. The morphology and physiology of *Rhizobium*at free-living condition is different from bacteroid of nodules. They are the most efficient biofertilizer for legumes as far as the quantity of nitrogen fixation is concerned (Jehangir *et al*. 2017) [23] Cross-inoculation plays an important role in host specificity of rhizobium and host (Table 4). The *nod*, *nif* and *fix* genes control the nodulation and nitrogen fixation by the bacterium.

Table 4: Major inoculation groups with inoculant and host plants **(**Ponmurugan and Gopi, 2006) [42]

Cross inoculation Group	***Rhizobium* species**	**Host Legume**
Pea group	*R. leguminosarum*	Pea, sweet pea
Alfalfa group	*R. meliloti*	Sweet clover
Clover group	*R. trifoli*	Clover / berseem
Bean group	*R. phaseoli*	All beans
Soybean group	*Bradyrhizobium japonicum*	Lupins
Cowpea group	*Rhizobium* sp.	Cowpea, arhar, urd, moong and groundnut

1.1.1.4 Cyanobacteria

Cyanobacteria (Blue Green Alga, BGA) are prokaryotes responsiblefor water blooms in stagnant waterbodies. They are also found in snow and hot springs. They are photosynthetic and can fix atmospheric nitrogen. They also promote plant growth by producing auxins (indole acetic acid), gibberllic acid and can fix around 20– 30 kg N/ha in submerged rice fields. Under low land rice conditions, application of BGA + Azospirillum proved beneficial in improving LAI and all yield attributes (Mishra *et al*. 2013) [38] Examples of certain BGA are *Nostoc, Anabaena, Cylindrospermum, Gloetrichia Tolypothrix, Aulosira* and *Aphanotheca*. Application of BGA can enhance rice productivity from 15% to 38%. Beneficial effects of cyanobacterial inoculation were also reported on a number of other crops such as barley, oats, tomato, radish, cotton, sugarcane, maize, chilli and lettuce (Thajuddin and Subramanian, 2005) [58]

1.1.1.5 *Azolla*

*Azolla*is a biofertilizer used in rice cultivation extensively. It is an aquatic fern sheltering a cyanobacteria (*Anabaena azollae*). It is an example for symbiotic association. Azolla as biofertilizer may be used as a partial substitute for synthetic fertilizer N due to its sustainable supplementation of N to rice crops and the associated improvement of soil fertility. Nitrogen fixing potential of Azolla varies between 30 and 50 kg N ha^{-1} which designates Azolla as an important biological N source for agriculture and animal industry (Yao *et al*., 2018). [63] Gupta (2004) stated that application of Azollabrought an impressive increase in rice yield by 0.5 -2 t /ha. Sundaravarathan and Kannaiyan (2002) [57] showed that application of *Azolla microphylla* @ 15 t/ha increased grain yield by 29.2%.

1.1.1.6 *Gluconacetobacter diazotrophicus*

G*luconacetobacter diazotrophicus* is a nitrogen fixing bacterium originally found in monocotyledon sugarcane plants in which the bacterium actively fixes atmosphere nitrogen and provides significant amounts of nitrogen to plants. This bacterium mainly colonizes intercellular spaces within the roots and stems of plants and does not require the formation of the complex root organ like nodule. The bacterium is less plant/crop specific and indeed *G. diazotrophicus* has been found in a number of unrelated plant species. Importantly, as the bacterium was of monocot plant origin, there exists a possibility that the nitrogen fixation feature of the bacterium may be used in many other monocot crops.

1.1.2 P-solubilization

Most of the phosphorus is insoluble and hence remain unavailable to promote plant growth as plants absorb it only in two soluble forms-monobasic and dibasic. Unavailable P is present in the form of organic matter (phosphodiesters, inositol phosphate, apatite). Immobilization of soluble inorganic P fertilizers makes it unavailable for the plants. Phosphorus solubilizing bacteria (PSB) in soil like *Pseudomonas putida* and *Bacillus megaterium* and fungi like *Aspergillus* and *Penicillium* solubilize bound phosphate and fixed phosphorus to make it available for plants. 20–25% phosphorus requirement of plants is fulfilled by PSB and PSF. Thus, its use in agriculture not only reduces manufacturing costs of phosphate fertilizers but also mobilizes insoluble fertilizers in soil (Chang and Yang 2009). [9] Among the P-solubilizing microbial population in soil, bacteria (PSB) constitute 1 to 50%, while fungi (PSF) are only 0.1 to 0.5% in P solubilization potential (Chen *et al.* 2006). [10] Some actinomycetes are known for P- solubilization and are gaining popularity due to added advantages like capability of surviving in extreme environments like drought, production of antibiotics and phytohormone-like compounds. Hamdali *et al*. (2008) had statedthat approximately 20% of actinomycetes can solubilize P, including those in the common genera *Streptomyces* and *Micromono spora*. Some of the P-solubilizing microorganisms are discussed here.

1.1.2.1 Bacteria

Also called as "Phosphobacterium", *Bacillus megaterium* are Gram +ve, rod shaped bacteria reported to enhance mineral phosphorus (P) solubilisation (Lach *et al*. 1990). [30] The proposed mechanisms of P- solubilisation are by the release of organic and inorganic acids, excretion of protons that accompanies to theNH4$^+$ assimilation and due to therelease of phosphatase enzymes that mineralize organic P compounds (Stevenson, 1986). It also has the Zn, K, Fe and Mn solubilizing potential (Amalraj *et al*. 2012). It also enhances root proliferation due to release of growth-promoting hormones. It supplies P to the plants to the extent of 10 to 15 kg / ha, saving up to 50% over the cost of P chemical fertilizer inputs. Modes of application of this biofertilizer can be seed treatment, root dipping for transplanting, soil application and drip system. *Pseudomonaserwinia* and *P. chlororaphis*were able to solubilize P sources strongly and HPCL analyses showed several organic acids (Diriba Muleta *et al*. 2013) [13]

1.1.2.2 *Fungi*

Fungi like *Aspergillus* and *Pencillium* are potent P-solubilizing fungi (Vassilev *et al*. 2007; Oleivera *et al*. 2009). Gunes *et al*. (2009) [19] reported that commonly found arable soil fungi such as *Penicillium* sp., *Mucor* sp. and *Aspergillus* sp. have shown to increase plant growth by 5–20%. Kucey and Paul (1982) [29] recognized and reported the ability of several species of *Aspergillus* and *Penicillium* to solubilize insoluble phosphate. *A. niger* increased dry biomass of chickpea plants by 22–33% compared with non-inoculated control (Kapri and Tewari 2010). [26] PSF gave significantly higher (by 12.6%) grain yield than the control alone by increasing solubility of the unavailable P forms in soil. The study showed potential of using PSF (*Penicillium bilaii*) as bio-inoculants along with 50% of recommended P fertilizer dose that produced wheat yield similar to 100% P when no PSF was used (Ram *et al*. 2015). [46]

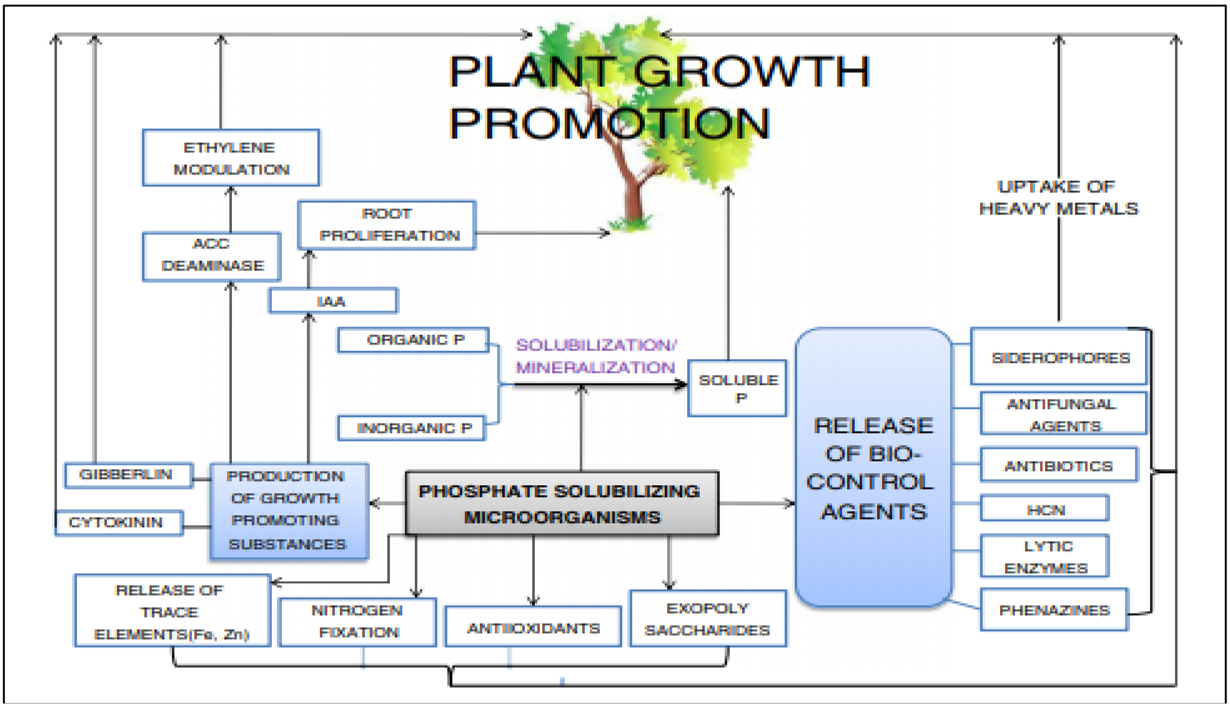

Fig 2: Possible Mechanisms involved in plant growth promotion by PSM (Sharma *et al*. 2013) [51]

1.1.3 P-mobilizers

Certain microorganisms increase the uptake of P by mobilizing it from a P rich environment to plants instead of solubilizing P. Such microorganisms are called P- mobilizers. While arbuscularmycorrhizae fungi are known to increase fertilizer P use efficiency by increasing its mobility in soil and reducing fixation, these fungi have only an advantage at low P availability in soil (Ghorbanian *et al*. 2012) [16] *Pseudomonas letiola* on mobilization of P and growth of Ligol (young apple trees) grown in a pot, significantly increased the total shoot length and solubilised insoluble P compounds (Ewa *et al*. 2013) [15]. While Mycorrhizaare the major contributors for P-mobilization, they are being discussed.

1.1.3.1 Mycorrhiza

Mycorrhiza is group of fungi in symbiotic association with plant roots in rhizosphere. They belong to phylum *Glomeromycota* and greatly enhance the uptake of relatively immobile soil nutrients, especially P, by plant root systems (Bolan, 1991) [7] They are known for exploitation of a larger soil volume and thus increase P uptake by the extensive external hyphae, Jakobsen *et al*. (1992) [22] showed that different amounts of external mycelium produced by AM Fungi supplied different amounts of P to the host plant. In a study conducted by Yao and others (2001), [63] the results proved that AMF gave increased contact with phosphates in the soil by mobilizing phosphates under conditions of low P availablility and also solubilized P by production of organic acids. The small diameter of hyphae (2-20 μm) makes P acquisition easier as compared to that of new roots formation. Transfer from fungus to plant occurs principally at the arbuscule interface, but likely also at the site of hyphal coils (Karandashov *et al*. 2004) [27].

1.1.4 K Solubilizing Bacteria

Potassium (K) is considered as an essential nutrient for plants. Naturally, soils harbour K in larger amounts than any other nutrients but in unavailable form for plant uptake. Only 1 to 2% of this element is available to plants (Sparks and Huang, 1985) [55] Many groups of microorganisms like bacteria, fungi and actinomycetes are reported for solubilization of soil K of which, bacteria solubilize by production of inorganic and organic acids, acidolysis, polysaccharides, complexolysis, chelation, polysaccharides, and exchange reactions (Archana *et al*. 2013; Meena *et al*. 2015). [4] In rice fields, *Bacillus licheniformis* and *Pseudomonas azotoformans* had higher K-solubilizing ability than other isolated rhizobacteria (Saha *et al*. 2016). [48] Prajapati and Modi (2016) [43] reported that K and chlorophyll content in cucumber was remarkably enhanced by *Enterobacter hormoechei*, which had the ability of K- solubilization. (Subhashini, 2015) [55] observed that soluble potassium and inoculated with strain *Frateuria* *aurantia* shown higher potassium content of the leaf was increased by 39%. K solubilization is carried out by a large number of bacteria such as *Bacillus mucilaginosus, B. edaphicus, B. circulans, Pseudomonas, Burkholderia, Acidithiobacillus ferrooxidans*, and *Paenibacillus* spp. The production and management of biological fertilizers containing KSB can be an effective alternative to chemical fertilizers.

1.1.5 Plant Growth Promoting Rhizobacteria (PGPR)

The group of bacteria that colonize roots or rhizosphere soil and beneficial to crops are referred to as plant growth promoting rhizobacteria (PGPR). They are also called as microbial pesticides e.g. *Bacillus spp.* and *Pseudomonas fluorescence.* PGPR inoculants currently commercialized that seem to promote growth through at least one mechanism; suppression of plant disease (termed Bio-protectants), improved nutrient acquisition (termed Bio-fertilizers), or phyto-hormone production (termed Bio-stimulants). Species of *Pseudomonas* and *Bacillus* can produce as yet not well characterized phyto-hormones or growth regulators that cause crops to have greater amounts of fine roots which have the effect of increasing the absorptive surface of plant roots for uptake of water and nutrients. These PGPR are referred to as Bio-stimulants and the phyto-hormones they produce include indole-acetic acid, cytokinins, gibberellins and inhibitors of ethylene production Sudheer *et al*., 1983. [56]. Rhizobacteria induce resistance through the salicylic acid-dependent SAR pathway, or require jasmonic acid and ethylene perception from the plant for ISR. Rhizobacteria belonging to the genera *Pseudomonas* and *Bacillus* are well known for their antagonistic effects and their ability to trigger ISR. Resistance-inducing and antagonistic rhizobacteria might be useful in formulating new inoculants with combinations of different mechanisms of action, leading to a more efficient use for biocontrol strategies to improve cropping systems. Olivera *et al*., 2009. [39]

1.1.6 Zinc solubilizers

The zinc can be solubilized by microorganism's viz., *B. subtilis, Thiobacillus thioxidans* and *Saccharomyces* sp. These microorganisms can be used as bio-fertilizers for solubilization of fixed micronutrients like zinc. The results have shown that a Bacillus sp. (Zn solubilizing bacteria) can be used as bio-fertilizer for zinc or in soils where native zinc is higher or in conjunction with insoluble cheaper zinc compounds like zinc oxide (ZnO), zinc carbonate (ZnCO3) and zinc sulphide (ZnS) instead of costly zinc sulphate. (Mahdi *et al*. 2010) [33].

1.2 Rejuvenation Strategies of Biofertilizers for Sustainable Agriculture Economic Development

The following strategies can be used for rejuvenating sustainable agriculture economic development: •Identifying and selecting efficient locations, crop, soil, and specific strains for N-fixing, P, Zn solubilizing and absorbing (mycorrhizal) to suit different agro climatic conditions. Strain improvement through biotechnological methods. Exchanging cultures between countries of similar climatic conditions and evaluating their performance for a better strain for particular crop as well as checking the activity of cultures during storage to avoid natural mutants • Developing suitable alternative formulations viz., liquid inoculants / granular formulations for all bioinoculants, to carrier based inoculants, standardizing the media, method of inoculation etc., for the new formulations. •Employing microbiologists in production units to monitor the production • Developing cold storage facilities in production centers •Technical training on the production and quality control to the producers and rendering technical advice and projects to manufacturers • Organizational training to the extension workers and farmers to popularize the technology •Disseminating information through mass media, publications and bulletins. (Gupta and Sen, 2013)

1.3 Conclusion and future prospects

As a boon for farmers, Bio-fertilizers being essential components of sustainable farming play vital role in maintaining long term soil fertility and sustainability. Bio-fertilizers would be the viable option for farmers to increase productivity per unit area in organic farming for an era of prosperity and clean environment. Currently there is a gap of ten million tons of plant nutrients between removal of crops and supply through chemical fertilizers. In context of both the cost and environmental impact of chemical fertilizers, excessive reliance on the chemical fertilizers is not viable strategy in long run because of the cost, both in domestic resources and foreign exchange, involved in setting up of fertilizer plants and sustaining the production. Biofertilizers are products that are likely to be commercially promising in the long run once adequate information becomes available to producers and farmers. The use of biofertilizers in India will not only have an impact on sustainable agriculture's economic development but it will also contribute to a sustainable ecosystem and the holistic well-being of the country.

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