ROLE OF PGPR AS MICROBIAL INOCULANTS IN IMPROVING FODDER CROPS PRODUCTIVITY AND QUALITY: A REVIEW

MEGHA VERMA[1], GULAB PANDOVE[2]* AND ANUREET KAUR[3]

[1]Department of Microbiology, Punjab Agricultural University, Ludhiana-141001 (Punjab), India
[2]School of Organic Farming, Punjab Agricultural University, Ludhiana-141001 (Punjab), India
[3]Punjab Agricultural University, Regional Research Station, Bathinda-151001 (Punjab), India
**(e-mail : gpandove@pau.edu)*

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SUMMARY

Global increase in human population poses a major threat to livestock as the area for the cultivation of fodder crop is shrinking over the time. In addition, biotic and abiotic stresses are other main barriers to the fodder crop yield, quality and global food security for the livestock. For the higher productivity of livestock products such as milk, meat and wool, it is necessary to provide green fodder in ample quantity and quality. It leads to indiscriminate use of agrochemicals for enhanced productivity. Plant Growth Promoting rhizobacteria (PGPR)/microbial inoculants can play pertinent role to counteract the detrimental environmental impacts exerted by chemical fertilizers and other agrochemicals. PGPR are the living micro-organisms which colonizes the rhizosphere and promotes growth by increasing the availability and supply of nutrients by multifarious methods such as biological nitrogen fixation, siderophore production, phosphate solubilization and phytohormone production. Likewise, PGPR releases their metabolites directly or indirectly into the soil, thus improves the soil fertility and sustainability. The potential of PGPR has gained momentum during the last few decades as these can be used in integrated manner to reduce the use of chemical fertilizers and pesticides and thereby helping to fulfil the demand of fodder crop in environmental, economical and climate resilient manner. The aim of this review is to discuss the important mechanisms and deliberate the prospects of using PGPR in fodder production.

Key Words : Plant Growth-Promoting Rhizobacteria (PGPR), Microbial Inoculants, Livestock, Fodder, Integrated Nutrient Management.

India makes use of 4.9% cropped land for the cultivation of the green fodders and facing a deficit of 26% of dry fodder and 35.6% of green fodder (Jemimah *et al.,* 2015). It is not easy to amplify fodder production due to ever growing human pressure on land for the production of oilseeds, pulses and cereals.

Although, Chemical fertilizers (Inorganic sources) are most effective and quick in response on increasing the yield and growth of crops but excessive exposure would deteriorate soil health and induces environmental impacts. In addition, variability in the climate has high impact on fodder crops, livestock, fisheries and animal husbandry. The constituent and quality of green fodder changes and results in the reduction of livestock goods (Milk, Egg and Meat) due to climate change (IPCC, 2014).

The need of the hour is integrated nutrient management which is very essential for the sustainability and productivity of the soil fertility.

Microbial inoculants are now being used worldwide in agriculture for the different crops. These form an integral part of integrated plant nutrient supply system (IPNS) because they are ecofriendly, cost effective, enhance and promote plant growth and their development by various methods. Microbial inoculants are also known as biofertilizers. Biofertilizers (Microbial inoculants) may be defined as a substance containing micro-organisms which colonizes the rhizosphere or the interior of plant and promotes growth by increasing the supply and availability of nutrients (Abdel ghany *et al.,* 2013). The beneficial micro-organisms in the microbial inoculants are called Plant Growth Promoting Rhizobacteria (PGPR) or Plant Growth Promoting Bacteria (PGPB). These micro-organisms can be rhizospheric or endophytic depending upon the location.

Mechanisms of action of Microbial Inoculants

1. Biological Fixation of Nitrogen

The component which is essential for productivity and the growth of a plant is nitrogen. Although, 78% of nitrogen is available in the atmosphere but it is unable to utilize by the plant kingdom because of the lack of enzyme that aids to convert dinitrogen (highly stable triple bond Structure) into ammonia, nitrate and nitrite. There is conversion of atmospheric nitrogen to those forms which can be easily utilized by the plant through the process of Biological Fixation of Nitrogen (BFN) which involves the conversion of dinitrogen to NH_3 in the presence of complex enzyme nitrogenase (Kim and Rees, 1994). The Biological Nitrogen Fixation takes place with the help of microbes through a reductive process. The microbes involved in it are Actinomycetes, Eubacteria and algae specially the blue-green. It was first discovered by Beijerinck in 1901 (Wagner, 2012). A total of 70% of the nitrogen fixation occurs in the atmosphere through the biological means. In return for fixing the atmospheric nitrogen, the bacteria are benefitted with favourable environment for their growth and source of energy i.e. the photosynthate.

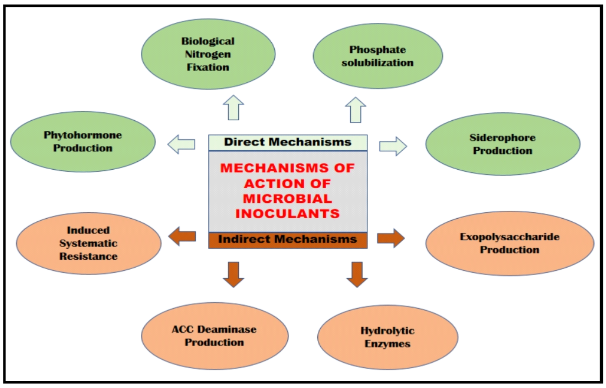

Fig. 1. Direct and Indirect mechanisms of action of Microbial Inoculants.

Azotobacter and *Azospirillium* sp. are potential biofertilizers that can improve the quality and yield of forage crops without any additional use of chemical fertilizers via the process of biological fixation of nitrogen (Mahdi *et al*., 2010). Wani *et al*., (2016) demonstrated that *Azotobacter* has been used as a biofertilizer for barley, oats and maize.

2. Phosphate solubilization

The second most vital element in the plant growth and development after nitrogen is phosphorous. It plays major roles in the various metabolic processes of plants which includes respiration, transfer of energy, photosynthesis, signal transduction, biosynthesis of macromolecules (Khan *et al*., 2010) and fixation of nitrogen in legumes (Saber *et al*., 2005). It is abundantly available in both inorganic and organic forms in the soil (Khan *et al*., 2009). However, it is a limiting factor for the growth of the plants as it is unavailable for root uptake (as it is available in the non-soluble forms) and the plants can take it in only in the forms of H_2PO_4 and $H_2PO_4^{2-}$ which are the soluble forms (Bhattacharyya and Jha 2012). The insoluble forms of Phosphorus includes inorganic mineral such as apatite or as organic form such as soil phytate (inositol phosphate), phosphotriesters and phosphomonoesters (Glick 2012). In general, the fertilizers of phosphates are used to increase the quantity of phosphorous in the soil, but only 30-35 % of the applied fertilizers of phosphates are taken up by the plant rest 65-70% changes to non-soluble form and cannot be solubilize by the plants. There are few microorganisms which has the capacity of solubilizing that non-soluble phosphate and makes it available to the plants in the soluble form. Microbes which help in the solubilization activity of phosphorus, are known as the Phosphate solubilizing microorganisms.

The Bacterial genera which solubilize the Phosphate are *Bacillus, Burkholderia, Rhizobium*, *Flavobacterium, Serratia, Pseudomonas, Azotobacter, Beijerinckia, Enterobacter, Erwinia*, *Microbacterium* etc. These microbes secrete the Phosphatase enzyme, acids which are organic in nature like citric, succinic, gluconic and oxalic acids (Kumari *et al.,* 2009), release proton and chelation takes place (Zaidi *et al.,* 2009b). The Phosphate Solubilizing Bacteria also stimulates the plant growth by increasing the efficiency of biological fixation of nitrogen and by enhancing the presence of other microelements (Zaidi *et al.,* 2009, Ahmad *et al.,* 2008).

3. Siderophore production

The essential microelement that affects various metabolic processes in Bacteria, fungi and plants is Iron. Siderophores are the low molecular compounds which are produced by various microorganisms (Microbial inoculants) under iron limiting conditions to satisfy the nutritional requirements of iron. These Siderophores have high affinity for Fe^{3+}, binds strongly with Fe^{3+} and is reduced to Fe^{2+} that is released into the cell via gated channels. When plant host incorporates these soluble Fe^{2+}, eventually total plant growth increases significantly (Mahanty *et al.,* 2017). Also, siderophore form complexes with heavy metals like Cu, Cd, Al, Pb and Zn that are of environmental concern. Iron is taken up by the plants with the help of various mechanisms from the bacterial siderophores (Neubauer *et al.,* 2000). Siderophore producing

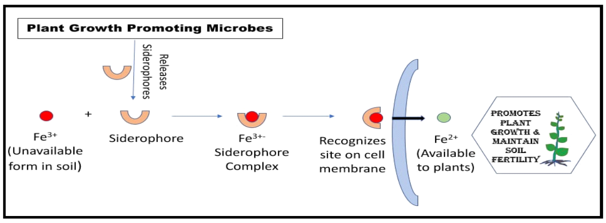

Fig. 2. Mechanism of Iron Sequestration through the production of Siderophore by Plant Growth Promoting Bacteria.

bacteria are *Bacillus, Agrobacterium, Rhodococcus, Aerobacter, Actbinomyctes, Streptomyces, Azotobacter, Arthrobacter, Nocardia, Mycobacterium, Pseudomonas, Enterobacter*, *Escherichia* etc.

4. ACC deaminase Activity

The hormone which is an important modulator required for the plant growth and development is ethylene (Khalid *et al.,* 2006) as well as it induces various physiologically changes in the plants. The level of ethylene increases under conditions of stress which are caused by the excessive water logging, pathogenicity, salinity, drought and heavy metals that results into defoliation and deteriorate of other cellular processes which affects crop performances and is responsible for reduction in the yield (Bhattacharyya and Jha, 2012).

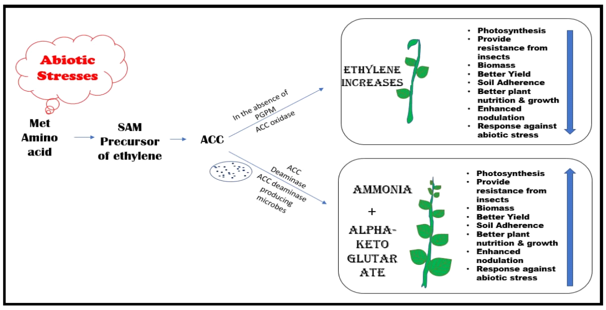

Fig. 3. Possible mechanism of ACC Deaminase reducing ethylene level to facilitate plant growth funded by PGPM.

Many microbial inoculants produces an enzyme called 1-amino-cyclopropane-1-carboxylic acid - deaminase. This enzyme breaks 1-amino-cyclopropane-1-carboxylic acid (ACC), which is a precursor of ethylene in plant under stress conditions (Glick, 2014). It leads to reduction in the ethylene level and relieves other stresses like resistance from polyaromatic hydrocarbons, radiations, high light density, insect predation, draft, flooding and effect of phytopathogenic microorganisms. (Mani and Kumar 2014, Glick 2012). ACC deaminase producing genera are *Burkholderia, Enterobacter, Serratia, Rhizobium, Achromobacter, Agrobacterium, Acinetobacter, Alcaligenes, Pseudomonas and Azospirillium* etc. Inoculation of these microorganisms induce perceptible effects on plants such as promotion of shoot growth, nutrient uptake, extensive root growth (Babalola, 2003), enhancement in rhizobial nodulation and mycorrhizal colonization in many crop species. (Glick, 2012, Nadeem *et al.,* 2009).

5. Phytohormones produced by the microbial inoculants

In PGPRs, the most important plant growth mechanism after Nitrogen fixation is the synthesis of phytohormones. Phytohormones (often regarded as plant regulators) are the signal molecules produced in extremely meagre quantity but able to regulate alot of processes especially the cellular one in plants. It also maintains abiotic stress and the interaction of plant-pathogen. Growth and development in plants occurs through phytohormones like Auxins (IAA), Gibberellins (GA), Ethylene (ETHY), Cytokinins (CKs), Abscissic Acid (ABA) and Brassinosteroids (BRs) which helps in the controlation of various physiological and biochemical processes in sessile plants (Iqbal et al 2014).

Role of Microbial Inoculants in improving growth parameters of fodder crops

India supports nearly 15 % of the world's livestock population with only 2.5 of the world's geographical area. India is the home for about 16% cattle and 5.5 % buffalo from 512.05 million livestock. However, animals facing 40-60% deficiency in energy and protein respectively (Sial and Aalam, 2008). The forage yield of our native varieties is insufficient to meet the demands of the livestock. Both the quality and quantity of feed mills must be raised to the required level. Major annual forage crops of our country are Oats, cowpea, barseem, maize, pearl millet, sorghum & lucerne (Garnett, 2010). Microbial inoculants can be a viable option to keep our production system running (Mahdi *et al.,* 2010). These microbial inoculants significantly increases the plant height (cm), germination count (m^{-2}), number of tillers (m^{-2}), number of leaves per plant, leaf area (cm^2) and leaf to stem ratio (Alori *et al.,* 2019). PGPR also promotes the growth of plants in terms of root and aerial parameters, yield, nutrient absorption and stress relief (Nantsiost *et al.,* 2019).

Saleem *et al.* (2015) conducted a field experiment in RCBD with three replicons with two integrated approaches. The first approach was of oat cultivators having four treatments *i.e.* V1(AVON), V2(S-2000), V3(S-2011), V4 (PD2L65) and second approach was of seed inoculation with three treatments viz S0 (control), S1 (*Azotobacter sp.*), S2 (*Azospirillium sp.*) and reported that *Azotobacter* and *Azospirillium sp.* with V3 oat cultivator significantly increases the plant height (cm), germination count (m^{-2}), no. of leaves per plant, no. of tillers (m^{-2}), leaf area (cm^2), green forage yield (85.2 t ha^{-1}), dry matter yield (14.0 t ha^{-1}).

Singh *et al.* (2016) demonstrated that co-inoculation of *Azotobacter chroococcum*, *Azospirillium lipoferum*, *Pseudomonas fluorescens*, *Acetobacter diazotrophicus* along with *Trichoderma viride* enhanced the plant height (163.54cm), dry weight (91.15g), stover yield (10.77t ha^{-1}) and grain yield (3.01 t ha^{-1}) of pearl millet over control.

Kaur *et al.* (2018) conducted a field experiment in RCBD and replicated thrice at two places (Bathinda and Ludhiana) with nine treatments in fodder sorghum. The result revealed that maximum growth and yield was observed with the application of RDF + *Burkholderia sp.* + *Azotobacter sp.* and percentage increase in the plant height, tillers per plant, leaves per plant, leaf/ root ratio, dry fodder yield and green fodder yield was 3.89, 9.6, 11.28, 11.2, 4.28, 4.11 and 4.72, 8.22, 12.44, 20.71, 7.37, 6.4 percent respectively at Bathinda and Ludhiana.

Aditi *et al.* (2019) conducted a field experiment with twelve treatment combinations consisting of two levels of bio-compost, two levels of biofertilizers and three levels of fertilizers in RBD with three replicons. Among these treatment combinations, 100% RDF was statistically at par with 75% RDF with biofertilizer and biocompost recorded the maximum yield of green and dry fodder, stem girth and plant height.

Haran and Thaher (2019) demonstrated that co-inoculation of PSB + *Azotobacter sp.* + *Pseudomonas flourescens* on maize plant enhances plant height (33.08%), dry weight of shoot (31.90%), weight of 100 grains (44.99%), grain weight in cop (62.06%), total grain yield (61.07%) and leaf surface area (36.94%) and Nitrogen, Potassium and Phosphorus concentration in the growth and grain with 12.8%, 156.5%, 31.5%, 272.3%, 75.1 % and 39.5 % over the uninoculated crop.

Kiran *et al.* (2020) performed a field experiment in RCBD with 13 treatments including control, 100% RDF, Microbial Consortia + 100% RDF, Microbial Consortia + 100% RDF + humic acid (0.25, 0.50%), Microbial consortia+ Recommended package of practices+ Algal extract (10, 20%) and these treatments were also repeated with 75% RPP. Under the treatment of 100% RPP with microbial consortia and algal extract (T_6 and T_7) gave higher plant height, leaf number, branch leaf area, SPAD value and dry matter accumulation in maize was observed which may be due to the presence of plant growth regulators.

Further Li *et al.* (2020) demonstrated that PGPR (*Providencia rettgeri, Advenella incenata, Serratia plymuthica and Acinetobacter calcoaceticus*) had significant impact on enzyme activity, nutrient content and on growth and physiology of oat, alfafa and cucumber.

Role of Microbial Inoculants in improving Quality Parameters of various fodder crops

Intensive agricultural practices are used worldwide to meet the food needs of the growing population (Sanchez-Santillan *et al.,* 2020). Due to rapid development of economy the demand for livestock products has also grown rapidly. Fodder is an important resource for the normal rumen development of ruminants and high quality feed is a key resource for improving animal performances (Kim *et al.,* 2012).

Ramya (2019) carried out a field experiment in random complete design with a total of eleven treatment combination of liquid microbial inoculants (*Bradyrhizobium* sp., *Burkholderia seminalis* and *Burkholderia* sp.) with 75% and 100% RDF and observed that the treatment T_{10} (75% RDF + *Burkholderia seminalis* + *Burkholderia* sp.) improved the quality parameters viz Acid detergent fibre (ADF), Neutral Detergent fibre (NDF), In vitro dry matter digestibility (IVDMD), total sugars and phenols in the forage cowpea.

Mishra *et al.* (2008) found that dual inoculation of *Rhizobium* (Nitrogen fixer) and *Arbuscular mycorrhizal fungi* improves the 91-92% nodulation, 89-91% biomass and 46-47% fodder production in terms of digestible nutrients (DN) and crude protein (CP) by decrease of Acid detergent fibre (ADF) and Neutral detergent fibre (NDF) in stylosanthes forage and concluded that multiple strains improvens the production and quality of fodder.

Shabbir *et al.* (2013) reported that application of phosphorus (0, 30, 60 and 90 kg ha^{-1}) along with Phosphate solubilizing microbe in oat resulted in improved growth, yield and quality parameters.

Lee *et al.* (2014) evaluated that the inoculation of *Lactobacillus Plantarum* increases the cutting height of forage during harvest and can be used to obtain the fibrous protein, thereby increasing the digestibility of dry matter (IVD) of silage barley.

Role of Microbial Inoculants in integrated nutrient management of fodder crops

According to World Health Organization (WHO) there are three million cases of the agrochemical poisoning in the developing countries. The long term, large-scale and indiscriminate use of agrochemicals will have adverse effects on agricultural sustainability, soil biodiversity and food security as well as prolonged negative effects on the health of animals and humans. Most agrochemicals can impair the soil microbial functions and their biochemical processes. Currently there is a need for innovative, high quality and demand oriented soil science research in the developing countries to promote eco-friendly research by creating a supportive and trustworthy work atmosphere (Meena *et al.,* 2020).

Among the various Agrotechniques, Integrated Nutrient Management (INM) has proven to be the best opinion to increase the production of better quality forage per unit area by maintaining soil fertility and productivity with limited soil resources. Integrated Nutrient Management is the amalgamation of mineral fertilizers with organic resources such as manure, compost, biofertilizers, green manures etc (Antil, 2012). Liquid biofertilizers is a special formulation containing high content of desired micro-organisms with high shelf life and zero chances of contamination. (Mazid and Khan, 2014) These formulations provide good field performance characteristics which uses low cost materials and easily manufactured by small manufacturers.

Divya *et al.,* (2017) reported that the application of 75% RDF and Biofertilizers (@5kg ha^{-1}) resulted in higher yield attributes, grain yield and stover yield of Pearl Millet as compared to 75% RDF + 25% N and 100 % RDF.

Devi et al (2014) reported that the combined application of *Azotobacter chroococcum* with 80 kg N ha^{-1} improves the yield attributes like grain yield, straw yield and green fodder yield and economic returns of oats. Similarly, Jena *et al* (2019) found that *Azotobacter* + PSB + 75% N, P and K and micronutrient Zn increases the crude protein content than both control and recommended dose of fertilizers applied treatment.

Bilal *et al.* (2017) carried out a experiment with four levels of nitrogen (0, 40, 80, 120 kg ha^{-1}) along with mixture of *Azospirillium* + *Azotobacter sp.* and found that inoculation mixture produces 6.58%, 2.58%, 10.26%, 9.58%, 16.94%, 14.02%, 66.18%, 17.59 % and 33.81% more number of tillers, leaf to stem ratio, mineral matter content, plant height, dry matter yield, crude protein, total digestible crude protein, crude fibre and crude protein yield of oat. The interaction effect showed that the inoculation without adding nitrogen produced 6.87% and 19.16% more crude protein and dry matter respectively. In addition, the application of 80 kg ha^{-1} nitrogen and *Azotobacter* + *Azospirillium* species appears to be most efficient for acquiring maximum yield of forage oat.

Narkhede *et al.* (2017) conducted a field experiment in soyabean-onion with seven nutrient treatments. Out of the seven treatments, five had organic resources, one had 100% fertilizer and another one had integrated nutrient management (50% Inorganic and 50% organic fertilizers) and found higher onion bulb yield (23.9 t ha^{-1}), soyabean grain yield (2.67t ha^{-1}), economic efficiency (510.07/ha/day) and production efficiency (110.0 kg/ha/day) by Integrated Nutrient management.

Thus, the application of microbial inoculants not only optimizes the use of chemical fertilizers but also improves the cost benefits ratio.

Conclusion and Future Prospective

The burgeoning population increases pressure on livestock to meet the animal product demand (milk, meat, wool etc). For the increase in livestock productivity, it is essential to provide green fodder.

However, the indiscriminate use of pesticides and chemical fertilizers severely damages the ecosystem therefore it is necessary to adopt more sustainable pest and nutrition management methods like microbial inoculants or Biofertilizers. They may also help to mitigate the effects of climate and other related abiotic and biotic stresses.

The use of microbial inoculants not only improves growth productivity and yield of fodder crops but also the quality. Moreover, long term use of microbial inoculants can improve soil fertility and sustainability.

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