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Possibilities and potential of Rhizobial inoculants in organic production of fenugreek in arid and semiarid regions of Rajasthan

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ABSTRACT

Arid and semiarid regions are known for erratic rainfall and low productive soils. Fenugreek is one of the important crop of these areas. These regions are having immense possibilities of organic production of fenugreek. The possibilities can be explored through use of specific, effective and efficient rhizobial inoculants for the fenugreek production. In the market commercial cross inoculated rhizobial inoculants are available which are not specific to fenugreek or a particular variety. Also, the growing awareness of health globally has led to demand of organically produced seed spices as there are free from synthetic chemical residues and health hazards and also bear high quality ecofriendly. The use of *Rhizobium* has proved beneficial in improving the soil nitrogen and getting higher yield. However, the use of effective *Rhizobium* cultures as inoculums for organic production of fenugreek crop has not gained much momentum due to lack of scientific information and awareness among the growers therefore, it is essential to understand interaction of *Rhizobium* to specific variations of fenugreek, bio-ecology and their association as well. Screening of effective *Rhizobium* strains for adverse environmental conditions (Drought, salinity and salt, temperature etc) prevailing in Rajasthan, may require a great attention on rhizobia research. This paper reviews the potential of fenugreek rhizobia symbiosis and discusses future potential of this crop for use in arid and semiarid regions of Rajasthan.

Keywords: Organic fenugreek, Potential, Rhizobial inoculants.

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INTRODUCTION

Arid and semiarid regions are known for stressed environment mainly because of erratic rainfall, low fertility of soil, resulted in low crop productivity with high uncertainty. These conditions are not supported for fenugreek culture. This crop has great contribution in the economy of the region and in export market of the country as well.

During the past four decades we have witnessed the doubling of the human population and a concurrent doubling of food production (43). Plant nutrition has played a key role in this dramatic increase in demand for and supply of food. Increases in crop production have been made possible through the use of commercial man-made fertilizers. With respect to seed spices, the growing awareness of health globally has led to demand of organically produced seed spices as they are free from synthetic chemical residues and health hazards. Fenugreek (*Trigonella foenum-graecum*) being a legume, fixes about 283 kg nitrogen per hectare per year from the atmosphere into the soil, thus has very good advantages in symbiotic interaction with a group of root nodulating bacteria collectively known as "Rhizobia". The use of *Rhizobium* has proved beneficial in improvement of the soil quality and getting higher seed yield, since field inoculation with *Rhizobium meliloti* bacterium is very useful for fenugreek cultivation (1). However, the use of effective *Rhizobium* cultures as inoculums for organic production of fenugreek crop has not gained much momentum due to lack of scientific information, although organic produce are of high quality, safe, nutritive and eco-friendly. Uses of appropriate agronomic practices are necessary to maximize forage and seed production of the fenugreek crop cultivar in arid and semi arid conditions of Rajasthan.

In the article, possibilities and potential of *Rhizobium* inoculants in organic production of fenugreek have been discussed.

Distribution of Fenugreek

Fenugreek, (*Trigonella foenum-graecum* L), is an annual legume crop mainly grown for use as a vegetable and spice in many parts of the world. Land races and species of *Trigonella* have been found in the continents of Asia, Europe, Africa and Australia. Fenugreek is also cultivated in parts of Europe, Northern Africa, West and South Asia, North and South America and Australia (27). The species name *"foenum-graecum"* means "Greek hay" indicating its use as a forage crop in the past. Carbonized fenugreek seed recovered from Punjab, India indicate its use in trade as far back as 2000-1700 B.C.

Importance of symbiotic association

Nitrogen is commonly the most limiting element in agricultural production and the most of expensive to purchase as fertilizer. Atmospheric air is rich with nitrogen (78%), but unfortunately in stable gaseous form, which can not be utilized directly by the plants. Hence, there is need to convert this atmospheric nitrogen to available form for plants. Biological Nitrogen Fixation (BNF) is a fascinating biological phenomenon, which involves some legumes, whether grown as pulses for seed, as pasture in agro-forestry or in natural ecosystems (14). This process involves a specialized and intricately evolved interaction between rhizobia and higher plants, which results in the formation of specific organs, called Nodules (35). Nodulation is very common on legume roots but in some cases nodules are also formed on the stems, where they harness and convert atmospheric nitrogen (6, 7, 22) into a form such as ammonium, nitrates and ammonia (13, 41, 45) that can be easily assimilated by the plants (14, 30). This process occurs by involving the enzyme complex i.e. nitrogenase, which is found only in prokaryotes. The ammonia produced then can be incorporated by the enzyme for the growth and maintenance of the cell. Many nitrogen fixing prokaryotes are diazotrophic, that is, they can grow using dinitrogen gas as their sole source of nitrogen while other organisms can fix nitrogen only in symbiosis with another eukaryotic organism. Many leguminous plants are able to utilize this atmospheric nitrogen through an association with rhizobia.

This is also an agriculturally important association since the symbiotic bacteria fix nitrogen, which the plant can use, thus reducing the need for nitrogen fertilizer. Legumes enhance the productivity and sustainability of farming systems. This has economic and environmental significance for the sustainability of farming (17). Globally, Terrestrial BNF is estimated at between 100 and 290 million t of nitrogen year^{-1} (19, 11). Out of this, 40-48 million t is fixed by agricultural field crops (12, 18). However, very little work has been carried out to estimate the nitrogen fixation by fenugreek crop.

Agricultural importance of rhizobial interaction with fenugreek

Rhizobium inoculation of fenugreek increased the biomass of plant and seed production (28). Fenugreek is reported to fix about 48% of its total nitrogen during the growing season. Apart from this, symbiosis between fenugreek and rhizobia is a virgin area for research and needs further attention due to following reasons:

- Unlike most forage crops, fenugreek is an annual crop therefore allowing easy incorporation into short term crop rotations.
- Under field conditions, crops grown in rotation with

fenugreek will be benefited from nitrogen fixed by the crop. Crop rotations also have a positive effect on conservation of soil by reducing the chance of depletion of crop-specific nutrients from the soil, soil erosion and incidence of soil born pathogens.

- In addition to soil conservation, crop rotation can reduce the need of fertilizer and insecticide use while fenugreek act like cover crop resultant reduce weed problems.
- Since fenugreek can produce a high biomass in a short period of time, it can be used as a “green manure” crop to increase the organic matter content in soil depleted of this important ingredient.
- Fenugreek can grow on marginal lands and may be useful for reclamation of land disturbed by industrial activity.
- As a dry land crop, its water requirement is low; use of fenugreek can reduce the cost of irrigation and save water.
- Fenugreek seed needs to be inoculated with an appropriate *Rhizobium* inoculum to optimize its growth potential while increasing biodiversity in the soil.

By viewing these things in mind a strategy or action plan may be develop to prepare suitable rhizobial inoculants for fenugreek.

Use of rhizobial fertilizer in fenugreek for soil health and environment quality

The increasing use of chemical fertilizer, pesticides and weed control agents has damaged soil quality, ecology, environment and most importantly human health. Use of inorganic nitrogen fertilizer always possess a risk of contamination of underground and surface water resources through leaching or run off. Nutrient rich water can lead to eutrophication of streams, lakes and reservoirs and an increase in harmful bacteria and associated microbes of these fresh water sources. The increasing use of inorganic fertilizers have also created other environmental problems such as deterioration of soil quality, air pollution, reduced biodiversity and suppressed ecosystem function (43). Fenugreek has the potential to help organic farming practices by reducing ever-increasing fertilizer costs to the producer and by addressing public demand for more environmentally friendly agricultural practices.

Biofertilizers are live formulations of agriculturally beneficial microorganisms, which upon application to seed, root or soil can mobilize the availability of nutrients by their biological activity and help to improve the soil health. In recent years fenugreek has attracted the attention of producers to meet manufacturing demands for “functional food” additives and natural health products in Canada. Some suggest fenugreek seed could be added to foods like ground meat and baked goods, as a nutritional supplement as well as a potential functional food. The recent great increases in crop yields and food production in developed countries have been achieved by intensive agricultural practices. These increases, however, have not come without tremendous environmental costs (43). In developing countries the problems are different. The lack of fertilizers and adequate agricultural practices do not allow intensive crop production and a vast segment of the population remains undernourished. Clearly, there is an urgent need of rhizobial inoculants of fenugreek for sustainable agricultural practices on a global level.

Success of rhizobial fertilizer In adverse arid and semiarid environmental conditions

Environmental stresses are the major force that governs the food production in the arid and semi-arid regions. Drought, high and low temperature, salinity and air pollution are most frequent abiotic stresses which are caused by various environmental factors. It is assumed that several abiotic factors in the soil, such as water stress, high soil temperature, salinity, - nutrient deficiencies, alkalinity and acidity may limit growth, nodulation and nitrogen fixation of the legumes grown in the tropics region (9, 10). Moreover, a given stress may also have more than one effect e.g., salinity may act as water stress, which affects the photosynthetic rate. An excess of salts in the soil leads to both osmotic and ionic stress (4, 25). Bacteria face same adverse environmental conditions as their legume partner. Population of root nodulating soil bacteria (rhizobia) vary in their tolerance to major environmental factors (2, 8, 15, 20, 23, 24, 33, 37). However, the inoculated strains may be susceptible to loss of symbiotic traits such as infectiveness and effectiveness due to environmental factors and may not be competitive with the indigenous strains already present in the soil (38, 40). To achieve sustainable crop production to feed growing human population, strategic measures should be taken in management of these environmental stresses. One of the approach/strategy may be the selection and application of abiotic stress tolerant rhizobial strains.

Increasing salt concentration may have a detrimental effect on soil microbial populations as a result of direct toxicity as well as through osmotic stress (42). However, legume plants are more sensitive to salt or osmatic stress than the rhizobia (5). Rao *et. al.,* (32) found that *Rhizobium* strains well adapted to form effective symbiosis with their hosts at saline conditions are not necessarily obtained from saline soils. Also, Nair *et.*

al., (26) noticed that some rhizobial strains tolerating extremely high levels of salt had a significantly lower symbiotic efficiency under salt stress.

Recent developments in new pulse and pasture legumes, particularly in Australia, have emphasized the need for the selection of compatible *Rhizobium* spp. to optimize legume production (36). As a consequence, rhizobial survival in soil and growth in the rhizosphere is most important. Working with strains of *Mesorhizobium cicer*, adapted to acid conditions and chickpea (31), demonstrated that only 5% of strains tested were suitable for nodulation and growth in strongly acidic environments.

The survival and multiplication of the bacteria in the soil is influenced by temperature. Rhizobial strains which are suited to survival under different environmental conditions, effect of temperature on rhizobial growth rate under such conditions must be considered (17). Recently, Pongsilp and Leelahawonge (29) studied the response of root nodule symbionts of *Derris elliptica* Benth under different environment including different incubation temperature and reported that the tested strains, even among the same genus varied in their response to the levels of temperature. Elboutahiri *et. al.,* (10) observed the most optimum temperatures for rhizobial culture as 28°C-31°C. The survival of *Bradyrhizobium japonicum* under hyper osmotic treatments at various temperatures was studied by Beney et. al., (3) and reported that low temperature (4°C to 10°C) exhibiting a protective ef fect against exposure to high levels of osmotic pressure to rhizobia. Because high temperatures decrease rhizobial survival and establishment in tropical soils, repeated inoculation of grain legumes and higher rates of inoculation may be required. Temperature also affects nodule function, nodule initiation, rhizobia release from the infection thread and bacteroid development (34) and accelerates nodule senescence (16). However, Kulkarni and Nautiyal, (21) has done ecological survey of *Rhizobium* species of *Sesbania* and stated that *Rhizobium* sp. NBRI0102 and *Rhizobium* NBRI 2505 survived at 50°C and 65°C in YEB at pH 7.0 for up to 4 and 2 h respectively.

Generally, rhizobial strains from arid regions are adapted to such adverse environmental conditions. Some rhizobial strains may be effective inoculant strains for crop growing in adverse conditions. The best *Rhizobium*-legume symbiosis for arid conditions where drought and salinity prevail would be the ones that grow rapidly under ideal temperature and moisture conditions, with high tolerance levels to unfavourable conditions. The ability of inoculum strain to adopts the soil conditions and persist into the next growing season are important factors to consider when choosing an inoculum. Besides rhizobial inoculant, other environmental factors such as soil nitrogen content, soil physiochemical constraints and climatic conditions are important factors in determining the yield of crops. These factors together with the suitability of the inoculum strain(s) determine success in inoculation programmes. Large scale application of effective and efficient rhizobial strains to legume crop as inoculants would be attractive to increase crop yield as it would substantially reduce the use of chemical fertilizers and pesticides, which often pollute environment and contaminate the foodstuffs. Research and field trials for screening of effective and efficient rhizobial strains over decade have opened up new horizons for the agricultural bioinoculant industry. These biotechnological approaches can be exploited as low input, sustainable and environment-friendly technology for the management of plant stresses.

Use of rhizobia as a bio-control agent

In another approach, rhizobia may be used as plant growth promoting rhizobacteria (PGPR) in agriculture. Interest in biological control arose during 1920s and 1930s, when some plant pathogens were suppressed by introducing some antibiotic producing microbes to the natural habitats. Microorganisms that can grow in rhizosphere are ideal for use as biocontrol agents, since rhizosphere is the front-line defense for root against attack by pathogenic fungi (44). Root nodule bacteria of fenugreek may also possess some antimicrobial activities, which is still need to be validated under various conditions. If fenugreek microsymbiont contain such activities, could be used other than nitrogen fixer.

Production of rhizobial inoculums

The aim of *Rhizobial* inoculants production in Vietnam is high quality of products and economical cost of production. The rhizobial inoculums can be produced and applied in numerous ways. Inoculum can be prepared as powder, liquid and granular formulations. Granular formulations are convenient as they allow control of placement and application rate (39). An additional important feature of inoculums is the selection of the carrier (e.g., peat, perlite, mineral soil, charcoal). Whichever the carrier, its sterilization is necessary to maximize inoculum survival and subsequent infection rate (39).

If the full benefit from grain legume crops is to be achieved in terms of maximum yield and soil improvement, each crop should be inoculated with its own specific strain of rhizobia bacteria before planting. Inoculants specific for various crops including fenugreek can be seen in the Table (1).

Current status and cost of fertilizer

Presently, there is a wide gap of about 10 MT of plant nutrients between removal by crops and replenishment by fertilizers. Application of only organic manures could not fill this gap. Moreover, the nutrients supply is unlikely to improve due to competitive demand for the use of agricultural waste residue as fuel. So, biofertilizer especially *Rhizobium* inoculants for fenugreek is one of the alternatives left for supplementing the need of plant nitrogen. Biological nitrogen fixation is by far a cheaper and more sustainable process. Inoculation of legume seeds with root nodulating bacteria can result in a large benefit cost ratio. The cost of inoculants is about 1% of the total cost of input.

The future success of organic fenugreek production would largely depends upon size of the farm and supplies of non–chemical inputs, which have to be thoroughly backed up by well proven package of practices addressing to the objectives of producing organically. These organic farming practices have to be turn to change traditional concept of farming.

CONCLUSION

With the above discussion it is clear that use of *Rhizobium* inoculants as the component of organic farming is becoming necessary not only for market point of view but also for the welfare of farmer and earth society as a whole.

Table 1. Legume crops and their treatment with rhizobial inoculums

Legume Crops	Kg of seed treated (per 250 g pkt of inoculum)	Legume Crops	Kg of seed treated (per 250 g pkt of inoculum)
Chickpea	100	**Lucerne and medics**	25
Vetch, Field pea, Lentil	100	**Cowpea, Adzuki bean**	100
Faba bean	100	**Pigeonpea**	100
Fenugreek	100	**Lablab**	100
Lupin	100	**Mungbean**	100

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