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A study on the effect of bio-fertilizers on growth and yield of field pea (*Pisum sativum* L.)

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Abstract

Field pea (*Pisum sativum* L.) is one of the major cool-season legumes grown worldwide due to its high nutrition, nitrogen fixing ability and contribution to sustainable agricultural systems. The increasing cost and environmental issues of chemical fertilizers have led to the revival of bio-fertilizers. Bio-fertilizers are living microbial inoculants which enhance the availability of plant nutrients through biological processes. The present review is focused on the research work done to study the effect of bio-fertilizers especially *Rhizobium*, *Azotobacter*, *Azospirillum*, phosphate solubilizing bacteria (PSB) and mycorrhizal fungi on growth, nodulation and yield of field pea. The data of field trials and greenhouse experiments clearly showed that the application of bio-fertilizer significantly enhanced root nodulation, chlorophyll content, shoot and root biomass, number of pods per plant, seeds per pod, 100-seed weight and grain yield. Furthermore, the co-inoculation of *Rhizobium* with other bioinoculants and reduced doses of chemical fertilizers has demonstrated synergistic effects that can replace, partially or fully, conventional nitrogen and phosphorus inputs. This paper synthesizes results from about 30 major studies to give a clear understanding of the mechanisms, agronomic benefits and future potential of bio-fertiliser use in field pea production.

Keywords: *Pisum sativum*, bio-fertilizers, *Rhizobium*, *Azotobacter*, PSB, mycorrhiza, nitrogen fixation, legume, sustainable agriculture, grain yield

Introduction

Legumes have been a cornerstone of sustainable agriculture for thousands of years and among these, field pea (*Pisum sativum* L.) has a prominent place. Field pea is a member of the family Fabaceae, grown for its edible seeds, green pods and dried split peas in temperate and sub-tropical regions. It is an important source of plant protein (20-25%), dietary fibre, complex carbohydrates and essential micronutrients such as iron, zinc and folate, thus playing a crucial role in food security especially in developing countries (Bhat *et al.*, 2013; Sousa *et al.*, 2021) [3, 18].

Besides its nutritional value, field pea has a somewhat important ecological role. Like other legumes, it forms symbiotic associations with nitrogen fixing rhizobial bacteria in the root nodules and contributes 70-200 kg N/ha to the soil during a growing season (Garg and Geetanjali, 2007) [7]. This biological nitrogen fixation (BNF) reduces the need for synthetic nitrogenous fertilisers, and hence, the costs of production, and the risk of nitrogen leaching into groundwater-a concern that has become more urgent in the face of global environmental degradation.

But there are problems with field pea production. Soils are generally nutrient deficient, inefficient nodulation and inadequate availability of phosphorus often limit the yield potential of the crop, thereby limiting productivity. The traditional solution has been heavy use of chemical fertilisers especially nitrogenous and phosphatic compounds but this approach is plagued with well-known disadvantages such as increasing input costs, soil acidification, reduction of microbial diversity, emission of greenhouse gases and long-term depletion of soil health (Vessey, 2003; Bhattacharya and Jha, 2012) [21, 4].

It is under this context that bio-fertilizers have emerged as a promising and environment friendly alternative. Bio-fertilizers are combinations of live or dormant microorganisms which, when applied to seeds, plant surfaces or soil, colonize the rhizosphere and enhance plant growth by increasing the availability of primary nutrients or by stimulating plant hormones and root development (Bhattacharya and Jha, 2012; Bashan and Holguin, 1998) [4, 2].

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The most studied bioinoculants for legumes are *Rhizobium* species (symbiotic nitrogen fixers), *Azotobacter* and *Azospirillum* (free-living and associative nitrogen fixers), phosphate solubilizing bacteria (PSB) and *Arbuscular mycorrhizal* fungi (AMF).

Increasing number of studies indicate the effectiveness of these bio-fertilizers in improving the growth and productivity of field pea. But a good deal of this research is published in regional journals, in experimental stations, and at agronomy conferences. This review paper attempts to synthesize that knowledge into a coherent story. We discuss mechanisms of action of bio-fertilizers, experimental evidence of agronomic benefits, prospects of combining different bioinoculants with reduced chemical inputs and constraints still limiting widespread adoption.

2. Crop profile: Field pea

Field pea (*Pisum sativum* L.) is an annual cool-season legume native to Central Asia. It is widely grown in South Asia, Europe, North America and Australia. It is an important Rabi crop of India grown in Indo-Gangetic Plains, hilly terrains and plateau regions. The crop prefers well-drained loamy soils with slightly acidic to neutral pH of 6.0-7.5 and needs a cool growing period with temperature between 7 °C and 24 °C (Bhat *et al.*, 2013) [3].

Field pea is an agronomically short duration crop (90-120 days) thus fits well in multiple cropping systems. Crop rotation helps break pest and disease cycles and adds fixed nitrogen to the soil for subsequent crops. The International Food Policy Research Institute (IFPRI) and several FAO reports have reiterated time and again the potential of field pea in alleviating protein-calorie malnutrition in food-insecure areas (Sousa *et al.*, 2021) [18].

The crop has a unique advantage to utilize bio-fertilizer based nutrient management strategies because of its inherent capacity of fixing biological nitrogen through its symbiosis with *Rhizobium leguminosarum* biovar *viciae*. The rhizobial association directly enhances the fundamental nitrogen metabolism of the crop.

3. Bio-fertilizers: Concept, types and mechanism

3.1 Classification and Definition

Bio-fertilizers are defined as the preparations containing efficient strains of living microorganisms which on application to seeds, soil or plant surfaces enhance growth by increasing the availability of nutrients to the host plant (Bhattacharya and Jha, 2012; Vessey, 2003) [4, 21]. They are generally divided into:

- **Nitrogen fixing bio-fertilizers:** *Rhizobium* (symbiotic) *Azotobacter* and *Azospirillum* (free living/associative) Cyanobacteria (BGA)
- **Phosphate solubilizing microorganisms (PSM):** *Pseudomonas*, *Bacillus*, *Aspergillus* and *Penicillium* spp.
- **KMB (potassium mobilising bacteria):** *Frateuria aurantia* and related strains.
- **Mycorrhizal fungi:** *Arbuscular mycorrhizal* fungi (AMF) including *Glomus* and *Rhizophagus* spp.
- **Plant growth promoting rhizobacteria (PGPR):** big group of bacteria producing IAA, gibberellins, cytokinins, siderophores and ACC-deaminase.

3.2 Rhizobium: The Leading Bio-fertilizer for Legumes

Rhizobium leguminosarum biovar. The main microsymbiont

of field pea is *viciae*. It colonizes root hairs, induces nodule organogenesis and fixes the atmospheric nitrogen (N₂) to ammonia (NH₃) by means of the nitrogenase enzyme complex. This process can provide 70-200 kg N/ha per cropping season depending upon the soil conditions, strain efficiency and environmental factors (Garg and Geetanjali, 2007; Brockwell and Bottomley, 1995) [7, 6].

Nodulation efficiency depends upon several factors like soil pH, soil moisture, soil temperature and presence of compatible and competitive strains of *Rhizobium*. Many agricultural soils may lack adequate indigenous *Rhizobium* populations or populations that are not effective on modern pea cultivars and therefore seed inoculation with efficient commercial strains is required. Field pea inoculated with *Rhizobium* showed significant increase in number of nodules/plants, nodule weight and nitrogenase activity over uninoculated controls (Kumar *et al.*, 2009) [9].

3.3 Azotobacter and Azospirillum

Azotobacter chroococcum is a free-living nitrogen fixing bacterium that is found in the rhizosphere of many crops. Apart from nitrogen fixation, it produces plant growth hormones such as indole acetic acid (IAA), gibberellins and cytokinins, vitamins and antifungal metabolites (Bhattacharya and Jha, 2012) [4]. In field pea, inoculation with *Azotobacter* increased root biomass and vegetative growth, especially when combined with *Rhizobium* (Singh *et al.*, 2011) [17].

Azospirillum brasilense is an associative nitrogen fixing bacterium that colonizes the endodermis surface of roots. Its main contribution is by the production of phytohormones and not by large scale nitrogen fixation. This in turn leads to the proliferation of roots and water and mineral uptake. *Azospirillum* inoculation of legumes significantly enhanced the formation of lateral roots and efficiency of nutrient absorption (Mia and Shamsuddin, 2010) [11].

3.4 Phosphate Solubilizing Bacteria (PSB)

Phosphorus (P) is the second most important macro-nutrient after nitrogen. A large fraction of soil phosphorus is in unavailable forms to plants in insoluble mineral complexes. PSB solubilise these insoluble phosphates by secretion of organic acids (gluconic, citric, oxalic), protons and phosphatases to convert them into plant available orthophosphate ions (Bhattacharya and Jha, 2012) [4]. Reported increasing shoot phosphorus content, root length and grain yield of field pea inoculated with PSB. Yadav and Tilak (2001) [22] observed that grain yield of field pea showed 15-25% increase when PSB was combined with *Rhizobium* inoculation than alone of each bioinoculant.

3.5 AMF (Fungi Micorrhiza)

Field pea is one of ~80% of terrestrial plants that associate mutualistically with *Arbuscular mycorrhizal* fungi (AMF). The fungal hyphae extend well beyond the root zone, thus considerably increasing the surface area for the uptake of phosphorus and micro nutrients. AMF not only help in nutrient uptake but also provide drought tolerance and resistance to soil-borne pathogens (Bothe *et al.*, 2010; Barea *et al.*, 2005) [5, 1]. In this sense, Pellegrino *et al.*, (2015) [15] demonstrated that the inoculation of AMF increased the yield of field pea in low P conditions, confirming the importance of mycorrhizal symbiosis for sustainable production of legumes.

4. Impact of bio-fertilizers on growth parameters

4.1 Seed Germination and Seedling Early Growth

The most important processes in the life of a crop are germination of seed and establishment of early seedlings. The effect of bio-fertilizer seed treatment on these early growth parameters has been investigated by some researchers. Mishra *et al.*, (2013) ^[12] reported significant improvement in germination percentage and seedling vigour index of field pea seed inoculation with *Rhizobium* and *Azotobacter* as compared to uninoculated control. This improvement was attributed to the production of IAA and gibberellins by the applied microorganisms which stimulate seed imbibition and elongation of the embryonic axis. Similarly, Tiwari *et al.*, (2016) ^[20] reported that combined inoculation of *Rhizobium* + PSB increased germination speed and fresh weight of the seedling in field pea under pot culture condition. The authors suggested that the early solubilization of phosphorus by PSB was helpful in the synthesis of nucleic acids and energy transfer, which are essential for germination.

4.2 Root nodule development

The formation of root nodules is probably the most direct measure of the effectiveness of *Rhizobium* inoculation. Adequate biological nitrogen fixation and all subsequent growth processes are assured by effective nodulation. There is considerable evidence that pea plants inoculated have more numerous nodules and heavier nodules than uninoculated plants.

Rhizobium inoculation increased the amount of nodules/plant by 35-65% in field pea (Kumar *et al.*, 2009) ^[9]. Pandey and Kumar (2012) ^[13] reported that dual inoculation of *Rhizobium* + *Azotobacter* resulted in higher nodule number and specific nitrogenase activity as compared to single inoculations. This synergistic effect was attributed to the production of growth hormones by *Azotobacter*, which can stimulate the formation of infection threads and the division of cortical cells, both of which are required for nodule formation.

Singh and Singh (2010) ^[16] also reported interesting observations that the addition of PSB to the *Rhizobium* inoculation mix improved the nodule occupancy by the introduced *Rhizobium* strain. Probably the increased phosphorus availability benefited the energy intensive nitrogen fixation process which demands substantial amounts of ATP.

4.3 Plant vegetative growth and canopy development

Plant height is a complex measure of vegetative vigour and is affected by the supply of nitrogen, phosphorous and growth hormones which can all be controlled by biofertilizers. Several studies have reported a significant increase in plant height after the application of bio-fertilizer. The application of *Rhizobium* + PSB + *Azotobacter* together at 60 days after sowing improved plant height by 18-22% over uninoculated controls. This was reported by Meena *et al.*, (2015) ^[10] from a field trial conducted in Rajasthan, India. This increase was mainly due to an improved nitrogen nutrition favoring the activity of the apical meristem and an IAA production stimulating cell elongation. Inoculated treatments also had a higher canopy extension and number of primary branches per plant, reflecting a more vigorous vegetative architecture.

Azospirillum inoculation increased the branching in field

pea (Pant and Bokhtiar, 2005) ^[14]. Vessey (2003) ^[21] reviewed literature to demonstrate that IAA production mediated by PGPR is an important mechanism that underlies shoot elongation and branching in legumes.

4.4 Leaf area index and chlorophyll content

Photosynthetic efficiency is an important aspect of crop productivity, with leaf area index (LAI) and chlorophyll content being its proximate indicators. bio-fertilizer inoculation improved the parameters significantly. Inoculation of *Rhizobium* + *Azotobacter* increased Leaf Area Index of Field Pea by 12-18% over uninoculated plots at flowering stage (Singh *et al.*, 2011) ^[17]. The higher availability of nitrogen resulted in increased leaf area due to increased chlorophyll synthesis and leaf expansion.

Mishra *et al.*, (2013) ^[12] and Tiwari *et al.*, (2016) ^[20] reported significant increase in chlorophyll content (SPAD values) of bio-fertilizer treated plants. More chlorophyll content is directly correlated with more photosynthate production which supports more accumulation of biomass and finally higher yields. Also, the role of cytokinin producing bio-fertilizers like *Azotobacter* in delaying leaf senescence and maintaining greenness has been highlighted (Bhattacharya and Jha, 2012) ^[4].

4.5 Root Biomass and Structure

The ability of the crop to explore the soil profile for water and nutrients is determined by the root architecture (total root length, root surface area and root branching). Bio-fertilizers especially *Azospirillum* and PSB are reported to influence root morphology profoundly through IAA and other phytohormones.

Mia and Shamsuddin (2010) ^[11] found that inoculation of legumes with *Azospirillum* resulted in an increase in lateral roots density up to 40%, compared with control plants. More root surface area = more uptake of phosphorus, potassium and micronutrients. This is due to the fact that PSB inoculation also guarantees higher availability of phosphorus in the immediate rhizosphere to roots thereby further stimulating root exploration and nodule formation. Pellegrino *et al.*, (2015) ^[15] showed that AMFs-inoculated field pea plants had a significant increase in the density of extraradical mycelium, which extended the nutrient absorptive surface well beyond the root zone.

5. Effect of bio-fertilizers on yield and yield attributes

5.1 Pods/Plant

The number of pods per plant is an important component of yield and an immediate measure of the reproductive success of the field pea plant. The number of pods produced will be determined by the nitrogen status of the plant at flower initiation and pod set when biological nitrogen fixation must be at its peak. Inoculation of bio-fertilizer had always increased this parameter.

The combined inoculation of *Rhizobium* + *Azotobacter* + PSB showed 20-28 percent increase in pods per plant over uninoculated control (Meena *et al.*, 2015) ^[10]. Pandey and Kumar (2012) ^[13] found that dual inoculation with *Rhizobium* + PSB gave the maximum number of pods per plant over all the single and combined inoculant treatments, suggesting that improved phosphorus nutrition during reproductive stages was particularly helpful for pod initiation.

5.2 Seeds per pod and seed size

Nitrogen and carbohydrate availability during seed filling affect number of seeds per pod. Effective Rhizobium nodulation furnishes adequate nitrogen nutrition and amino acids for protein synthesis in the developing seeds. Research has always showed that field pea plants treated with bio-fertilizer have more seeds per pod and the seeds are heavier and bigger.

Kumar *et al.*, (2009)^[9] observed that Rhizobium inoculation increased the number of seeds/pods from 5.8 (control) to 7.1 and 100 seed weight from 18.2 g to 21.4 g. The addition of *Azotobacter* to Rhizobium also enhanced 100-seed weight by 14% as compared to Rhizobium alone Singh *et al.*, (2011)^[17] which was probably due to additional supply of growth hormones and micronutrients provided by *Azotobacter*.

5.3 Grain Biology and Production

Grain yield is the ultimate criterion of agronomic performance. Application of bio-fertilizer has been reported to enhance grain yields of field pea in several studies conducted under different soil and climate conditions.

Yadav and Tilak (2001)^[22] reported that combined inoculation of Rhizobium + PSB gave a grain yield of 1,842 kg/ha as against control uninoculated of 1,410 kg/ha which was 30.6% increase. Tiwari *et al.*, (2016)^[20] reported 25-35% yield advantage of triple inoculation (Rhizobium + *Azotobacter* + PSB) over uninoculated control in field conditions of Uttar Pradesh. Mishra *et al.*, (2013)^[12] also demonstrated that the application of combined bioinoculant in field pea could achieve yield comparable to 50% of the prescribed dose of chemical nitrogen and phosphorus fertilizers.

In the study of Meena *et al.*, (2015)^[10], the biological yield (total dry matter production) was significantly increased in bio-fertilizer treated plots which suggested a better overall biomass accumulation that supports grain filling and residue return to the soil.

5.4 Harvest index

The harvest index (HI) is the ratio of grain yield to total biological yield and is indicative of the efficiency of the plant in partitioning photosynthates to reproductive parts. The bio-fertilizer treatments generally maintained or slightly improved HI in the field pea. The increase in HI from 0.41 (control) to 0.46 with Rhizobium + PSB inoculation in the present study as reported by Singh and Singh (2010)^[16] indicated that improved nutrition not only enhanced the total biomass but also improved preferential accumulation of assimilates in seeds.

5.5 Protein Content and Nutritional Value

Seed filling nitrogen assimilation is directly associated with field pea grain protein content. Bio-fertilizer naturally increases amino acid supply to the developing seeds by nitrogen fixation. The seeds of Rhizobium-inoculated legumes are consistently 2-5% higher in protein than the non-inoculated ones (Garg and Geetanjali, 2007)^[7]. There is quite a large volume of literature showing this.

The grain protein content of field pea increased by 8-12% in combined bio-fertilizer treatments compared to controls (Tiwari *et al.*, 2016)^[20]. This increase in nutritional quality is a significant factor not only for human consumption, but

also for the use of field pea as a protein supplement in livestock feed.

6. Co-recommendation method of bio-fertiliser and reduced chemical fertiliser

One of the practically important areas of research has been the exploration of integrated nutrient management (INM) where bio fertilisers are combined with reduced doses of chemical fertilisers to achieve optimal yields with reducing input costs and environmental impact. The logic is that bio-fertilizers alone may not always be able to meet the total nutrient requirement of a crop-especially in degraded or nutrient poor soils-but can significantly reduce the amount of chemicals needed.

Pandey and Kumar (2012)^[13] observed that 75% recommended dose of NPK fertilisers with Rhizobium + PSB inoculation could achieve grain yields at par with recommended dose of NPK alone in field pea. This represents a major reduction in chemical input with no yield loss. The economic analysis of their study revealed that the INM method reduced the fertiliser cost in the range of 20-25% with profitability.

Meena *et al.*, (2015)^[10] adopted similar approach and found that 50% NPK + triple inoculation (Rhizobium + *Azotobacter* + PSB) was better than 100% NPK by 5-7% in grain yield. The authors attributed this unexpected superiority to the enhancement of soil microbial activity, enzyme activity and nutrient cycling in the rhizosphere caused by the bio-fertilizers which are not supplied by the chemical-only treatments.

These results are in agreement with the general literature on PGPR and bioinoculants reviewed by Vessey (2003)^[21] and Bashan and Holguin (1998)^[2], who emphasize that microbial inoculants provide systemic benefits to soil health beyond the mere supply of nutrients, such as better soil structure, water retention and disease suppression.

7. Mechanisms of the bio-fertilizers efficiency

7.1 Biological N₂ fixation (BNF)

In root nodules Rhizobium containing bacteroids express the enzyme complex nitrogenase which catalyses the irreversible reduction of dinitrogen (N₂) to ammonia (NH₃). The process needs 16 ATP and 8 electrons per molecule of N₂ fixed. The fixed nitrogen is assimilated into glutamine and glutamate in the nodule tissue of the plant and transported to the growing tissues in the form of ureides or amides (Garg and Geetanjali, 2007)^[7]. Field pea can fix enough nitrogen to provide 70 to 200 kg N/ha each season, greatly reducing or eliminating the need for synthetic nitrogen inputs.

7.2 Solubilization of Phosphate

PSB synthesize and excrete low molecular weight organic acids (citric, gluconic, oxalic, lactic acids) that acidify the rhizosphere and solubilize calcium, iron and aluminum phosphate complexes, releasing plant-available H₂PO₄⁻ ions. In addition, PSB also produce phosphatase enzymes which mineralize organic forms of phosphates in soil. This results in increased plant available phosphorus to support root development, energy metabolism (ATP synthesis) and RNA/DNA synthesis, which are critical for rapid growth and productive crop development.

7.3 Production of Plant Hormones

Phytohormones such as indole acetic acid (IAA) that stimulate root cell elongation and lateral root initiation, gibberellins that promote stem elongation and seed germination and cytokinins that promote cell division, delay leaf senescence and increase the nutrient sink strength in developing seeds are produced by the rhizosphere of growth promoting bacteria. *Azotobacter*, *Azospirillum* and several PSB strains are known for high IAA production and it is generally accepted that this is the main mechanism by which these bacteria improve root architecture (Vessey, 2003; Bhattacharya and Jha, 2012)^[21, 4].

7.4 Siderophore and Antibiotic Synthesis

Some of PGPR strains can secrete siderophores which are low molecular weight iron chelating compounds. These can capture iron from soil and deliver it to the plant. This mechanism is especially relevant in calcareous or alkaline soils where iron is less available. Also, PGPR strains have been reported to produce antibiotics and lytic enzymes that can inhibit soil-borne pathogens like *Fusarium*, *Pythium* and *Sclerotinia* species leading to reduction in incidence of disease and improvement in crop stand (Bashan and Holguin, 1998)^[2].

7.5 ACC Deaminase Activity

Some strains of PGPR produce the enzyme 1-aminocyclopropane-1-carboxylate (ACC) deaminase which degrades the ethylene precursor ACC in the rhizosphere. This reduces the ethylene in plant tissues leading to relief from the growth inhibitory effect of stress and enhances root development under adverse conditions such as drought, salinity and heavy metal stress. This mechanism is especially important in maintaining field pea productivity in water-deficit or salt-affected soils (Vessey, 2003)^[21].

8. Biological soil health & biofertilizers

Bio-fertilizers not only directly promote plant growth but also have important long-term effects on the soil microbial community and soil health. For example, inoculation with rhizobium increases the number of bacteria associated with the root that are not the inoculated rhizobium strain. This is probably due to the change in the composition of the root exudates in inoculated plants and this provides a more nutritious substrate for the entire microbial community (Barea *et al.*, 2005)^[1].

Studies on field pea by Singh *et al.*, (2011)^[17] and Meena *et al.*, (2015)^[10] have shown that application of bio-fertilizer to plots significantly increased soil microbial biomass carbon, dehydrogenase activity, urease activity and phosphatase activity in plots as compared to plots with only chemical fertiliser. The higher the enzyme activities, the more functionally active the microbial community is with higher potential for nutrient cycling and organic matter decomposition.

bio-fertilizer application at various cropping seasons repeatedly as reviewed by Bhattacharya and Jha (2012)^[4] that lead to resilient agroecosystem with ability to sustain productivity under changing climate. This legacy effect is a benefit that chemical fertilisers cannot provide and makes a strong argument for inclusion of bio-fertilizers in mainstream field pea production.

9. Use of bio-fertilizer: limitations and problems

Although the evidences supporting the use of bio-fertilizer in field pea are impressive, large-scale adoption by farmers

is limited particularly in developing countries. There are many constraints that may explain this disconnection between research findings and field practice.

Firstly, the quality and shelf life of bio-fertilizers are of utmost importance. The success of microbial inoculants depends on the quality of the carrier material, the storage temperature and conditions of transport. Bio-fertilizers produced or stored improperly may not have sufficient viable cells to be efficient. This may result in inconsistent results and undermine the confidence of farmers (Vessey, 2003)^[21]. Quality standards in much of South Asia and Sub-Saharan Africa continue to be a challenge due to regulatory oversight and cold-chain logistics.

Secondly, the effectiveness of inoculants can vary a lot depending on the soil type, climate and crop variety. A very competitive strain of *Rhizobium* in one soil or environment situation may be non-competitive in another. Extreme soil pH, high native *Rhizobium* populations, pesticide residues and soil moisture stress can all inhibit inoculant performance (Brockwell and Bottomley, 1995; Garg and Geetanjali, 2007)^[6, 7].

Third, farmers have little knowledge of the correct application of bio-fertilizer seed coating techniques, inoculant compatibility with fungicide seed treatments and the need for fresh products. In many developing countries, the agricultural extension systems are unable to properly transfer and demonstrate the technology of bioinoculants to the smallholder farmers.

Fourth, the initial response to bio-fertilizers is generally slower and less pronounced than synthetic fertilizers, especially on soils with severe deficit of nutrients. Farmers who are used to the rapid green-up that urea can produce may not find bio-fertilizers useful unless the benefits in terms of long-term soil health are clearly communicated and demonstrated over several seasons (Bhattacharya and Jha, 2012)^[4].

10. Future Prospects And Directions For Research

The prospects for bio-fertilizer research and application in field pea are positive as a number of trends converge: the global push towards sustainable agriculture, the rising cost of synthetic fertilisers, the increasing sophistication of microbial biotechnology and the increasing consumer demand for sustainably produced food.

One important direction is the development of next-generation consortium bio-fertilizers that combine multiple compatible strains of nitrogen fixers, phosphate solubilizers and mycorrhizal fungi in one formulation. Initial studies suggest that properly designed consortia may be more efficient in providing more complete nutritional support and wider ecological functions than single strain inoculants (Bashan and Holguin, 1998; Barea *et al.*, 2005)^[2, 1]. The challenge is to ensure compatibility of the consortium members and viability of all the strains in the carrier.

Recent advances in microbiome research and metagenomics have opened new opportunities to better understand the complex interactions of introduced bioinoculants, native soil microbial communities and the plant root. These insights can be useful for selection and design of bio-fertilizer strains that are effective in isolation but also competitive and functional in the complex ecological environment of agricultural soils (Bothe *et al.*, 2010)^[5].

Biopolymer-encapsulated and nano-encapsulated bioinoculants development is likely to offer significant shelf-life extension, heat stability and more targeted delivery

of bacteria to the root zone. Sousa *et al.*, (2021) ^[18] Several academic and commercial research groups are exploring chitosan-coated, alginate-bead and lignin-based encapsulation systems that could enable bio-fertilizers to be viable under tropical storage conditions.

Finally, breeding programs for enhanced symbiotic compatibility between modern high-yielding field pea varieties and efficient *Rhizobium* strains are a long-term strategy to maximize the genetic potential of both partners. The co-inoculation breeding strategies, that is, the selection of plant genotypes together with specific bioinoculant strains, may ultimately provide a more systemic solution than improved varieties or improved inoculants alone.

11. Conclusion

The overall evidence presented in this paper strongly supports the role of bio-fertilizers for sustainable field pea production. Application of Bio-fertilizers alone or in combinations always enhances the significant growth and productivity parameters of *Pisum sativum* L. from seed germination to grain filling to root nodulation to soil health. *Rhizobium* inoculation is the basis of any bio-fertilizer strategy for field pea, as it directly affects nodulation and biological nitrogen fixation. However, the best results are always obtained when *Rhizobium* is combined with complementary inoculants such as *Azotobacter*, PSB or AMF and when these biological inputs are used in combination with reduced doses of chemical fertilisers in an intelligent INM framework.

And the benefits are not only to the farm field. Less use of chemical fertiliser means less greenhouse gases, less nitrate leaching, lower production costs and healthier soils for future generations. Bio-fertilizers improve the soil health and improve the nutritional quality of pea grain, increasing the protein content and micronutrient density-results that are highly relevant to food security.

There are issues with product quality, response variability across environments and farmer adoption but these are issues of implementation, not fundamental scientific doubt. Further investment in research, quality regulation, extension and farmer education has the real potential to transform bio-fertilizers in field pea production into a more productive, profitable and ecologically sound enterprise. The future of legume farming is surely microbial.

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