EFFECT OF AM FUNGI AND PSB INOCULATION ON THE PER CENT ROOT COLONIZATION, AM FUNGAL SPORE NUMBER AND PSB POPULATION IN THE RHIZOSPHERE SOILS OF BRINJAL (SOLANUM MELONGENA L.)

K. SIVAKUMAR, Assistant Professor Department of Agricultural Microbiology Faculty of Agriculture Annamalai University, Annamalainagar – 608 002

N. SUGAPRIYA, PG Student Department of Agricultural Microbiology Faculty of Agriculture Annamalai University, Annamalainagar - 608 002

Abstract

A pot culture experiment was conducted to study the inoculation effect of AM fungi (*Glomus fasciculatum*) and Phosphobacteria (*Bacillus megaterium*) on the per cent root colonization, AM fungal spore number and Phosphobacterial population in the rhizosphere soils of brinjal (*Solanum melongena* L.) var PLR 2 at different levels of phosphorus. In all the treatments, *G. fasciculatum* and *Bacillus megaterium* inoculated brinjal recorded higher values than uninoculated plants. The root colonization percentage (84.52) and AM fungal spore number (129 100 g⁻¹ soil) in the rhizosphere soil were the highest in 75 per cent phosphorous level followed by 50 per cent phosphorous (71.28, 124.00 spores 100 g⁻¹ soil). The highest number of phosphobacterial population (11.33 × 10⁻⁶ Cfu) was recorded by the co-inoculation of *G. fasciculatum* and *B.megaterium* at 75 per cent phosphorous levels.

Key words: AM Fungi, Phosphobacteria, brinjal cultivation, agricultural produce, etc.

1. Introduction

Eggplant (*Solanum melongena*) or aubergine, is a specie of nightshade, grown for its edible fruit. Eggplant is the common name in North America, Australia and New Zealand, but British English uses the French word aubergine. It is known in South Asia and South Africa as brinjal. Brinjal has an important nutritional value due to its composition, which includes minerals like potassium, calcium, sodium and iron as well as dietary fiber. It contains 92.7 per cent water, 4 per cent carbohydrates, 1.4 per cent protein, 1.3 per cent fiber, 0.3 per cent fats, 0.3 per cent minerals and vitamin A in a negligible quantity. The varieties of *Solanum melongena* L. show a wide range of fruit shapes and colours, ranging from oval or egg –shaped to long club shaped and form white, yellow, green through degrees of purples pigmentation to almost black. India is the second largest producer of brinjal in the world next to China and produces 14 MT from an area of 711.3 ha. In India, the farmers use huge more amounts of chemical fertilizers for crop production especially for vegetables,

thus the soil which leads to soil pollution and ground water contamination, ultimately causing health hazards. In order to avoid the environmental pollution especially soil pollution, most of the scientists are recommending the use of bio fertilizers along with inorganic fertilizers in a sustainable manner to maintain the soil health and also the productivity, diseases and increasing the yield. The AM fungus varies with host ranges. Though they are ubiquitous, they showed that the every taxonomic group of plants and the list of species not infected is probably far of microorganisms like bacteria, fungi and actinomycetes which may help in increasing crop productivity by way of helping in solubilization of insoluble phosphorus, stimulating plant growth by providing hormones, vitamins and other growth promoting substances. Phosphate Solubilizing Bacteria (PSB) are capable of hydrolyzing organic and inorganic phosphorus from insoluble compounds and PSB produce phosphatase like phytase that hydrolyse organic forms of phosphate compounds efficiently.

2. Materials and methods

Isolation and screening of AM fungi: Brinjal rhizosphere soil sample were collected from twenty different locations in Cuddalore District of Tamilnadu. Four different AM fungal species *viz.*, *Glomus fasciculatum, Glomus mosseae, Gigaspora margarita* and *Acaulospora laevis* were isolated, characterized and identified under stereo zoom microscope according to Gerdemann and Trappe, 1974. Isolated AM fungi are screened for the efficiency by root colonization percentage, AM fungal spore numbers in soil. All the four AM fungal species colonized the roots of brinjal. However, the degree of root infection and colonization varied considerably between them. The response of brinjal in terms of root colonization by AM fungi was the highest with *Glomus fasciculatum* followed by *Glomus mosseae, Gigaspora margarita* and *Acaulospora laevis* in soils.

Enumeration of Phosphobacteria: Phosphobacteria enumerated from the rhizosphere soils of different brinjal grown fields by serial dilution plate technique. The soil samples were serially diluted upto 10^{-4} dilution. One ml of aliquots of last dilution was plated in using Sperber's hydroxy apatite medium. The plates were incubated upto two weeks at $28\pm2^{\circ}$ C. The bacterial colonies showing clear zone were enumerated and expressed as cfu g⁻¹ of oven dry soil.

3. Treatment details

- T₁ Control
- T_2 -RDF
- T₃ Glomus fasciculatum
- T₄ Bacillus megaterium
- T_5 -75% of P+ G. fasciculatum

- T_6 -75% of P + B. megaterium
- T₇-75% of P + G. fasciculatum + B. megaterium
- T_8 -50 % of P+ G. fasciculatum
- T₉-50 % of P + *B. megaterium*
- T_{10} -50 % of P + G. fasciculatum + B. megaterium

4. AM fungal colonization in brinjal roots

The percentage mycorrhizal colonization of the roots was determined by the method of Phillips and Hayman. The roots were washed gently in tap water. The washed roots were cut into one cm length and then immersed in 10 per cent KOH solution for clearing the host cytoplasm and nuclei for stain penetration. Then it was autoclaved at 15 lbs/sq, inch pressure for about 20 minutes. Then the root bits were taken out and washed with tap water for about three times or until no brown colour appeared in the rinsed water. The roots were acidified with two per cent hydrochloric acid (3-4 minutes) for proper staining. The acid was poured off without rinsing with water and root bits were stained with 0.05 per cent tryphan blue in lacto phenol solution and boiled for 10 minutes. These root bits were examined under compound microscope. Fifty root segments in each replication were used to determine AM fungal colonization per cent.

Per cent root colonization = $\frac{\text{Number of root bits with infection}}{\text{Total number of root bits examined}} \times 100$

5. Survey for the occurrence of AM fungal spores

AM fungal spore population was estimated by wet sieving and decanting method of Gerdemann and Nicolson. One hundred gram of rhizosphere soil samples were taken from the rhizosphere of brinjal mixed thoroughly in one lt. of tap water to settle down the heavier particles for few seconds. The suspension was decanted through a coarse soil-sieve (500-800 µm sieve) to remove large pieces of organic matter. The liquid which passed through the sieve was collected separately and stirred to resuspend all particles. The suspension was decanted through a sieve fine enough to retain desired spores (38-250 µm sieve). The material retained on the sieve was washed with a stream of water to ensure that all colloidal materials were passed through the sieve. The small amount of remaining debris were transferred to a shallow layer of water in a petridish and examined under a Stereo zoom microscope. The spore numbers from each soil sample were counted and expressed per 100 g of soil.

6. Results

The per cent root infection by inoculated brinjal, increased with the advancement age of the plants. The maximum root infection AM fungal spore and PSB population were observed at 130 DAT. The effect of inoculation of *G. fasciculatum* on the per cent root colonization and AM fungal spore population was recorded at 130 DAT and the results are presented in Table 1. The AM fungal root colonization and AM fungal spore number increased with increase in plant age. In all the treatments, *G. fasciculatum* and *Bacillus megaterium* inoculated brinjal recorded higher values than uninoculated plants. The root colonization percentage (84.52) and AM fungal spore number (129.00, 100 g⁻¹ soil) in the rhizosphere soil were the highest in 75 per cent phosphorous level followed by 50 per cent phosphorous (71.28, 124.00spores 100 g⁻¹ soil) respectively. From this study, it was observed that addition of low levels of phosphorous increased the mycorrhizal colonization and AM fungal spore number in PLR 2 brinjal.

TABLE 1

Co-inoculation Effect of *G. fasciculatum* and *B. megaterium* on the per cent Root Colonization and Spore Population in the rhizosphere soil of Brinjal at different Levels of Phosphorus

GN	Treatments			AMF root colonization (%)			AMF spore numbers (100 g ⁻¹ soil)		
S.No.				45 DAT	90 DAT	130 DAT	45 DAT	90 DAT	130 DAT
1.	T_1	:	Control	25.01	44.71	52.53	48	58	70
2.	T_2	:	RDF	25.98	48.91	56.79	72	77	98
3.	T ₃	:	Glomus fasciculatum	25.93	47.56	56.68	69	74	92
4.	T_4	:	Bacillus megaterium	25.53	46.75	56.13	62	71	91
5.	T ₅	:	75% of P+ G. fasciculatum	30.23	51.31	62.17	92	110	122
6.	T ₆	:	75% of P + B. megaterium	29.17	51.17	69.00	70	101	94
7.	T ₇	:	75% of P + G. fasciculatum + B. megaterium	33.18	55.32	84.52	106	113	129
8.	T ₈	:	50% of P + G. fasciculatum	27.18	49.17	70.20	81	90	124
9.	T 9	:	50% of P + B. megaterium	26.28	48.17	60.12	77	82	93
10.	T ₁₀	:	50% of P + G. fasciculatum + B. megaterium	27.75	50.49	71.28	83	97	124

The rhizosphere soil phosphobacterial population was estimated on 45, 90 and 130 DAT and the results are presented in Table 2. In all the periods, inoculation of *G. fasciculatum* and *B.megaterium* both single and combined at different phosphorous levels increased the rhizosphere soil phosphobacterial population compared to uninoculated control. Among the single inoculation, *B.megaterium* inoculated treatment, recorded more number of phosphobacterial population (10.33×106 cfu) followed by *G. fasciculatum* (8.00×106 cfu). The highest number of phosphobacterial population (9.66×106 cfu) was recorded by the co-inoculation of *G. fasciculatum* and *B.megaterium* at 75 per cent phosphorous levels. Addition of low levels of phosphorous with co-inoculation of *G. fasciculatum* and *B.megaterium* highly enhanced the survival of phosphate solubilizers in the rhizosphere soils of brinjal.

S.No.			Treatments	Phosphobacterial population (×10 ⁶ cfu g ⁻¹ oven dry soil)			
				45 DAT	90 DAT	130 DAT	
1.	T_1	:	Control	5.33	6.66	7.33	
2.	T_2	:	RDF	6.33	7.33	8.00	
3.	T ₃	:	Glomus fasciculatum	6.66	8.00	8.01	
4	T_4	:	Bacillus megaterium	9.00	10.33	9.33	
5.	T_5	:	75% of P+ G. fasciculatum	9.66	10.00	8.33	
6.	T ₆	:	75% of $P + B$. megaterium	9.33	9.66	11.00	
7.	T ₇	:	75% of P + G. fasciculatum + B. megaterium	8.66	10.66	11.33	
8.	T ₈	:	50% of P + G. fasciculatum	8.00	8.66	9.50	
9.	T 9	:	50% of P + B. megaterium	8.66	9.00	10.20	
10.	T ₁₀	:	50% of P + G. fasciculatum + B. megaterium	9.00	10.00	10.66	

1.CA

TABLE 2Co-inoculation Effect of G. fasciculatum and B.megaterium on Phosphobacterial Population in
the Rhizosphere soil of Brinjal at different Levels of Phosphorus

7. Discussion

The synergistic effect of G. fasciculatum and Pseudomonas striata on the growth, nutrient uptake and total dry weight of neem seedlings were found to be superior over individual inoculation at different stages of Neem seedlings. Interaction of phosphate solubilizing bacteria and AM fungi on tomato growth, soil microbial activity and production of organic acids in non-sterile soil containing hydroxyapatite and glucose was examined. The P concentration was greatest in all treatments and total N and P uptake in plants were higher in treated ones compared to control. A tripartite symbiosis between Azospirillum sp., Pseudomonas straita and G. fasciculatum enhanced the growth of rhizosphere microflora of cotton. Similar interaction occurred between P solubilization and AM fungi. The rhizosphere population of AM fungi and phosphate solubilizing bacteria were determined from soil samples collected from mixed and monocropped coffee and cardamom. The population of phosphorous solubilizing bacteria and fungi was higher in coffee and cardamom, respectively in both cropping systems. The interaction between mineral phosphate solubilizing bacterium and AM fungus and Azotobacter at different levels of fertilizers increases the mycorrhizal colonization, when all the three biofertilizers were added at 75 per cent of the recommended dose of NPK on sweet basil Ocimum basilicum. Interactive efficacy of phosphobacteria Bacillus megaterium and arbuscular mycorrhizae inoculated at different soil types were analysed. Among the treatments dual inoculation of AM fungi and phosphobacteria gave the most satisfactory outcome in Amaranthus tritis.

8. Conclusion

In the present study, the maximum root colonization percentage and spore numbers and Phosphobacterial population in the rhizosphere soils of brinjal at different levels of phosphorus. The maximum root colonization percentage and spore numbers was recorded in treatment 7 (84.52), (129.00) and the minimum in control (52.53), (70.00) which was closely followed by treatment 10 (71.28), (124.00). The maximum Phosphobacterial population in treatment 7 (11.33) and the minimum in control (7.33) which was closely followed by treatment 6 (11.00).

9. Reference

- Ajimuddin (2002). Productivity and Quality of Sweet Basil (Ocimum Basilicum) as Influenced by Integrated Nutrient Management and Biofertilizers, M.Sc (Ag.). Dissertation Submitted to University of Agricultural Sciences, Bangalore.
- Gurumurthy, S.B., & Sreenivasa, M.N. (2000). Effect of VAM Fungus and P-solubilizer on Shisham. *Myforest*, 35, 73-80.
- Karthikeyan, B. (1994). The Response of Neem Azadirachta Indica to VA-Mycorrhizae Inoculation. M.Sc (Ag.). Dissertation Submitted to Tamil Nadu Agricultural University, Coimbatore.
- Lekberg, Y., & Koids, R.T. (2005). Arbuscular Mycorrhizal Fungi, Rhizobia available P and Nodulation of Groundnut (Archis hypogea L.) in Zimbabwe. Agriculture, Ecosystems & Environment, 110, 143-148.
- Prathiba, C.K., Alagawadi, A.R., & Sreenivasa, M.N. (1994). Establishment of Inoculated Organisms in Rhizosphere and their Influence on Nutrient Uptake and Yield of Cotton. *Karnataka Journal* of *Agricultural* Sciences, 8 (1), 22-27.
- Raigon, et al. (2008). Comparison of Eggplant Landraces and Commercial Varieties for Fruit Content of Phenolics, Minerals, Dry Matter and Protein. *Journal of Food Composition and Analysis*, 21 (5), 370-376.
- Sperber, J. I. (1958). Solubilization of Apatite by Soil Microorganisms Producing Organic Acids. *Australian Agricultural Research*, 9, 782-787.
- Zehra Ekin (2010). Resurgence of Safflower Utilization: A Global View. *Journal of Agronomy*, 4 (2), 83 87.