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Synergistic effect between phosphate solubilizing bacteria and vesicular-arbuscular mycorrhizal fungi on growth and p uptake in *Cajanus cajana* L. (Pigeon pea)

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Abstract: The influence of phosphate solubilizing bacterium (*Bacillus polymyxa*) and arbuscular mycorrhizal fungi (*Rhizophagus fasciculatus*) on growth and phosphorus uptake in *Cajanus cajana* were studied, in green house conditions. Experiments were carried out by using both sterilized and unsterilized garden soil (sandy loam). Mycorrhiza with P-solubilizing bacteria inoculated with sterilized soil produced significantly higher growth, dry matter and increase in nodule number and P uptake in shoot. Moderate or lower growth response was observed among the plants grown in unsterilized soil either PSB or AMF inoculation. On the contrary non-inoculated plants in sterilized garden soil did not showed meager growth and higher total P uptake. A synergistic effect was recorded with increased plant dry matter, nodule number and P uptake in the plants treated with both the inoculum and in sterilized soil.

Key words: Cajanus cajana; Bacillus polymyxa; Arbuscular mycorrhizal fungus; Rhizophagus fasciculatus; nodulation.

Introduction

Recent research on plant nutrition through PSB and AM fungi have amply demonstrated that these organisms play an important role in uptake of nutrient from the marginal soils. Research in the last three and half decades has established that dual inoculation of phosphate solubilizing bacteria and AM fungi stimulates plant growth. Phosphate solubilizing bacteria solubilize insoluble P and help plants to absorb and translocate more soluble phosphate (Lakshman Kadam *et al.*, 2004; Aruna and Lakshman, 2007; Lakshman, 2009).

The importance of other soil microorganism for plant growth has been well documented by many workers (Rekha Naik and Lakshman, 2010; Madgaonkar and Lakshman, 2013). *Cajanus cajana* L. is a largest producer and a second most important pulse in India. The crop is drought resistance, soil improving crop growing in semi-arid region. It is an important food grain in South India. This crop is oftenly used for counter hedge plant for controlling soil erosion. Synergistic studies were meager on this plant. Hence, the present study investigated the efficiency of phosphate solubilizing bacteria and arbuscular-mycorrhizal fungi on the growth and P uptake of pigeon pea plants.

Materials and Methods

The Green house experiments were conducted in the Post Graduate Department of Botany, Microbiology laboratory, Karnataka University, Dharwad-580003, India. *Bacillus polymyxa*, a phosphate solubilizing

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Dr. Chaitra B. Negalur, Microbiology laboratory, P.G. Department of Botany, Karnatak university, Dharwad, Karnataka, India. **E-mail:** <u>chainegalur@gmail.com</u> bacterium, was procured from the Department of Agricultural Microbiology, University of Agricultural Sciences, Dharwad-580005, India. The AM fungus *Rhizophagus fasciculatus* was multiplied with Sudan grass (*Sorghum vulgare* var. *sudaneese*) as a host plant. Soil based AM fungal inoculum was established and maintained in separate pot culture.

Seedlings were raised in earthen pots measuring 25 X 30cm diameter containing 4Kg of sterilized and unsterilized garden soil. The soil separately used in the experiments were a sandy loam with a pH of 6.8, EC 0.13 mmhas/cm, organic carbon 0.37%, available N 197 kg/ha, available K 205 kg/ha and available P 4.5 kg/ha. One week old seedlings of *Cajanus cajana* L. were inoculated with PSB and AM fungi around the root system. PSB (0.5mg) and mycorrhizal colonized chopped root bits (5g) and 7.5g soil of the host plants which consisted of spores (approximately 150/250g soil) and external hyphae was used as the inoculum singly or in combination. The treatments were as follows;

- 1. Un-inoculated control
- 2. Bacillus polymyxa alone
- 3. Rhizophagus fasiculatus alone
- 4. B. polymyxa + R. fasciculatus

The following observations were recorded on 60 days old seeding grown in green house conditions.



Plants growth parameters:

Plant height, dry weight of shoot and root were recorded after keeping plant material in hot air oven at 70°c constantly for 48hrs. Nodule number were recorded and P uptake in shoots was determined. The per cent of AM fungal colonization of Pigeon pea roots were estimated according to Phillips and Hayman (1970). The extra matrical chalamydospores were isolated by adopting wet sieving and decanting technique outlined by Gerdemann and Nicolson (1963). Phosphorus content of shoots was estimated by vanadomolybdate phosphoric yellow colour method outlined by Jackson (1973).

Results and Discussion

The data in terms of plant height, dry matter and P uptake in shoot and roots were recorded at 60 days of plants grow in both sterilized and unsterilized soils are presented (Tables 1 and 2). Pigeon pea plant biomass, per cent of root colonization.

Table 1: Effect of PSB and AM fungal inoculation on growth, dry matter, nodule number and P uptake of 60 days old *Cajanus cajana* L. in sterilized garden soil.

Treatments	Plants height (cm)	Shoot dry weight (g/pl.)	% Root colonization	Spore count/50 g soil	Nodule number/ plant	Root Dry weight (g/plant)	P uptake in shoot (ppm)
Un-inoculated (control)	19.7a	3.2b	-	-	0.51a	0.71b	37c
PSB	33.2c	9.3a	14.3a	31.3c	4.3b	0.86b	91b
AMF	36.8b	11.4d	52.4e	147.2b	6.9c	1.4d	94a
PSB+AMF	53.1g	14.5b	66.8a	158.5c	11.3d	1.7e	107c _

Table 2: Effect of PSB and AMF inoculation on growth, dry matter, nodule number and P uptake of 60 days old *Cajanus cajana* L. in unsterilized garden soil.

Treatments	Plants height (cm)	Shoot dry weight (g/pl.)	% Root colonization	Spore count/50 g soil	Nodule number/ plant	Root Dry weight (g/plant)	P uptake in shoot (ppm)
Un-inoculated (control)	7.6a	1.1b	-	-	0.4b	0.562a	27b
PSB	26.8b	3.7a	24.3b	33.2b	3.4a	0.968b	59c
AMF	29.3ab	4.9b	37.2d	98.0e	3.1c	0.971b	81b
PSB+AMF	31.5a	9.8e	49.6g	107d	7.1c	0.984b	92a

Spore population was greatly improved by simultaneous with the inoculation of PSB and AM fungi. This was been noted down in number of nodules, dry weight of plants and P uptake. This kind of progressive improvement was not recorded among the plants grown in unsterile soil. However, there was an increased plant height, biomass yield, P uptake, nodule number over the non-inoculated or control plant. This kind of variation has been observed in both inoculated and un-inoculated plants.

The bacterium Bacillus polymyxa in combination with Rhizophagus fasciculatus brought a significant increase in plant height, per cent AM fungal colonization, spore population and P uptake in plant grown in sterilized than unsterilized soils. The response was higher due to mixed (dual) inoculation than single inoculation indicating the synergistic effect of two organisms. The results indicated that dual inoculation could be used for better growth and production of root nodules in Cajanus cajana L. plants in greenhouse condition. The establishment of AM fungal symbiosis in plants is known to change physiological and biochemical properties of the host plant and these changes may alter the composition of the root exudates which play a key role in the modification of the microbial population quantitatively and qualitatively in the rhizosphere. (Dobbelaere et al., 2003; Lakshman, 2015). Interaction between AM fungi and certain rhizospheric microorganisms results in synergistic effect leading to

the growth and enhancement of various plant growth parameters (Bagyaraj, 1984; Gupta *et al.*, 1998a, 1998b; Gyaneshwar *et al.*, 1998; Suman *et al.*, 2003). Similar interactions between PSB and VAM observed in the present study are in confirmation with results of earlier workers (Paul and Sundara Rao, 1971; Barea *et al.*, 1975; Sattar and Gaur, 1989) on lavender, maize and lentils. The present study concludes that PSB under specific conditions mobilized-unavailable form of soil and fertilizer P and AM fungus provided soluble nutrients to plants, which improved plant nutrient uptake and growth and improvement of biomass yield.

References

- 1. Aruna S, Charanthimath and Lakshman HC. "AMF and PSB an effective tool to improve growth and nutrient status of *Tinospora cordifolia* Willd". *Nat. J. Life. Sci.* 4 (2007):37-40.
- Azcon-Aguilar, Gianinazzi-Pearson V, Fardean JC, Gianinazzi S. "Effect of vesicular-arbuscular mycorrhizal fungi and phosphate solubilizing bacteria on growth and nutrition of soybean in a neutral calcarious soil amended with ³²P ⁴⁵Ca-tricalcium phosphate". *Pl Soil* 93 (1986):3-15.
- Barea JM, Azcon R, Hayman DS. "Possible synergistic interaction between endogone and phosphate solubilizing bacteria in low phosphate soils". In: *Endogone and Phosphate* (Eds. F.E. Sanders, B. Mosse and P.B. Tinker) Academic Press, London, New York, (1975):409-418.

- Bayaraj DJ. "Biological interactions with VA-mycorrhizal fungi". In: VA-Myworrhiza, Eds. C.L. Powell and D.J. Bagyaraj. CRC press. Boco Raton (1984):131-153.
- Dobbelaere S, Vanderleyden J, Okon Y. "Plant growth promoting effects of diazotrophs in the rhizosphere". Crt Review plant Sci. 27(2003):107-149.
- 6. Gerdemann JW, Nicolson TH. "Spores of mycorrhizal endogone species extracted from soil by wet sieving and decanting". *Trans. Brit. Mycol. Soc.* 46(1963):235-244.
- Gupta A, Saxena AK, Gopal M, Tilak KVBR. "Bacterization of greengram with rhizosphere bacteria for enhanced plant growth". J. Sci. Indus. Res. 57(1998a): 726-731.
- Gupta A, Saxena AK, Gopal M, Tilak KVBR. "Enhanced nodulation of greengram by introduced Bradryhizobium when inoculated with plant growth promoting rhizobacteria". J. Sci. Indus. Res. 57(1998a):720-725.
- Gyaneshwar P, Naresh Kumar G, Parekh LJ. "Effect of buffereing on the phosphate- solubilizing ability of microorganisms". World J. Micobiol. Biotecnol. 14(1998):669-673.
- Jackson ML. "Soil Chemical Analysis". Prentice Hall of India (P) Ltd. New Delhi. (1973): 498.
- Lakshman HC. "AM Fungi with rhizosphere soil influence on Jatropha curcas L". Int. J. Plant Sci. 1.1(2009):120-123.
- 12. Laxman BK, Lakshman HC, Prakash A, Hosamani and Manjunath Doddamani. 2004. "Interaction between VAM and Phosphate solublizing bacteria (*Bacillus polymyxa*) on growth and seed yield of Foxtail millet (*Setaria italic* L. Beau) under graded levels of phosphorus". *National J. of Life Sciences.* 1.2(2004): 305-308.

- Madgaonkar SC, Lakshman HC. "Effect of AM fungi, Azotobacter and Phosphate solubilizing bacteria in improvement of *Amaranthus paniculatus* L." - a leafy vegetable. *Research Journal of Biotechnology*. 8.3(2013): 36-39.
- 14. Paul NB, Sundara Rao WVB. "Phosphate dissolving bacteria and VAM fungi in the rhizosphere of some cultivated legumes". *Plant soil*. 35(1971):127-132.
- Phillips JM, Hayman DS. "Improved procedure of clearing and staining of vesicular- arbuscular mycorrhizal fungi for rapid assessment of infection". *Trans. Brit. Mycol. Soc*, 55(1970):158-161
- Rekha RN, Lakshman HC. "Effect of AM fungi PGPR and different soil nitrogen sources to improve growth and yield of paddy (cv. Jaya)". *Int. J. of Plant Sci.* 5.1(2010): 165-169.
- Sattar MA, Gaur AC. "Effect of vesicular-arbuscular mycorrhiza and phosphate dissolving microorganisms on the yield and phosphate uptake of 2Lentil, *Lens esculenta* Moench". *Thai. J. Agri. Set.* 22(1989): 129-136.
- Suman DA, Bagyaraj DJ, Arpana J. "Interaction between *Glomus mosseae, Azotobacter chroococcum* and *Bacillus* coagulants and their influence as growth and nutrition of Neem". J. Soil. Biol. Ecol. 23(2003): 80-86.

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