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Symbiotic Response of Pigeon Pea to VA Mycorrhizal Fungi and Rhizobium

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ABSTRCT

Pulses play a very important role in agricultural systems providing high protein grain and herbage and also maintaining and improving soil fertility. The process of nitrogen fixation depends greatly on phosphorous availability. Vesicular arbuscular mycorrhizae (VAM) have been reported in wide range of crop plants stimulating plant growth, through uptake of phosphorus and other micronutrients. Interaction between mycorrhizal fungus (*Glomus* spp.) and *Rhizobium* spp. in Pigeon pea was studied using Leonard Jar, pot culture and field experiments. These experiments revealed that dual inoculation significantly enhanced total dry matter, nodule formation and productivity over that in control.

Keywords: Pigeon pea, Rhizobium, VA Mycorrhiza.

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INTRODUCTION

Pigeon pea play a significant role in agricultural systems providing high protein grains and herbage. It also play an important role in maintaining and improving soil fertility. Pigeon pea , because of it's restricted root system, can not absorbs sufficient soil phosphate. The scarcity of available phosphorus is a critical limiting factor, because it affects not only plant growth but also nodulation and nitrogen fixation[1]. The Vesicular- Arbuscular mycorrhizal fungi are known to increase nutrient uptake (Zn, Cu, S) particularly Phosphorus to the host plant[2]. The eukaryotic fungus supplies the excess phosphorous required for nitrogen fixation along with water and other nutrients[3]. Bagyraj reports the interaction between mycorrhiza and Rhizobium in relation to productivity of pulses[4].

MATERIALS AND METHODS

Seeds of Pigeon pea (Cajanus cajan (L) Mill. Var.ICPL-87) were obtained from College of Agriculture, Pune. Surface sterilized earthen pots (10 inch diameter) were filled with steam sterilized (20 lbs for 2 hrs.) soil. VA mycorrhiza spores multiplied on Jawer root stock having approximately 25 spores/10 g soil was used as inoculums. *Rhizobium leguminosarum* from Pigeon pea were isolated, authenticated and used as inoculum. The seeds were inoculated with *Rhizobium leguminosarum* by slurry method. Soil with 25 spores/10 g soil were added in a 2 inch deep hole made in the centre of the soil in each pot. Pregerminated seeds were sown. Alight irrigation was given immediately after sowing. Controls were with either of the symbionts and without both symbionts. Observations were recorded at harvest stage.

Modified Leonard jar assemblies were set up using amber coloured beer bottles. These bottles were cut cross wise into two halves and the upper was used to hold rooting medium and heavy specimen jars were used as reservoirs. The beer bottles were inverted in the reservoir and its mouth was plugged with absorbent cotton. A lamp wick was made to pass through the plug in such a way that part of it remained in the upper half of the assembly and the other part in the reservoir. The beer bottles were filled with washed oven dried river sand, The wick was placed centrally in the sand. The reservoir was field with one fourth strength of Reading's N_2 free solution and Hoagland solution respectively to saturate the rooting medium[5]. The whole assembly was set up and it was sterilized by autoclaving at 15 lbs pressure for two hours. After sterilization and cooling , pre germinated seeds of Pigeon pea were sown (one each) in each set. The sets includes Control, With Rhizobium alone, With mycorrhiza alone and with Rhizobium and mycorrhiza (25 spores / 10 g soil).

For field experiments 400 sq.feet ploughed and leveled plots were selected for the present study. Four parallel beds were prepared to sow seeds. Experiments were set up after onset of monsoon during kharif season. In the first row, seeds of Pigeon pea was sown without any treatment. For the second row seeds were mixed with the compatible strains of *Rhizobium leguminosarum* by slurry method. In the third row small pits were made at a distance of 1 ft each. Into these pits 10 g soil with approximately 25 spores were added. The fourth row was sown with seeds coated with Rhizobium and approximately 25 VAM spores/10 g soil in each pits. A uniform plant population was maintained by thinning the excess seedlings.

Observation on shoot and root biomass, nodulation, nodule weight, VAM sporulation , percentage infectivity were recorded at flowering stage of the crop by sampling five plants per row and average values along with standard deviation were recorded. Mycorrhizal colonization in roots was assessed by Phillips and Hayman method[6]. Yield was recorded at maturity. All experiments were run in triplicates.

RESELTS AND DISCUSSION

The result of pot culture experiments (Table 1), showed maximum increase in the dry matter and productivity in plants inoculated with both symbionts. The plants inoculated with VAM spores and Rhizobium had maximum growth with the dry weight of shoot 6.90 g and that of root 0.90 g. The number of nodules produced also was 83 where as the plants with Rhizobium alone had 56 nodules. Most of the nodules were large and pink, indicating active nitrogen fixation. Percentage infectivity was 73.33 in pots with dual symbionts and was only 55.34 in pots with VAM alone.

Table 1 : Pot Culture

Synergistic effects of VA Mycorrhiza and Rhizobium on Pigeon pea

Treatment	Plant Height Shoot + Root (cm)	Dry wt.of Plant (g)	No. Of pods	No. Of nodules	Percentage infectivity
Control	85.12 ± 4.49	02.77 ± 1.50	18.30 ± 1.60	--	--
R.leguminosarum	101.23 ± 3.78	04.40 ± 1.85	30.60 ± 2.47	56.00 ± 1.60	--
G. fasciculatum	96.26 ± 5.66	03.50 ± 1.50	26.00 ± 3.81	--	55.34 ± 2.05
R.leguminosarum + G. fasciculatum	119.56 ±4.49	7.80 ± 1.72	34.30 ± 3.24	83.60 ± 2.50	73.33 ± 2.33

(± indicates standard deviation) (Average of three readings)

In Leonard jar experiment, plants were slowly removed after two months growth and were studied to note the biomass and productivity. (Table 2) The length, fresh and dry weights of the shoots and roots, nodulations, percentage infectivity of VA mycorrhizal fungi were maximum in jars having dual symbionts. Addition of compatible strains of rhizobia and VAM in the vicinity of the root system of Pulses showed better growth[7]. The results of Leonard jar experiments indicated the benefits of dual symbiosis, This was also indicated by an increase in the number of nodules. This proved that VAM increased nodulation and accelerated nitrogen fixation leading to better growth and productivity.

Table 2 : Leonard jar experiment

Treatment	Plant Height Shoot + Root (cm)	Dry wt.of Plant (g)	No. Of nodules	Dry wt. Of nodules (mg)	Percentage infectivity
Control	60.43 ± 2.96	0.98 ± 0.14	--	--	--
R.leguminosarum	74.79 ± 3.65	1.99 ± 0.18	42.00 ± 3.26	66.60 ± 3.09	--
G. fasciculatum	57.89 ± 4.67	0.92 ± 0.18	--	--	61.66 ± 2.35
R.leguminosarum + G. fasciculatum	86.66 ±4.08.	2.12 ± 0.22	45.33 ± 2.05	70.60 ± 4.10	83.33 ± 2.35

(± indicates standard deviation) (Average of three readings)

In field experiments the inoculum present as natural soil flora was supplemented with slurry of compatible strains of Rhizobium and VAM spores. Considerable increase in the biomass and productivity was observed in field experiments (Table 3) compared to Pot experiments. The dry mass of plants sown in fourth row with seeds coated with Rhizobium and VAM spores was 33.87 g (control 24.70 g). While average number of pods were 203.3 per plants, control were growing in soil with natural inoculum of Rhizobium as will as VAM, even then the number pods produced were 48.3. The increase in the number of nodules from 18.00 to 27.66 was because of the addition of compatible strains of Rhizobium. With the addition of VAM alone, nodules increased to 18.6. However, when more inoculum of compatible strains of symbionts were added, there was a remarkable increase in biomass, nodulation, VAM infectivity, and productivity. This was followed by plants inoculated with Rhizobia alone. Plants inoculated with VAM alone and control did not have much growth, However, the growth with VAM alone was coparatively better, indicating that the nutrients required for plant growth and even fixed form of nitrogen is transported by mycorrhiza to the plants. Several positive indications have come forward in the case of Chick pea [5]

Table. 3 : Field experiment

Treatment	Plant Height Shoot + Root (cm)	Dry wt.of Plant (g)	No. Of pods	No. Of nodules	Percentage infectivity
Control	96.73 ± 4.97	24.70 ± 1.31	48.3 ± 6.40	18.00 ± 2.49	64.33 ± 3.65
R.leguminosarum	135.98 ± 5.05	32.75 ± 2.22	126.0 ± 7.10	27.66 ± 2.05	75.00 ± 8.71
G. fasciculatum	115.69 ± 5.05	31.20 ± 3.94	56.6 ± 9.03	18.66 ± 2.82	68.60 ± 4.11
R.leguminosarum + G. fasciculatum	142.83 ± 5.15	33.87 ± 3.65	209.3 ± 8.86	32.33 ± 3.05	87.40 ± 6.23

(± indicates standard deviation) (Average of three readings)

From the results obtained from pot culture, Leonard Jar experiments and field trails, concluded that VAM with Rhizobium substantially helped Pigeon pea crop and significantly increased their biomass and productivity as reported by Bagyaraj[5]. A complementary effects of Rhizobium was seen with the increase in VAM infection and sporulation.

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