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Response of Black Gram Crop to Rhizobacteria

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Abstract

Field experiments were conducted during summer seasons in two consecutive years (2020-21 and 2021-22) at Experimental Farm of College of Agriculture, Tripura to find out the best *Rhizobacteria* inoculants for stimulation of nodulation and increasing yield of blackgram crop. From the findings of two years' experiment, it can be noted that inoculation of *Rhizobium* significantly increased the number and dry mass of nodules (32.8 plant^{-1} and 35.8 mg plant^{-1} at 30 DAS), plant dry mass (26.8 g plant^{-1}) and grain yield (861.3 kg ha^{-1}) of black gram. *Azotobacter chroococcum* alone and with *Rhizobium* numerically favoured the nodulation (29.6 and 34.2 plant^{-1} respectively) in black gram. *Rhizobium* + *A. chroococcum* treatment produced significantly more plant dry mass (29.6 g plant^{-1}) and grain yield (930.6 kg ha^{-1}) of black gram over the uninoculated control (23.2 g plant^{-1} and 747.9 kg ha^{-1} respectively) and *Rhizobium* alone, respectively. *Bacillus* sp. in conjunction with *Rhizobium* were statistically comparable to *Rhizobium* alone inoculation in respect of nodule mass and grain yield. All the three inoculants together gave the 890.3 kg ha^{-1} black gram yield which was statistically comparable to *Rhizobium* + *A. chroococcum* in black gram. So, from the results of experiment, it can be concluded that dual inoculation of black gram seeds with *Rhizobium* + *A. chroococcum* promotes significant nodulation of black gram crop with in turn increases the plant biomass and seed yield of summer blackgram crop.

Keywords: *Bacillus*, Black gram, Inoculation, Nodulation, *Rhizobium*

Introduction

Seed treatment with effective *Rhizobium* inoculant is recommended for pulse crop to ensure adequate nodulation and N_2 fixation for maximum growth and yield of pulse crops. Many researchers have reported that rhizobacteria belonging to groups of phosphate solubilising bacteria (PSB) and plant growth promoting rhizobacteria (PGPR) influence the symbiosis particularly at early growth stages. Many of these rhizobacteria were found synergistic to *Rhizobium* and their co-inoculation with *Rhizobium* showed improvement in root nodulation, N_2 fixation, nutrient uptake and yield of pulse crops (Habete and Buraka, 2016) and (Otieno *et al*., 2009). Since dual inoculation practice is coming up with an important management in maximization of yield of pulse crops, it becomes worthwhile to study compatibility among different microorganisms and their inoculation effect under field conditions in order to identify promising combinations of such microorganisms for different pulse crops. Keeping the above objective in view, effect of dual inoculation of different rhizobacteria (*Azotobacter chroococcum* and *Bacillus* sp.*)* with *Rhizobium* was examined in black gram [*Vigna mungo* (L.) Hepper] crop under field conditions.

Materials and Methods

Inoculants of black gram *Rhizobium* spp., *Azotobacter* sp. and *Bacillus* sp. were obtained from State Agricultural Research Station, Government of Tripura.

Field experiments were conducted during summer seasons in two consecutive years (2020-21 and 2021-22) at Experimental Farm of College of Agriculture, Tripura. Characteristics of the experimental soils in the different years are given in Table 1. Treatments consisted of inoculation

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with *Rhizobium, Azotobacter chroococcum* and *Bacillus* sp., either alone or in combinations. The treatments were laid out in Randomized Block Design (RBD) in four replications in plots of 4 m × 3 m size. Seeds were treated with the desired inoculant(s) (at 25 g kg^{-1} of seed) as per treatments and sown in lines at 30 cm apart. Dual inoculation, wherever required, was done by mixing the required quantity of the inoculants at the time of seed treatment. The test crop variety of black gram was T-9. The black gram crop was raised following recommended agronomic practices with one pre-sowing irrigation.

Table 1: Soil properties of the experimental sites in two years

Soil properties	First year	Second year
Texture	Sandy loam	Sandy loam
Soil pH	5.42	5.47
Organic Carbon (%)	0.47	0.46
Total N (%)	0.041	0.043
Available N (kg ha^{-1})	322.67	327.17
Available P_2O_5 (kg ha^{-1})	9.12	9.87
Available K_2O (kg ha^{-1})	165.90	169.90

Randomly twenty plants were dug out care fully from each plot at 30, and 45 days after sowing (DAS). The soil was gently washed off from the roots with a water jet, nodules were removed and counted. Dry mass of the nodules and plants at each observation was recorded after drying the material at 80 °C for 72 h. Grain yield and plant dry mass were recorded at final harvest. The data were subjected to analysis of variance following *F*-test and the critical difference was determined at 5% level of significance.

Results and Discussion

Table 2 showed the pooled data of two years experimental results which clearly indicated that inoculation of black gram seeds with *Rhizobacteria* stimulated the nodulation and increased seed yield of crop. Dual inoculation of *Rhizobium* + *A. chroococcum* gave the highest number (34.2 $plant^{-1}$ and 33.5 $plant^{-1}$ at 30 and 45 DAS respectively) and dry mass of nodules (40.9 and 26.2 mg $plant^{-1}$ at 30 and 45 DAS respectively) over the years and was comparable to the combined inoculation of *Rhizobium, A. chroococcum* and *Bacillus* sp. (26.7 and 27.4 nodules $plant^{-1}$ at 30 and 45 DAS respectively) (Table 2). Dual inoculation of *Rhizobium* + *A. chroococcum* recorded the maximum plant dry mass (29.6 g $plant^{-1}$) and grain yield (930.6 kg ha^{-1}) of black gram in the different years was statistically similar to *Rhizobium* + *A. chroococcum* + *Bacillus* sp. and significantly better than *Rhizobium* alone. Dual inoculation with *Rhizobium* + *A. chroococcum* resulted significant number and dry mass of nodules than *Rhizobium* alone. Nevertheless, this treatment recorded significant increase of 10.4% in plant dry mass and 8.0% grain yield over *Rhizobium* inoculation. The results are in close conformity with the earlier reports that indicated significant effect of dual inoculation of *Rhizobium* + *A. chroococcum* on grain and straw yield of chickpea (Abdiev *et al*., 2019) and Wheat (El-Sawah *et al*., 2018) due to synergistic interaction. *A. chroococcum* is also known to secrete plant growth hormones in rhizosphere (Tilak *et al*., 2006) leading to more plant biomass and yield in the study. *Rhizobium* alone inoculation gave significant increase of 19.3-49.5% in nodule number, 106.9-61.7% in nodule dry mass, 15.5% in plant dry mass and 15.2% in grain yield over the control. Number and dry mass of nodules (32.8 $plant^{-1}$ and 35.8 mg $plant^{-1}$ at 30 DAS), plant dry mass (26.8 g $plant^{-1}$) and grain yield (861.3 kg ha^{-1}) by the treatment *Rhizobium* alone were also significantly more than the control. Such positive effects of *Rhizobium* inoculation in black gram have also been reported by (Otieno *et al*., 2009) and may be due to the presence of either ineffective and/or low population of native rhizobia in the soils. *A. chroococcum* treatment favoured the nodule and plant dry mass slightly and recorded 7.3% increase in grain yield over the control; however, there was no significant difference. *Bacillus* sp. alone was comparable to the uninoculated control in terms

Table 2: Effect of Rhizobacteria on nodule number, nodule mass, plant dry mass and grain yield of black gram (Pooled data of two years)

Treatment	Nodule number $plant^{-1}$		Nodule dry mass $plant^{-1}$ (mg)		Plant dry mass $plant^{-1}$ (g)	Grain yield (kg ha^{-1})
	30 DAS	45 DAS	30 DAS	45 DAS	at Harvest	at Harvest
Uninoculated (Control)	27.5	20.6	17.3	14.9	23.2	747.9
Rhizobium sp.	32.8	30.8	35.8	24.1	26.8	861.3
Azotobacter chroococcum	29.6	23.3	22.3	18.3	24.4	802.8
Bacillus sp.	25.7	20.9	17.8	15.8	24.0	755.1
Rhizobium + *A. chroococcu*m	34.2	33.5	40.9	26.2	29.6	930.6
Rhizobium + *Bacillus* sp.	28.6	30.4	31.5	22.0	24.8	811.7
A. chroococcum + *Bacillus* sp.	27.3	21.0	24.4	16.2	25.5	808.9
Rhizobium + *A. chroococcum* + *Bacillus* sp.	26.7	27.4	29.8	23.1	25.7	890.3
SEm ±	1.1	3.1	3.3	2.6	0.89	22.5
CD at 5%	3.2	9.4	9.6	7.9	2.70	68.3

of number and dry mass of nodules and it adversely affected the nodulation, compared to their individual performance, when inoculated with either *Rhizobium* or *A. chroococcum* probably due to its antagonistic effect on these organisms. This bacterium with *Rhizobium* also showed reduction in grain yield. Such antagonistic interaction between *Rhizobium* sp. and *Bacillus* sp. has also been reported by (Jimtha John *et al.*, 2016) due to synthesis of some toxic metabolites by *Bacillus* sp.

Conclusion

From the findings of the experiment, it can be concluded that dual inoculation of *Rhizobium* + *Azotobacter* is superior than inoculation of *Rhizobium* alone for increasing the yield of black gram and this combined artificial inoculation can be promoted to the farmers of Tripura for higher blackgram yield as well as improvement of soil health.

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