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Effect of dual inoculation of salt tolerant *Rhizobium* and PSB in cowpea

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Abstract
A pot culture experiment was conducted to evaluate the effect of salt tolerant *Rhizobium* and PSB on growth and nutrient uptake of Cowpea (cv. Phule Pandhari). The inoculation with isolates of *Rhizobium* and PSB in combimation showed significant increase in growth parameters viz. seed germination, plant height, number of branches per plant, dry matter weight, nitrogen and phosphorus uptake per plant. The inoculation of *Rhizobium* (RHB_5) x Phosphate solubilizing bacteria (PSB_4) recorded significantly more germination percentage, growth character, nitrogen, phosphorus uptake and microbial population in soil over absolute control.

Keywords: Salt tolerance, Rhizobium, PSB, Cowpea.

Introduction
In India, about seven million hectares of land is affected by salinity and alkalinity (Yadav, 1976). The problems of such soils are old, but they are spreading worse by development of irrigation system without adequate provision of drainage and aggravated by poor water management practices and unsound reclamation procedures. Imbalanced use of chemical fertilizers reduces the infiltration ability of the soil and increased the salt content. Nearly 40 per cent of 4 world's surface has salinity problems. The productivity of crops is greatly affected by salt stress. Highly alkaline (pH >8.0) soils tending to be high in NaCl, bicarbonate, carbonate, are often associated with high salinity.
Due to higher salt content the activity of *Rhizobium* and PSB strains is reduced. In such soils, *Rhizobium* and PSB strains tolerating high concentration of salt and yet capable of fixing nitrogen and solubilizing phosphorus more effectively are of importance in increasing its fertility.

Materials and Methods
A pot culture experiment was conducted at the Department of Plant Pathology and Agricultural Microbiology, College of Agriculture, Pune during 2011-2013. The lignite based inoculants of native salt tolerant *Rhizobium* and PSB (10^7cfug-1) isolates and the lignite based commercial *Rhizobium* and PSB (10^7cfug-1) obtained from BNF scheme, College of Agriculture, Pune as check were used to treat the seeds separately by slurry method @ 250 g per 10 kg seed. Such seeds were dried in shade and used for sowing immediately.
The sowing was carried out by dibbling, five seeds in each pot. Immediately after sowing a light irrigation was given. After ten days, thinning was done and maintained only three plants in each pot. The treatments offered were : T_1 : Rhizobium (RHB_5), T_2 : Rhizobium (RHB_3), T_3 : Phosphate Solubilizing Bacteria (PSB_4), T_4 : Phosphate Solubilizing Bacteria (PSB_4), T_5 : RHB_5 x PSB_4, T_6 : RHB_5 x PSB_8, T_7 : RHB_3 x PSB_4, T_8 : RHB_3 x PSB_8, T_9 : Commercial Rhizobium, T_{10} : Commercial PSB, T_{11} : Commercial *Rhizobium* X PSB, T_{12} : Absolute control.
Observations on different parameters viz, Seed germination (%), number of branches, plant height, Nodulation (at 45 DAS), dry weight of root and shoot, PSB population in pot soil at initial, flowering and at harvest. Expriment was arranged in a completely randomized block design. The nitrogen and phosphorus uptakes were determined.

Results and Discussion
The treatments differences with respect of Plant height, number of root nodules / plant, total dry weight / plant, population of *Rhizobium* and PSB, available N and P in soil, N and P uptake per plant were statistically significant at harvest. Treatment T_5 (RHB_5 x PSB_4) was significant over all other treatments. The treatment T_6 (RHB5 x PSB_8) recorded the second

best treatment for almost all parameters followed by treatment T_7 (RHB_3 x PSB_4).

The increase in germination percentage due to inoculation of different salt tolerant *Rhizobium* and PSB isolates may be due to ability to suppress the growth of antagonists present in soil and on seed coat and release of plant growth promoting substances around seed sown. Rudresh *et al.* (2005) reported the effect of combined inoculation of salt tolerant *Rhizobium* and phosphate solubilizing bacteria in increased germination of chickpea.

Rudresh *et al.* (2005), Afzal and Bano (2008), Dastager *et al*. (2010), Muhammad *et al*. (2013) reported that, the effect of combined inoculation of *Rhizobium* and phosphate solubilizing bacteria increased plant height in chickpea.

The nodulation was further improved due to more availability of soluble phosphorus due to phosphate solubilizing microorganisms used as seed inoculation. Hashem *et al.* (1998), Rai and Singh (1999), Akhtar *et al.* (2009) also reported significantly greater nodulation due to salt-tolerant *Rhizobium* strains. Tomar *et al.* (2001) on effect of *Rhizobium* and PSB (phosphate solubilizing bacteria) inoculation, with and without P, increased nodulation in blackgram.

Neves *et al.* (1982), Alagawadi and Gaur (1988) reported that combined inoculation of *Rhizobium* and *P. striata* or *B. polymyxa* increased the dry matter content. Afzal and Bano (2008), Vikram and Hamzehzarghani (2008), Muhammad *et al.* (2013) observed that co-inoculation with rhizobia and plant growth promoting rhizobacteria (PGPR) improved the total dry matter (TDM).

Table 1: Effect of various Combination of Rhizobium and PSB on Cowpea.

Treatments	Germination %	Plant height (cm)	No. of branches plant^{-1}	No. of Nodules plant^{-1}	Total Dry Weight (g plant^{-1})	Nitrogen uptake (g plant^{-1})	Phosphorus uptake (g plant^{-1})
T_1	**86.67**	76.07	3.67	48.00	11.05	0.32	0.07
T_2	83.33	76.07	3.33	48.00	10.95	0.31	0.07
T_3	80.00	76.00	3.67	38.00	10.83	0.30	0.09
T_4	83.33	75.67	3.67	37.67	10.61	0.30	0.08
T_5	96.67	80.00	4.00	53.33	13.33	0.39	0.15
T_6	93.33	78.67	4.00	52.33	12.77	0.37	0.12
T_7	93.33	77.97	3.67	52.00	12.50	0.36	0.13
T_8	90.00	77.77	3.67	49.67	12.13	0.35	0.11
T_9	80.00	75.33	3.33	46.33	10.55	0.30	0.06
T_{10}	83.33	75.00	3.33	37.00	10.40	0.29	0.08
T_{11}	83.33	77.50	3.33	49.00	12.03	0.34	0.11
T_{12}	70.00	67.67	3.00	35.67	8.90	0.20	0.05
SE + -	3.10	1.07	0.30	0.75	0.70	0.02	0.01
CD at 5 %	9.31	3.12	NS	2.18	2.06	0.06	0.02

The N and P uptake in cowpea crop due to seed inoculation with *Rhizobium* and PSB isolates increased significantly over the absolute control. Seed inoculation with *Rhizobium* and PSB isolates increase the nitrogen fixation and its accumulation in plant system which increased the plant biomass, nitrogen content in biomass and reflected in its uptake. Phosphorus is the integral part of nitrogenase enzyme therefore; increase in phosphorus in rhizosphere increases the nitrogen fixation by *Rhizobium*. Hence *Rhizobium* coupled with PSB increased the nitrogen fixation there by plant biomass. Rashid *et al.* (1999) reported that the increase in nitrogen uptake by plant due to inoculation of *Rhizobium* and PSB. Govindan and Thirumurugan (2005), Rudresh *et al.* (2005), reported that the effect of *Rhizobium* and phosphate-solubilizing bacteria (PSB) significantly increase the NPK uptake. Son *et al.* (2006), Mahdi *et al.* (2011) reported that, use of phosphate solubilizing bacteria as inoculants increases phosphorus uptake.

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