Influence of Efficient Strain of PSB on Growth and Yield of Maize (*Zea mays* L.) Under Black Cotton Soil Condition

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A field experiment was conducted with local strains of phosphobacteria (PSB-S-2) in order to improve the growth and yield of maize in black cotton soil. This PSB isolate performed better with combination of 75 % recommended dose of phosphorus fertilizer with respect to stimulation of beneficial rhizosphere micro flora *viz.*, Bacteria, Fungi, Actinomycetes, Azotobacter and PSB as compared to rest of the treatment combinations. However, it was found on par to 100 % RDP in combination with PSB and 100 % RDP alone. The shoot P concentration was significantly high and it increased with increase in the age of the host due to inoculation of efficient isolate of PSB in combination with 75 % RDP, the plant growth parameters *viz*., plant height, number of leaves, shoot and root biomass and yield of maize was also found significantly increase over other treatments when it was inoculated with 75 % RDP + PSB.

Key words: Biofertilizer, RDP, Maize, PSB, Phosphobacteria.

Maize (*Zea mays* L.) is one of the important cereal crop next to wheat and rice in the world. In India, maize ranks fourth after rice, wheat and sorghum and is cultivated in an area of 8.71 million hectare with an annual production of 21.57 million tones and with a productivity of 24.76 q ha^{-1} (Anon, 2012). Among the three major plant nutrients, phosphorus next only to nitrogen is important in crop production. It is directly involved in energy transfer and protein metabolism in plant. An adequate supply of phosphorus in the early growth stage of the plant is important in initiation of flower primordia for its reproductive parts (Tisdale and Nelson, 1968). Deficiency of phosphorus leads to development of purple pigment on leaves, stunted plant growth and delay in plant development.

Price hike in chemical fertilizers, the environmental pollution caused by them and also depletion of fossil fuel resources; raw material for chemical fertilizer have called for more attention to the use of bio-inoculants to supplement chemical fertilizers. With this regard PSB bio-inoculant was selected (PSB-S-2) which has already proved their efficiency in terms of *in vitro* N_2 fixation, IAA and GA production potential was used in the present study with a view to select environment friendly microbial inoculants capable of supplying phosphorus and improving the growth and yield of maize under field condition.

MATERIALS AND METHODS

The field experiment was conducted during *Kharif* season 2013, at college of agriculture, Raichur (Karnataka).

The pure culture of PSB-S-2 and reference PSB was collected from the Dept. of

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Agric. Microbiology, college of Agriculture, Raichur. PSB strain was grown on Pikovskaya's broth for 48 hr to 72 hr to achieve 10^9 cells/ml of the broth. The broth culture was mixed with the lignite in such a way that the moisture content was maintained at 30 % and cured for 24 hr at room temperature. Later 24 hr of curing, the microbial load in the product was determined which was estimated to be 10^9 cells g^{-1} of the inoculants. And the inoculant was used for treating of maize seeds.

Maize seeds of variety Hema were used for sowing. The observations were recorded at the intervals of 30 DAS, 60 DAS, 90 DAS and At harvest of the crop. The treatments **(Table 1)** given to maize were replicated thrice in randomized completely block design.

The initial bacterial, fungal, actinomycetes, azotobacter and phosphobacterial population in the rhizosphere soil were isolated by following serial dilution agar plate method. Five plants were randomly selected from each treatment during different growth stages for recording various observations on growth and yield parameters. The plant samples were also analysed for shoot P concentration following standard methods.

RESULTS AND DISCUSSION

Biological properties of the experimental plots

The initial soil microbial population (Bacteria, fungi, actinomycetes, Azotobacter and PSB) was found non significant among all the treatments. at 30 DAS the microbial population was significantly increased rhizosphere soil, with increase in the age of the host the microbial population was increased up to 60 DAS (Bacteria 2.44 X 10^9 cfu g^{-1} of dry soil, Fungi 4.73 X 10^5 cfu g^{-1} of dry soil, Actinomycetes 4.60 X 10^5 cfu/g of dry soil, Azotobacter 6.23 X 10^4 cfu g^{-1} of dry soil and PSB 6.60 X 10^4 cfu g^{-1} of dry soil), The population decreased at harvest of the crop (Bacteria 2.10 X 10^9 cfu g^{-1} of dry soil, Fungi 3.93 X 10^5 cfu g^{-1} of dry soil, Actinomycetes 4.37 X 10^5 cfu g^{-1} of dry soil, *Azotobacter* 5.35 X 10^4 cfu g^{-1} of dry soil and PSB 5.90 X 10^4 cfu g^{-1} of dry soil). Jones and Sreenivasa (1992) also reported that, decrease in the population of bacteria, fungi, actinomycetes, Azotobacter and PSB in the rhizosphere soil of sunflower inoculated with *Glomus fasciculatum* on 83 days after sowing. It is likely that amount of root exudates and its nutrient status might have decreased with the advancement in the age of the host especially during harvest which, in turn, perhaps caused a decreased in the rhizosphere microbial population Gajda and Martyniuk (2004). The microbial population in the rhizosphere soil of maize were significantly highest due to inoculation of efficient strain of

Table 1. Influence of efficient strain of Phosphate solubilising bacterium on the population of bacteria and fungi in the rhizosphere soil of chilli at different growth stages

Treatments	Bacteria cfu x 10^9 / g				Fungi cfu x 10^5 / g			
	30 DAS	60 DAS	90 DAS	At harvest	30 DAS	60 DAS	90 DAS	Atharvest
T_1 - Control	1.37	1.56	1.53	1.50	2.03	2.17	2.13	2.07
T_2 - PSB	1.46	1.62	1.60	1.58	2.23	2.50	2.40	2.33
T_3 - Reference PSB	1.43	1.59	1.56	1.52	2.10	2.27	2.20	2.17
T_4 - RDP 100%	1.71	2.33	1.95	1.90	3.47	4.27	3.97	3.67
T_5 - RDP 100% + PSB	1.75	2.37	2.05	2.01	3.53	4.30	4.07	3.80
T_6 - RDP 75 %	1.58	1.75	1.71	1.68	2.67	3.17	3.00	2.87
T_7 - RDP 75 % + PSB	2.09	2.44	2.13	2.10	3.67	4.73	4.40	3.93
T_8 - RDP 50 %	1.55	1.67	1.65	1.63	2.50	2.93	2.70	2.60
T_9 - RDP 50 % + PSB	1.62	1.82	1.76	1.72	2.97	3.53	3.27	3.10
S.Em±	0.14	0.06	0.06	0.08	0.08	0.22	0.15	0.09
C.D. at 5%	0.39	0.18	0.18	0.23	0.24	0.64	0.43	0.28

Values are mean of three replications DAS – Days after sowing

PSB (PSB-S-2) with RDP 75 % as compared to rest of the treatment combinations and treatment having standard PSB strain and uninoculated control. However, T_5 (RDP 100 % + PSB) and T_4 (RDP 100 %) was on par to T_7 (RDP 75 % + PSB) (Table 1, 2 respectively). It's a well known fact that soil microbial activity and nutrient concentration are found in the rhizosphere area (Brown, 1975). Any change in this region might affect the rhizosphere microflora and in turn plant growth Oswald and Ferchau (1968). Rovira (1956) and Peterson (1958) reported that host plant and its growth stages are principle determinates of rhizosphere microflora. Kundu and Gaur (1980) observed a synergetic interaction between Azotobacter and Phosphate solubilising bacteria when the two organisms were inoculated together in cotton.

Influence of efficient strains of PSB on the plant growth and yield parameter of maize.

Plant height increased with the increase in the age of the plant 30 DAS (43.15 cm), 60 DAS (154.47 cm), 90 DAS (177.87 cm) and at harvest (178.43 cm) plant height increased due to inoculation of PSB (PSB-S-2) with 75 % RDP compared to rest of the treatment combinations (Table 3). Plant height increased because of nutrient availability, increase nutrient uptake (N, P and K) and enzyme activity in the rhizosphere soil as well as rhizoplane. Insoluble inorganic

Table 2. Influence of efficient strain of PSB on the population of Actinomycetes, Azotobacter, PSB in the rhizosphere soil of maize at

Treatments	Actinomycetes cfu x 10^5 / g				Azotobactercfu x 10^4 / g				PSBcfu x 10^4 / g			
	30 DAS	60 DAS	90 DAS	At harvest	30 DAS	60 DAS	90 DAS	At harvest	30 DAS	60 DAS	90 DAS	At harvest
T_1 - Control	2.54	2.80	2.60	2.58	2.25	3.83	3.50	2.64	2.17	3.03	2.70	2.30
T_2 - PSB	2.82	3.20	3.00	2.90	3.15	4.51	4.13	3.76	3.00	3.80	3.63	3.33
T_3 - Reference PSB	2.66	3.10	2.80	2.70	2.86	4.14	3.85	3.37	2.60	3.40	3.20	2.90
T_4 - RDP 100%	4.00	4.30	4.30	3.90	4.46	5.73	5.37	5.00	5.17	6.17	6.00	5.43
T_5 - RDP 100% + PSB	4.10	4.40	4.40	4.10	4.53	5.81	5.60	5.28	5.33	6.30	6.23	5.70
T_6 - RDP 75 %	3.30	3.67	3.50	3.40	3.88	5.18	4.88	4.54	3.73	4.40	4.20	3.90
T_7 - RDP 75 % + PSB	4.20	4.60	4.53	4.37	4.73	6.23	5.80	5.35	5.70	6.60	6.37	5.90
T_8 - RDP 50 %	3.07	3.37	3.017	3.310	3.48	4.87	4.37	4.51	3.23	4.07	3.70	3.30
T_9 - RDP 50 % + PSB	3.57	3.80	3.70	3.60	4.05	5.53	5.13	4.76	4.23	4.70	4.50	4.33
S.Em±	0.09	0.18	0.09	0.18	0.16	0.17	0.17	0.13	0.18	0.20	0.14	0.17
C.D at 5%	0.25	0.54	0.25	0.52	0.46	0.49	0.49	0.37	0.53	0.58	0.42	0.48

Values are mean of three replications DAS - Days after sowing

Table 3. Influence of efficient strain of PSB on the plant height, shoot and root biomass of maize at different growth stages

Treatments	Plant height (cm)				Shoot biomass (g / plant)				Root biomass (g / plant)			
	30 DAS	60 DAS	90 DAS	At harvest	30 DAS	60 DAS	90 DAS	At harvest	30 DAS	60 DAS	90 DAS	At harvest
T_1 - Control	32.13	135.35	156.27	157.27	76.67	152.33	153.00	154.00	21.00	45.00	56.00	58.33
T_2 - PSB	36.32	139.10	158.93	159.27	82.67	157.00	161.00	161.67	24.67	48.00	60.00	61.00
T_3 - Reference PSB	34.30	137.83	158.17	159.00	79.67	154.33	157.33	158.00	21.00	46.33	57.67	59.67
T_4 - RDP 100%	42.77	153.00	176.27	176.53	94.00	164.00	170.63	171.33	36.00	57.67	65.67	68.00
T_5 - RDP 100% + PSB	43.05	153.47	177.10	177.60	94.33	166.67	171.00	172.00	36.67	58.00	66.33	68.33
T_6 - RDP 75 %	38.90	144.70	164.37	165.60	91.00	161.67	166.00	166.67	31.00	52.67	62.00	63.67
T_7 - RDP 75 % + PSB	43.51	154.47	177.87	178.43	95.67	168.33	172.33	173.00	37.00	58.67	67.00	69.33
T_8 - RDP 50 %	37.33	142.13	164.27	165.40	87.00	162.00	165.00	166.00	27.00	50.00	61.67	62.00
T_9 - RDP 50 % + PSB	39.84	148.77	167.87	169.13	92.00	163.00	168.00	168.67	31.67	53.00	63.33	65.33
S.Em±	1.13	1.05	1.17	1.14	1.26	1.50	0.64	0.68	0.76	1.21	1.03	0.75
C.D at 5%	3.30	3.06	3.42	3.32	3.69	4.37	1.86	2.00	4.46	3.53	3.02	2.19

Values are mean of three replications DAS – Days after sowing phosphate compounds are made available to the plants when PSB was inoculated into soil. The increased cell elongation and multiplication due to enhanced nutrient uptake by plants following inoculation of P solubilising bacteria may have caused the increased plant height Black (1968). Iman (2008) reported that the inoculation of the efficient PSB strains significantly increased the plant height up to 81%. The obtained results in our studies is agreed with those of Sherif *et al.* (1997) who mentioned that phosphate dissolving bacteria posses to the ability in bring a insoluble phosphate in soluble forms by secreting organic acids which lower the pH and bring about the dissolution of bonds of phosphate and render then available for growing plants.

Shoot and root biomass of maize plant was significantly higher in treatment T_7 (RDP 75% + PSB) followed by T_5 (100% RDP + PSB) and T_4 (RDP 100 %) however T_5 and T_4 was on par to T_7. Both the treatment showed increase in shoot and root biomass when it compared with the control. The increase in shoot and root biomass due to inoculation of PSB (PSB-S-2) with 75 % RDP has been well established Vassilev *et al.* (2006). Similar results of increase in yield parameters were because of several factors such as release of plant growth promoting substances Madhaiyan *et al.* (2004). Similarly Lin (2002) recorded increase in the shoot and root biomass by 125 per cent. Effect of plant growth promoting rhizobacteria (PGPR) including phosphate solubilising bacteria (PSB) as biofertilizers as a sustainable solution to improve plant nutrients and production Vessey (2003). Similar increase in root biomass of canola seedlings due to inoculation of *Pseudomonas putida* was observed by Lifshitz *et al.* (1987) who attributed such an increase in root biomass to the orthophosphate uptake by the inoculated plants. Khamparia (1995) was of the opinion that the positive effects of PSB are due to secretion of different organic and inorganic acids as well as phytase enzyme which act upon the residual insoluble phosphate and convert them into orthophosphates very near to root surface and thus make it available to crops.

The inoculation of efficient PSB isolates showed significantly increased the grain yield of maize than control and reference PSB inoculation alone (Table 4). The cob weight and grain yield due to inoculation of efficient isolate of PSB + 75 % RDP showed higher cob weight and grain yield as compared to other treatment combinations, however 100 % RDP with PSB and 100 % RDP alone was on par to T_7. Combination of RDP with PSB treated plot showed increased grain yield over the application of RDP alone and control. The results are in agreement with Wu *et al.* (2005) who recorded increase in yield significantly in maize crop and also improve soil properties such as organic content due to co inoculation of PSB. The results are also comparable with Balasubramanian and Subramanian (2006), whose studied the increase rice grain yield in a field experiment due to effect of silicate solubilising bacteria recorded 5218 kg ha grain yield than control 4419 kg per ha.

Table 4. Influence of inoculation efficient strain of PSB on the grain yield of maize

Treatments	Yield (kg / ha)
T_1 - Control	4972.67
T_2 - PSB	5084.87
T_3 - Reference PSB	5054.01
T_4 - RDP 100%	5941.35
T_5 - RDP 100% + PSB	5949.07
T_6 - RDP 75 %	5424.38
T_7 - RDP 75 % + PSB	5964.50
T_8 - RDP 50 %	5293.20
T_9 - RDP 50 % + PSB	5594.13
S.Em±	14.12
C.D at 5%	41.22

Table 5. Influence of inoculation efficient strain of PSB on the shoot P concentration

Treatments	Shoot P concentration (%)			
	30 DAS	60 DAS	90 DAS	At harvest
T_1 Control	0.31	0.33	0.39	0.39
T_2 PSB	0.35	0.38	0.42	0.42
T_3 Reference PSB	0.33	0.35	0.41	0.41
T_4 RDP 100 %	0.44	0.46	0.52	0.52
T_5 RDP 100% + PSB	0.45	0.47	0.53	0.53
T_6 RDP 75 %	0.39	0.42	0.48	0.48
T_7 RDP 75 % + PSB	0.46	0.48	0.54	0.54
T_8 RDP 50 %	0.38	0.41	0.46	0.46
T_9 RDP 50 % + PSB	0.42	0.45	0.51	0.51
S.Em±	0.01	0.01	0.01	0.02
C.D. at 5%	0.03	0.02	0.02	0.06

The plants that received PSB inoculations along with RDP in the present study showed higher Shoot P concentration of the plant over RDP alone and control (Table 5). The shoot P concentration was significantly higher in maize plants and increased with increase in the age of the host up to 90 DAS and then it decreased slightly. Shoot P concentration at 30 DAS (0.46 %), 60 DAS (0.48 %), 90 DAS (0.54 %) and At harvest (0.52 %) due to inoculation of efficient isolate of PSB with 75 % RDP, which was followed by treatment which received 100 % RDP with PSB and 100 % RDP alone. However, T_5 (RDP 100 % + PSB) and T_4 (100 % RDP) was on par to T_7 (RDP 75 % + PSB). The increased Shoot P concentration due to PSB are in confirmation with the earlier findings that phosphate solubilizers increase the shoot P content and uptake in several crop plants (Azcon *et al.*, 1976; Raj *et al*., 1981; Kucey, 1988; Asea *et al.*, 1988; Gaind and Gaur, 1991; Anthoniraj *et al.*, 1994; Pal, 1998; Appanna, 2001; Kadapatti, 2001). The increase in shoot P concentration by the efficient PSB isolates over reference strains indicate the superiority of the isolate to establish and perform better in field conditions. Gaur (1972) reported that the significant increase in the yield and shoot P concentration of wheat due to inoculation of P solubilising organisms like *Pseudomonas striata* and *Aspergillus awamori* in combination with rock phosphate.

In conclusion, inoculation of local isolates of phosphate solubilising bacteria (PSB-S-2) improved the plant growth and yield parameters such as plant height, root and shoot biomass, shoot P concentration and grain yield of maize significantly over the control. These local strains would be potential microbial inoculants for the local farmers as it is evident from the experimental results.

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