

Associative effect of nitrogen-fixing and phosphate-solubilizing bacteria in rainfed soybean (*Glycine max*) grown in Vertisols*

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Phosphorus promotes N_2 fixation in legume crops and is vital for photosynthesis, energy transfer and formation of sugars (Gaur 1990). Legumes need high amount of P in readily available form around their roots for rhizobia and the host plant. Only a small fraction of phosphate fertilizer is utilized by crops, while remaining portion of applied P gets fixed in the soil or rendered unavailable to crop plants (Mandal and Khan 1972). Rock phosphate being available in plenty in the country is good source of P for acid soils, but it is ineffective in neutral to alkaline soils (Awasthi *et al.* 1977). Continuous efforts have been made to increase the efficiency in soil with pH more than 7 by adding P-solubilizing micro-organisms (Gaur 1990). In the present study, an attempt was made to explore the possibility of direct use of rock phosphate as a P fertilizer and increase the efficacy of superphosphate with the help of P-solubilizing bacteria (*Pseudomonas striata*) on soybean [*Glycine max* (L.) Merr.] under rainfed condition in Vertisols.

A field experiment was carried out during the rainy seasons (*khari*) of 1988-97 (10 years) on Vertisols (typic chromustert), having sand 10.5%, silt 35.3%, clay 54.2%, with 7.4 pH, electrical conductivity 0.73 dS/m, organic carbon 0.49%, available N 203.6, P 5.63 and K 383 kg/ha, cation-exchange capacity 44.0 Cmol (p+)/kg soil. The treatments were: control; inoculation with phosphate-solubilizing bacteria (PSB); superphosphate @ 26.4 kg P/ha; superphosphate @ 13.2 kg P/ha; superphosphate @ 13.2 kg P/ha + PSB inoculation; rock phosphate @ 13.2 and 26.4 kg P/ha alone as well as with PSB inoculation. Inoculation with *Bradyrhizobium japonicum* was common for all the treatments. For co-inoculation, phosphate-solubilizing bacteria and *Bradyrhizobium japonicum* inoculants were applied in equal amount (each of 5g/kg seed) as seed treatment. A basal dose of N @ 20 and K @ 33.2 kg/ha

was applied. Rock phosphate [$Ca_{10}(PO_4)_6F_2$] Ihabual grade, citrate soluble, contained 28.6% total P_2O_5 , 41.84% CaO, 0.18% K_2O ; 0.07% SO_4 ; 3% F; 0.75% CO_3 ; 1.92% Al_2O_3 ; 19.65% Si_2O_2 ; 0.05% MgO and 0.04% Na_2O , was also applied at the time of sowing. The experiment was laid out in randomized block design with 3 replications. The 'JS 75-46' soybean was sown in 4.0 m $\times$ 2.75 m plots at 0.4 m distance between rows and 0.05 m between the plants with 0.4 million/ha plant population. At harvest the seed yield was recorded.

Agronomic efficiency, phosphorus-use efficiency, availability coefficient ratio, relative agronomic efficiency of rock phosphate and relative economic efficiency were calculated as per Mole *et al.* (1982), Manickam and Ramaswami (1985), Mishra and Gupta (1978), Engelstad *et al.* (1974) and Black and Scott (1956) respectively. Economics of the treatments was also computed.

The average rainfall, maximum and minimum temperature during the crop season (June-October) were 743.9 mm, 30.9°C and 19.96°C in 1995; 1437.2 mm, 32.21°C and 19.24°C 1996 and 1021.7 mm 31.27°C and 19.18°C in 1997 respectively.

A significant increase was observed in mean seed yield of soybean with all the treatment as compared to control except rockphosphate alone (either of dose). The increase with superphosphate @ 13.2 kg P/ha, rock phosphate @ 13.2 and 26.4 kg P/ha with PSB inoculation was 5.52, 15.3 and 10.85% compared with superphosphate @ 13.2 kg P/ha, rock phosphate @ 13.2 and 26.4 kg P/ha alone respectively. The seed yield recorded with recommended dose of P through superphosphate @ 26.4 kg P/ha was maximum, but it was at par with half dose of superphosphate @ 13.2 kg P/ha with PSB inoculation. The response of applied P can be expected in low phosphorus soil as reported by Gaur (1990) and Bagavathi *et al.* (1997). The increase in yield with PSB inoculation was due to the production of organic acids by phosphorus-solubilizing microbes (PSM) which bring about dissolution of bound form of P (Gaur

* Short note

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Table 1 Nodulation, shoot weight, seed yield, profitability of soybean, average total P uptake, phosphorus-use efficiency (kg/ha), relative agronomic efficiency (RAE), relative economic efficiency (REE), availability coefficient ratio (ACR) and soil P as influenced by treatment (pooled over 10 years 1988-97)

Treatment	Nodules/ plant	Nodule dry biomass (mg/plant)	Shoot dry biomass (g/plant)	100- seed weight (g)	Seed yield (tonnes/ ha)	Average total P-uptake pattern (seed+ straw) (kg/ha)	Net profit (Rs/ha)	Benefit:cost ratio	Yard stick value (kg/ha)	P-use efficiency (kg/ha)	RAE of rock phosphate (%)	REE of rock phosphate (%)	ACR of rock phosphate (%)	Average available soil-P- after crop harvest (kg/ha)
Control	29	140.4	3.78	11.24	1.63	8.9								5.1
Phosphate - solubilizing bacterial inoculation (PSB)	42	200.8	5.39	12.16	1.79 (9.8%)	10.8	1 595	29.0						6.8
Superphosphate @ 26.4 kg P/ha	52	249.1	6.73	12.28	1.99 (22.1%)	14.3	2 560	2.35	6.07	6.0				9.8
Superphosphate @ 13.2 kg P/ha	45	186.1	6.38	11.57	1.88 (15.3%)	12.6	1 982	3.56	8.47	8.33				7.3
Superphosphate @ 13.2 kg P/ha+	56	202.0	6.82	11.81	1.95 (22.1%)	13.7	2 967	4.84	12.27	10.66				8.6
PSB inoculation	36	139.6	4.64	11.28	1.64 (0.6%)	9.3	(-)-1.27	(-)-1.27	0.37	0.33	87.09	391.90	0.043	6.0
Rock phosphate @ 13.2 kg P/ha	47	169.1	5.56	11.73	1.88 (15.3%)	11.6	2 317	12.04	8.37	0.33	87.31	392.89	0.310	8.8
PSB inoculation	35	193.7	4.93	13.22	1.74 (6.7%)	10.2	870	3.48	1.87	1.83	94.14	423.63	0.682	6.5
Rock phosphate @ 26.4 kg P/ha	42	258.9	5.75	12.55	1.93 (18.4%)	13.9	2 705	8.87	5.02	5.00	96.79	45.55	0.825	8.4
PSB inoculation+														0.61
LSD (P=0.05)	8.34	43.04	1.62	0.52	0.165	0.57								

Data in parentheses indicate the increase in yield (%) over the control. Net return based on cost of PSB culture @ Rs 37.50/ha; superphosphate @ Rs 18/kg P₂O₅; rock phosphate @ Rs 4/kg P₂O₅; cost of culture application (1/4 labour/day/ha) @ 12.50/ha; fertilizer application (1 labour/day/ha) @ Rs 50/ha; soybean Rs 9.50/kg

*Initial available soil P 5.63 kg/ha

Table 2 Balance sheet of average available soil P (kg/ha) after soybean crop

Treatment	Initial soil P (1)	P-added through fertilizer (2)	P-removed by crop (3)	Difference (4)=(2-3)	Available soil P at initiation + (P added-removed) (5)=(1+4)	Available P after harvest (6)	Net P left in the soil (7)=6-(1+2)	Loss - or gain (+) of (8)=(6-5)
Control	5.6		8.9	(-) 8.9	(-) 3.3	5.1	(-) 0.5	(+) 8.9
PSB inoculation	11		10.8	(-) 10.8	(-) 5.2	6.8	(+) 1.2	(+) 120
Superphosphate @ 26.4 kg P/ha	11	26.4	14.3	(+) 12.1	17.7	9.8	(-) 22.2	(-) 7.9
Superphosphate @ 13.2 kg P/ha	11	13.2	12.6	(+) 0.6	6.2	7.3	(-) 11.5	(+) 1.1
Superphosphate @ 13.2 kg P/ha + PSB inoculation	11	13.2	13.7	(-) 0.5	5.1	8.6	(-) 10.2	(+) 3.5
Rock phosphate @ 13.2 kg P/ha	11	13.2	9.3	(+) 3.9	9.5	6.0	(-) 12.8	(-) 3.5
Rock phosphate @ 13.2 kg P/ha + PSB inoculation	11	13.2	11.6	(+) 1.6	7.2	8.8	(-) 10.0	(+) 1.6
Rock phosphate @ 26.4 kg P/ha	11	26.4	10.2	(+) 16.2	21.8	6.5	(-) 25.5	(-) 15.3
Rock phosphate @ 26.4 kg P/ha + PSB inoculation	11	26.4	13.9	(+) 12.5	18.1	8.4	(-) 23.6	(-) 9.7

1990). Total average phosphorus uptake was significantly increased with all the treatments except with rock phosphate @ 13.2 kg P/ha and the increase was 4.6–60.7%, maximum being in superphosphate @ 26.4 kg P/ha. But it was at par with superphosphate @ 13.2 kg P/ha + PSB inoculation and rock phosphate @ 26.4 kg P/ha + PSB inoculation, which was due to more solubilization from rock phosphate and superphosphate in presence of P-solubilizing bacteria, maximum agronomic efficiency and P-use efficiency were observed under half dose of superphosphate applied @ 13.2 kg P/ha with PSB inoculant, followed by rock phosphate @ 13.2 kg P/ha with PSB inoculant. The results confirm the finding of Bagavathi *et al.* (1997). The relative agronomic efficiency and relative economic efficiency and availability coefficient ratio of the rock phosphate were maximum under 13.2 kg P/ha with PSB inoculant, followed by higher dose of rock phosphate applied @ 26.4 kg P/ha with PSB inoculant. Data further indicated the greater utility of PSB inoculum at lower levels particularly with rock phosphate. Bagavathi *et al.* (1970) also reported similar results. Highest net profit and yard stick value were recorded with half dose of superphosphate @ 13.2 kg P/ha applied with PSB inoculum. However, rock phosphate @ 13.2 and 26.4 kg P/ha with PSB inoculation gave net profit of Rs 2 317/ha and Rs 2 705/ha and 8.37 and 5.02 kg/ha yardstick value respectively, which were almost at par with full dose of superphosphate @ 26.4 kg P/ha which gave only net profit of Rs 2 560/ha with 6.08 kg/ha yard stick value. Maximum

benefit : cost ratio and minimum cost of production/kg seed were recorded under PSB inoculation alone. Among the P sources, rock phosphate @ 13.2 kg P/ha with PSB inoculation recorded highest benefit : cost ratio and less cost of production/kg seed. The results are in close agreement with the findings of Dubey (1996). A significant correlation coefficient (*r*) of nodule number with nodule dry weight (0.55*), nodule dry weight with shoot dry weight (0.66*), seed index (0.66*), seed yield (0.77*) and shoot dry weight with yield (0.82*) was observed.

Average available soil phosphorus indicated a significant increase due to P application alone as well as with PSB inoculation compared with the control. This may be ascribed to addition of P and there may be some sort of triggering action of native soil P resulting increased P availability. Inoculation with P-solubilizing bacteria caused a further improvement in soil P possibility due to the solubilization of fixed or the added P (Gaur 1990). But, when compared to initial status of available P it was observed that all the treatments showed gain of P, which was higher in treatment received 26.4 kg/ha and lowest under the control.

Phosphorus balance indicated that P application alone as well with PSB inoculum had marked influence on the residual P status (Table 2). The net P left in the soil showed negative trend with all the treatments except with PSB inoculation which showed gain (1.2 kg P/ha) of P. The depletion in net amount of P was -0.5 to -25.6 kg P/ha, which was maximum in rock phosphate @ 26.4 kg P/ha,

might be due to more removal of P from furrow layer, poor availability and rapid chemical fixation of P from rockphosphate (Chatterjee *et al.* 1972). Inoculation with P-solubilizing bacteria increased P availability through dissolution of fixed native P. Similarly, data on loss or gain of P showed both trends. But it was observed that addition of higher doses of P caused more loss of P due to more utilization and demands by soybean crop for root proliferation, N₂ fixation, growth and higher yield and protein synthesis (Gaur 1990).

These results indicate that about 30–40% phosphorus could be saved with the use of half dose of superphosphate @ 13.2 kg P/ha along with *B. japonicum* + *Pseudomonas striata* (PSB) inoculants. It was also observed that rock phosphate @ 13.2 kg P/ha along with *B. japonicum* + PSB inoculants was found to be an effective source of P, hence can be safely used in phosphorus deficient Vertisols.

SUMMARY

A long-term (10 years) field study was carried out on phosphorus-deficient Vertisols during the rainy seasons from 1988 to 1997 to generate information on biocoenotic association between *Bradyrhizobium japonicum* and P-solubilizing bacteria on rainfed soybean [*Glycine max* (L.) Merr.] with and without superphosphate and rock phosphate. The maximum average seed yield (1.99 tonnes/ha) was recorded with superphosphate @ 26.4 kg P/ha, and was equally effective with 50% dose of superphosphate @ 13.2 kg P/ha (1.88 tonnes/ha) and rock phosphate @ 26.4 kg P/ha (1.93 tonnes/ha) applied with phosphate-solubilizing bacteria (*Pseudomonas striata*). However, 50% reduction in superphosphate, ie @ 13.2 kg P/ha, along with P-solubilizing bacterial inoculum recorded highest agronomic efficiency, P-use efficiency, net profit (Rs 2 967/ha) and yard stick value (12.27 kg/ha). Rock phosphate applied @ 13.2 and 26.4 kg P/ha with PSB inoculum also proved to be an effective source of P than the superphosphate alone for seed yield and superior in respect of agronomic efficiency, P-use efficiency, net profit benefit : cost ratio and low-cost of production/kg seed. Inoculation also improved the availability coefficient ratio, relative agronomic efficiency and relative economic efficiency of rockphosphate compared

with the rock phosphate alone. Nodule number had significant positive correlation with nodule dry weight, nodule dry weight with shoot dry weight, seed index, seed yield and shoot dry weight with seed yield.

REFERENCES

- Awasthi P K, Luthra K L and Jaggi J N. 1977. Use of Indian rock phosphates for direct application as phosphatic fertilizers. *Fertiliser News* 12 : 44–4.
- Bagavathi Anmol, U Rani Perumal and Mehimairaja S. 1997. Phosphorus availability from tunis rock phosphate as influenced by organic amendments and biofertilizers. *Journal of Indian Society of Soil Science* 45(4) : 837–9.
- Black C A and Scott C O. 1956. *Fertilizer Evaluation. Fundamental Principles of Soil Science. Proceeding. Soil Science Society of America* 20 : 176–9.
- Chatterjee B N, Raquib A and Maiti S. 1972. Phosphorus manuring of soybean and its effect on wheat crop. *Journal of Indian Society of Soil Science* 20 : 176–8.
- Dubey S K. 1996. Response of soybean to rock phosphate applied with *Pseudomonas striata* in a Typic chromustert. *Journal of Indian Society of Soil Science* 44 (2) : 252–6.
- Engelsted O P, Jugsuinda A and Dutta S K De. 1974. Response of flooded rice to phosphorus rock varying in Citrate solubility. *Proceedings of Soil Science Society of America* 38 : 524–9.
- Gaur A C. 1990. *Phosphorus Solubilizing Micro-Organisms as Biofertilizer*, pp 63–90. Omega Scientific Publishers, New Delhi.
- Mandal L N and Khan S K. 1972. Release of phosphorus from insoluble phosphatic materials in acidic low and rice soils. *Journal of Indian Society of Soil Science* 20 : 19–25.
- Manickam T S and Ramaswami P P. 1985. Importance and methods of evaluation of phosphatic fertilizers. (in) *Proceedings of National Seminar on Rock Phosphate in Agriculture*, pp 183–191, held during 22–23 August at Tamil Nadu Agricultural University, Coimbatore.
- Mishra B and Gupta R P. 1978. Evaluation of Mussoorie rock phosphate as phosphorus source for maize on acid soil of Kumaon hills. *Indian Journal of Agricultural Sciences* 48(4) : 239–44.
- Mole R H, Kamprath E J and Jackson W A. 1972. *Agronomy Journal* 78 : 526.
- Singh R S and Ram Hari. 1977. Effect of organic matter on the transformation of inorganic phosphorus in soils. *Journal of Indian Society of Soil Science* 25 : 118.