

PRODUCTIVITY OF DIRECT SEEDED RICE-WHEAT SYSTEM AS INFLUENCED BY PHOSPHORUS LEVELS AND PHOSPHATE SOLUBILIZING BACTERIA

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ABSTRACT

Phosphorus (P) is an important macronutrient required for the growth and development of crop plants. However, the majority of P in soil is present in insoluble forms. Phosphate solubilizing bacteria (PSB) has the capacity to convert insoluble phosphates into a form which is readily available to crop plants. Thus, the present investigation was conducted during *rabi* 2015-17 and *kharif* 2016-17 to investigate the effect of PSB and its inoculation methods (seed and/or soil) on productivity of direct seeded rice (DSR) - wheat system under varying P levels. For both the crops, improved growth and yield parameters were observed with the application of 60 kg P₂O₅/ha (P₆₀) followed by P₄₅ (45 kg P₂O₅/ha), P₃₀ (30 kg P₂O₅/ha) and control (no P). P₆₀ level of phosphorous resulted in significantly higher grain and straw yield for both wheat and DSR during both the years, which was statistically similar with P₄₅, but significantly better than P₃₀ and control. Although, no significant influence of PSB inoculations on yield and yield components of DSR and wheat were observed, but single (seed/soil) and mixed inoculation (seed+soil) of both the PSB strains (*Bacillus* spp. and PSB IARI) not only improved the grain yield and yield components but also increased the availability of soil nutrients (N, P and K), as compared to non-inoculated control plots. Application of PSB strains needs special emphasis for increasing production and productivity of DSR-wheat cropping system.

Keywords: Direct seeded rice, Growth, P levels, Productivity, P solubilizing bacteria, Wheat

The rice-wheat cropping system is the world's largest agricultural production system that occupies 13.5 million hectares (M ha) of cultivated land in South Asia (Nawaz *et al.*, 2019). Millions of people in this region are dependent on rice-wheat cropping system for their livelihood. Both rice and wheat are exhaustive feeders and continuous cultivation of this system rapidly depletes the soil of its primary nutrients. Benbi *et al.* (2006) reported that a rice-wheat system with production of 7.0 and 4.0 t/ha of rice and wheat, respectively depletes 300 kg N, 30 kg P and 300 kg K per hectare from the soil.

Among plant nutrients, phosphorus (P) is the second most important macronutrient after nitrogen, required by plants. It plays a very important role in several biochemical and physiological processes viz., photosynthesis, respiration, energy storage and cell division/enlargement. Despite abundant amounts of P present in parent material, the soil phosphorus availability is limited for plants. In calcareous soil, P is precipitated and plants are unable to utilize the precipitated P. Further, because of their higher soil retention, the mobility of H₂PO₄⁻ and HPO₄²⁻ (phosphate ions) in soil is extremely slow. The processes like adsorption, precipitation and transformation into organic

forms results in recovery of about 10–30% of P from the applied phosphate fertilizers by the crops following fertilizer application (Syers *et al.*, 2008). Remaining 70 to 90% P gets fixed and become part of the soil pool (Singh *et al.*, 2016). Hence, there is dire need to mobilize the unavailable soil phosphorus to make it available for plants.

Phosphate solubilizing bacteria (PSB) play a significant role in solubilization of inorganic P from insoluble compounds and has been recommended for the agricultural use as a bio-fertilizer. These are the microorganisms that releases low molecular weight organic acids from mineral phosphate by binding phosphate to the chelates of hydroxyl and carboxyl groups, which lowers soil pH resulting in increased P availability. There are numerous methods used worldwide to apply the inoculant to the crop, but two methods namely seed and soil inoculation are most commonly used. In seed inoculation method, seeds are mixed with inoculant before sowing, whereas soil inoculation is used for inoculation of rhizosphere. Under transplanted rice-wheat system having flooded conditions during the *kharif* season solubilization of fixed phosphorus into available forms is more as compared to direct seeded rice (DSR)-wheat system. Thus, there is ample scope to increase P availability in DSR-wheat system using PSB under varying P levels. Keeping this in view, the current study was carried out to

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evaluate the effect of PSB and its inoculation methods on phosphorus availability under different P levels in DSR-wheat system.

MATERIALS AND METHODS

The present field study was conducted at the Research Farm of Department of Agronomy, Punjab Agricultural University, Ludhiana (latitude 30°54'N, longitude 75°48'E, altitude 247 m) during 2015-17. The site is situated in the central plain region of Punjab under Trans-Gangetic agro-climatic zone of India. The climate of the site is sub-tropical and semi-arid with mean annual precipitation of 650 mm and 75% of the average annual rainfall is received as south-west monsoon. The experimental field soil was calcareous alluvial in texture with pH 7.1, organic C 0.45%, available N, P and K of 234.1, 19.9, 336.2 kg/ha, respectively. The experiment was laid out in a split plot design with three replications keeping four P levels in main plots (0, 30, 45 and 60 kg P₂O₅/ha in wheat without any P application in DSR) and seven inoculation treatments {without inoculum (control), seed inoculation with *Bacillus* spp. (SI₁), seed inoculation with PSB (SI₂), soil inoculation with *Bacillus* spp. (Sol₁), Soil inoculation with PSB (Sol₂), Seed and soil inoculation with *Bacillus* spp. (SI₁ + Sol₁), Seed and soil inoculation with PSB (SI₂ + Sol₂)} in sub-plots (Fig. 1).

After pre-sowing irrigation, wheat variety HD-2967 was sown on 24th November, 2015 and 11th November, 2016, respectively at optimum soil moisture using 100 kg seed ha⁻¹ with seed drill, in rows 15 cm apart. After harvesting of wheat, rice variety PR 115 was sown in the same plots on 13th June 2016 and 6th June 2017, respectively with seed rate of 25 kg/ha in 20 cm apart rows at a depth of 2-3 cm deep. For the seed inoculation treatment, wheat seeds were moistened with water and then mixed with biofertilizers (liquid-based *Bacillus* spp. and PSB IARI) whereas, for soil inoculation, the recommended dose of biofertilizers were mixed with soil and broadcasted in the field before wheat sowing as per treatments. Treatment wise P was applied to wheat crop (0, 30, 45 and 60 kg P₂O₅/ha) as basal at the time of sowing and N was applied in two equal splits, first at sowing and second just before first irrigation through chemical fertilizers i.e. Urea (46 % N), DAP (18 % N and 46 % P₂O₅) and MOP (60 % K₂O). No phosphorous (P) was applied to rice crop and thus was sown on residual soil P. However, recommended dose of N was applied through urea (46 %) in three equal splits i.e. 1/3rd two weeks after sowing, 1/3rd five weeks after sowing and 1/3rd nine weeks after sowing. Recommended dose of potassium and zinc were applied at the time of sowing through MOP and zinc sulphate heptahydrate (21%), respectively. Recommended plant protection measures were adopted to protect both the crops from various

biotic stresses. At maturity, crops were harvested manually with sickles and bundles were made from the net area of each plot and left in the field for drying in sun. Finally, after threshing and winnowing, seed yield was recorded and expressed in q/ha.

Analysis of variance (ANOVA) was done using CPCS1 software developed by Punjab Agricultural University, India (Cheema and Singh, 1991) based on the procedure of Cochran and Cox (1967). Comparisons of difference between means were made 5% level of significance.

RESULTS AND DISCUSSION

Growth attributes

Significant influence of P fertilization treatments on growth attributes viz., plant height, dry matter accumulation (DMA) and leaf area index (LAI) of wheat and direct seeded rice (DSR) were observed during both the years (Table 1). P₆₀ recorded the highest plant height (89.2, 86.6 and 92.5, 92.2 cm), DMA (726.8, 722.6 and 965, 1019.9 g m⁻²) and LAI (3.8 and 4.2), which were statistically similar with P₄₅ and significantly higher than P₀ for both the crops, during both the years, respectively. The main reason for the positive effect of higher P doses on the growth attributes is that only 0.1% of the total P present in soil is available for plant uptake. Because of higher tendency of P to get fixed in soil either through adsorption or precipitation or conversion to organic forms, only 10-30% of applied P fertilizer can be recovered by the crop plants after the application of fertilizer (Syers *et al.*, 2008). In addition, P is also considered as a root growth promoting nutrient and with well-developed root system, a plant has an enhanced growth rate and thus more profuse growth attributes are observed with higher levels of phosphorus. Similar results were reported by Amanullah *et al.* (2017) and Aatif *et al.* (2017).

No significant effect was observed for PSB inoculation treatments and interaction between P fertilization and PSB inoculation for any of the growth attributes. However, single and mixed inoculation of both the PSB strains (*Bacillus* spp. and PSB IARI) improved the growth attributes as compared to non-inoculated control (Table 1).

Yield attributes

Different P fertilization treatments had significant effect on yield attributes viz., effective tillers/m², grains/ear except 1000-grain weight of wheat and DSR during both the years (Table 2). P₆₀ had the highest number of effective tillers/m² and grains/ear which were statistically at par with P₄₅ and both P₆₀ and P₄₅ treatments were significantly better than P₀ (control).

Table 1. Effect of phosphorus levels, PSB inoculations and inoculation methods on growth attributes of wheat and DSR at 90 DAS

Treatment	Wheat						DSR					
	Plant height (cm)		DMA (g/m ²)		LAI		Plant height (cm)		DMA (g/m ²)		LAI	
	2015-16	2016-17	2015-16	2016-17	2015-16	2016-17	2016	2017	2016	2017	2016	2017
Phosphorus (kg ha ⁻¹)												
P ₀	78.3	78.5	674.7	667.5	3.4	3.3	84.9	85.1	910.3	927.8	3.79	3.87
P ₃₀	84.5	83.8	692.6	686.9	3.6	3.5	87.2	87.3	921.9	954.0	3.89	3.96
P ₄₅	86.9	84.2	718.8	710.7	3.7	3.6	90.8	90.7	954.5	1001.0	4.01	4.11
P ₆₀	89.2	86.6	726.8	722.6	3.8	3.8	92.5	92.2	965.0	1019.9	4.15	4.18
CD (p=0.05)	2.9	2.8	20.4	21.9	0.2	0.3	1.8	2.1	19.4	32.9	0.24	0.18
PSB inoculation												
Control	83.9	81.8	701.3	694.8	3.5	3.5	87.7	87.5	936.1	973.8	3.90	3.99
S I ₁	85.9	84.8	703.0	697.4	3.6	3.6	88.9	88.4	937.8	975.6	3.99	4.06
S I ₂	84.8	82.6	703.5	697.8	3.7	3.6	88.8	89.1	938.8	975.5	3.98	4.04
So I ₁	84.5	82.3	702.3	695.7	3.6	3.6	87.9	88.1	936.1	974.9	3.97	3.97
So I ₂	83.9	82.6	702.7	695.8	3.6	3.6	88.0	87.9	936.9	974.7	3.94	4.03
S I ₁ + So I ₁	85.6	84.7	704.4	698.0	3.6	3.6	90.0	90.1	940.0	976.7	3.92	4.10
S I ₂ + So I ₂	84.5	84.3	705.4	698.9	3.7	3.6	90.7	90.9	939.9	978.5	4.01	4.03
CD (p=0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Interaction	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Table 2. Effect of phosphorus levels, PSB inoculations and inoculation methods on yield attributes of wheat and DSR

Treatment	Wheat						DSR					
	Effective tillers/m ²		Grains/ear		1000-grain weight (g)		Effective tillers/m ²		Grains/ear		1000-grain weight (g)	
	2015-16	2016-17	2015-16	2016-17	2015-16	2016-17	2016	2017	2016	2017	2016	2017
Phosphorus (kg ha ⁻¹)												
P ₀	305	298	43	45	39.9	39.8	298	294	114	116	22.2	22.1
P ₃₀	318	310	45	47	40.3	40.3	311	306	115	117	22.4	22.3
P ₄₅	322	323	47	49	40.3	40.4	325	320	116	118	22.5	22.4
P ₆₀	329	330	49	50	40.5	40.8	332	328	116	118	22.5	22.5
CD (p=0.05)	8	8	3	2	NS	NS	8	10	NS	NS	NS	NS
PSB inoculation												
Control	313	310	45	47	39.5	39.7	315	310	114	116	22.3	22.2
S I ₁	321	318	46	48	40.6	40.6	317	313	116	117	22.5	22.4
S I ₂	319	316	45	47	40.4	40.2	316	312	115	117	22.4	22.3
So I ₁	317	313	45	47	40.2	40.5	316	311	115	117	22.3	22.3
So I ₂	315	314	45	47	39.8	39.8	316	312	115	117	22.3	22.2
S I ₁ + So I ₁	321	319	47	49	40.7	40.9	319	313	117	120	22.5	22.4
S I ₂ + So I ₂	322	318	46	48	40.5	40.7	317	310	116	118	22.4	22.4
CD (p=0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Interaction	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

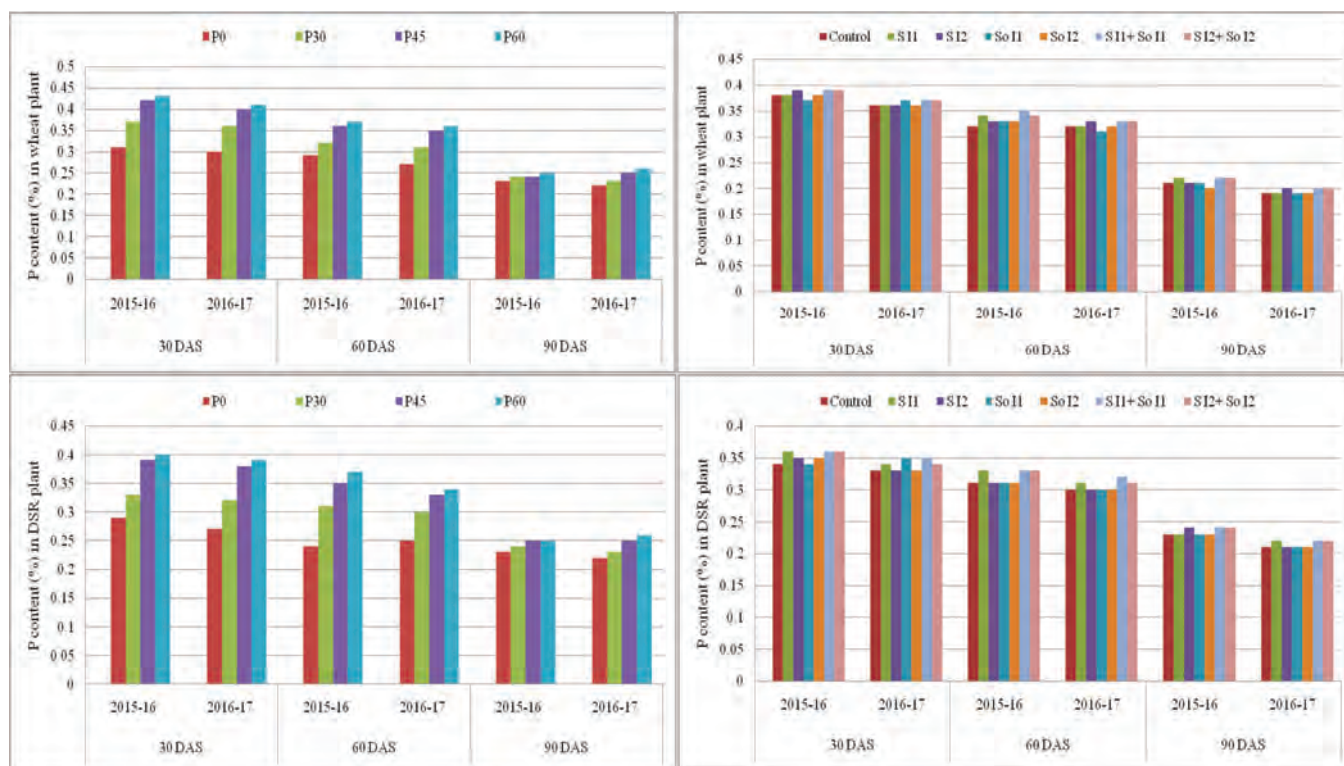


Fig. 1. Effect of phosphorus levels, phosphate solubilizing bacteria and method of inoculation on the periodic phosphorus content (%) in wheat and DSR plants

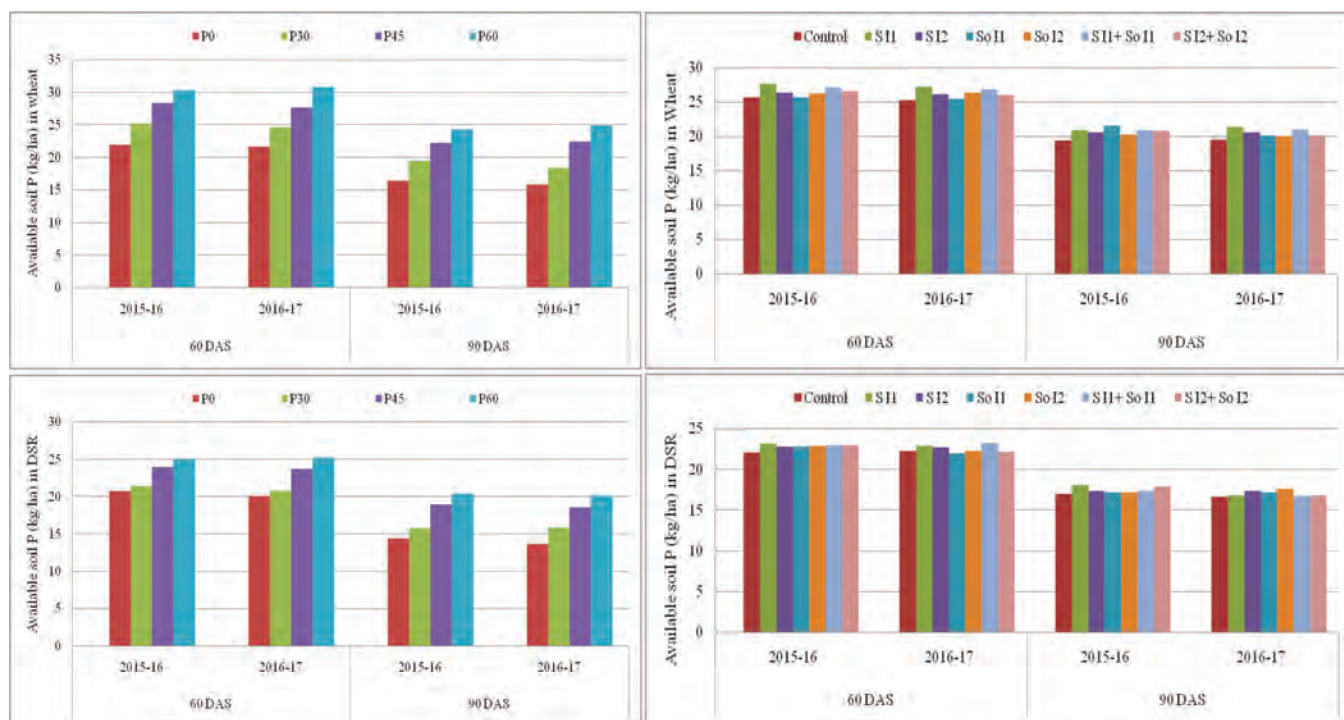


Fig. 2. Effect of phosphorus levels, phosphate solubilizing bacteria and method of inoculation on periodic available soil P (kg/ha) in wheat and DSR plots

Phosphorus is considered as root growth promoting nutrient and with well-developed roots a plant has an enhanced growth rate and thus more profuse tillering is observed with higher levels of phosphorus. Similar findings were reported by Amanullah *et al.* (2017), Aatif *et al.* (2017) and Rahim *et al.* (2010) who observed significant increase in tiller count with increase in phosphorus levels. In addition, inoculated plants showed higher number of yield attributes than non-inoculated control plants, although the difference was non-significant. The highest value in PSB inoculation treatments were recorded in seed and soil inoculation with *Bacillus* spp. (S I₁+ So I₁) followed by S I₂+ So I₂ (seed and soil inoculation with PSB IARI) and S I₁ (soil inoculation with *Bacillus* spp.) treatment for both wheat and DSR. Interaction among P and PSB inoculation treatments exhibited non-significant statistically during both the years.

Grain and straw yield

The crop yield per unit area is a direct measure of the economic returns to the farmer. Among P fertilization, P₆₀ level of phosphorus recorded highest grain yield (58.7, 59.0 and 66.7, 69.4q/ha) for both wheat and DSR during 2015-17, which were 24.9, 29.9 and 16.8, 15.1 per cent more than P₀ treatment, respectively (Table 3). P₆₀ was found to be statistically at par with P₄₅ (53.7,

56.1 and 65.1, 68.4q/ha) and statistically better than the P₀ (control) for both wheat and DSR during both the years. Despite abundant amounts of phosphorus stored in parent material, the soil phosphorus availability is limited for plants. The significant effect of P fertilizers on the dry matter accumulation (DMA) and effective tillers m⁻² has helped to increase the grain yield of both the crops during 2015-17 (Tables 1 and 2). Naima *et al.* (2020) reported that wheat is highly responsive to P fertilization in terms of grain yield and growth parameters. This effect of P fertilizers is basically due to of lesser amount of available P for the plants. Thus, with higher doses of P an enhancement of grain yield is observed. Amanullah *et al.* (2017), Singh *et al.* (2002) and Singh *et al.* (2000) also documented significant increase in grain yield with higher P levels.

The PSB inoculation treatments were non-significant during first year of wheat trial but during second year, these inoculations improved grain yield of wheat crop significantly. Treatment S I₁+ So I₁ recorded maximum grain yield (53.2q/ha), which was statistically similar with S I₁ and S I₂+ So I₂ (52.9 and 52.3 q/ha) but significantly better than un-inoculated control plots (50.5q/ha) during 2016-17. Notably all the PSB inoculation treatments recorded higher grain and straw yield than non-inoculated control plants, irrespective of the crop and year. Although, the interaction between P

Table 3. Effect of phosphorus levels, phosphate solubilizing bacteria and method of inoculation on grain and straw yield of wheat and DSR

Treatment	Wheat						DSR					
	Grain yield (q/ha)		Straw yield (q/ha)		Harvest index (%)		Grain yield (q/ha)		Straw yield (q/ha)		Harvest index (%)	
	2015-16	2016-17	2015-16	2016-17	2015-16	2016-17	2015-16	2016-17	2015-16	2016-17	2015-16	2016-17
Phosphorus (kg ha ⁻¹)												
P ₀	47.0	45.4	69.2	68.0	41.1	40.9	57.1	60.3	80.3	79.1	41.8	41.8
P ₃₀	50.3	47.3	71.2	72.6	41.2	41.1	61.4	65.0	84.7	82.9	42.2	42.1
P ₄₅	53.7	56.1	77.2	77.0	41.4	41.5	65.1	68.4	88.6	87.8	42.4	42.6
P ₆₀	58.7	59.0	80.8	80.7	41.6	41.7	66.7	69.4	91.4	91.7	42.6	42.7
CD (p=0.05)	7.3	3.5	8.2	5.7	0.2	0.3	4.9	6.5	4.5	5.4	0.3	0.4
PSB inoculation												
Control	51.2	50.5	73.3	73.0	41.2	41.2	60.2	62.5	85.1	82.4	42.1	42.0
S I ₁	53.0	52.9	75.7	74.6	41.5	41.4	63.5	67.7	87.6	86.7	42.3	42.5
S I ₂	52.7	51.7	74.4	74.8	41.3	41.3	63.2	66.3	85.6	86.4	42.2	42.2
So I ₁	52.9	51.7	74.2	74.3	41.2	41.3	61.9	64.6	85.7	85.2	42.3	42.2
So I ₂	51.3	51.4	73.6	73.9	41.2	41.2	60.9	64.3	85.8	83.1	42.2	42.1
S I ₁ + So I ₁	53.1	53.2	76.2	76.7	41.5	41.4	64.3	68.0	87.7	87.3	42.5	42.6
S I ₂ + So I ₂	52.9	52.3	75.1	74.7	41.4	41.3	64.1	67.0	86.2	86.6	42.4	42.6
CD (p=0.05)	NS	2.8	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Table 4. Effect of phosphorus levels, PSB inoculations and method of inoculation on available soil nutrients (kg/ha) in wheat and DSR plots

Treatment	Wheat						DSR					
	N (kg/ha)		P (kg/ha)		K (kg/ha)		N (kg/ha)		P (kg/ha)		K (kg/ha)	
	2015-16	2016-17	2015-16	2015-16	2016-17	2015-16	2016	2017	2016	2017	2016	2017
Phosphorus (kg ha ⁻¹)												
P ₀	246.7	245.3	15.7	15.1	359.7	350.7	238.4	236.7	13.9	13.3	348.2	344.6
P ₃₀	242.8	243.4	17.0	16.4	355.4	345.4	232.6	235.6	15.0	14.9	343.9	342.8
P ₄₅	238.8	237.7	19.5	19.3	348.0	337.7	227.2	224.8	18.3	18.4	334.1	336.7
P ₆₀	236.6	235.3	20.6	21.1	343.2	335.2	225.4	220.6	20.2	20.5	332.2	333.7
CD (p=0.05)	NS	NS	2.4	2.6	NS	NS	NS	NS	2.3	2.8	NS	NS
PSB inoculation												
Control	236.9	236.5	17.0	17.5	347.6	338.5	226.6	224.9	16.4	16.5	337.4	339.8
S I ₁	243.3	242.8	19.1	18.3	351.5	342.3	233.5	231.5	17.3	17.0	338.3	339.0
S I ₂	240.8	240.6	17.8	17.7	350.5	348.6	230.9	229.0	16.8	16.7	340.7	338.9
So I ₁	239.1	238.5	18.5	17.6	353.5	341.5	228.7	227.3	16.6	16.4	339.0	338.6
So I ₂	236.7	238.9	18.1	17.7	348.9	341.4	227.0	225.1	16.6	16.6	335.3	339.8
S I ₁ + So I ₁	248.2	243.1	18.5	18.7	352.2	342.8	235.7	236.4	17.5	17.6	345.4	339.3
S I ₂ + So I ₂	243.6	242.5	18.4	18.1	356.9	340.6	233.7	231.8	16.8	16.6	341.2	340.5
CD (p=0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Interaction	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Initial status	234.1	-	19.9	-	336.2	-	-	-	-	-	-	-

fertilization and PSB inoculation treatments were found to be non-significant.

Available soil nutrients in wheat and DSR plots

P fertilizers had a non-significant effect on available soil N and K for initial and after harvest soil status during both the years in both the crops (Table 4). However, the initial and after harvest soil P status was significantly affected by P fertilization treatments during 2015-17. Highest dose of P i.e. P₆₀ was found to be the best in increasing the available soil P at harvest. However, P₄₅ fertilizer treatment was found to be statistically at par with P₆₀ and significantly better than control. Khan *et al.* (2007) reported similar observations of increase in available soil P with higher doses of P fertilizers.

The PSB inoculation treatments and the interaction between P fertilizer and PSB inoculation treatments were found to be non-significant in affecting before and after available soil NPK during both the years for both the crops.

Periodic P content in wheat and DSR plants

Higher levels of P treatments had a positive effect on P content in wheat and DSR plants during both the crop years (Fig. 1). Application of P₆₀ resulted in maximum P content in wheat and DSR plants at 30, 60 and 90 DAS

during both the years. Treatment P₄₅ was found to be statistically at par with P₆₀. However, both P₆₀ and P₄₅ treatments were better than the control for both crops during both the seasons. The reason for the increased P content in crop plants with higher doses of P fertilizer is the increased amount of available soil P with the application of higher P doses. As the soil available P is higher, the plant is able to take up more P and thus increased plant P content. The increased root growth with higher doses of P fertilizers resulted in higher P uptake. Similar conclusions of increased P content with higher residual P fertilizer doses are reported by Singh *et al.* (2000). The PSB inoculation treatments were found to be non-significant for both wheat and DSR crops. The interaction was also non-significant.

Periodic available soil P in wheat and DSR plots

The P fertilization treatments had a significant effect on available soil P at 60 and 90 DAS (Fig. 2). Application of P₆₀ treatment had maximum soil available P. However, the available soil P with P₆₀ treatment was found to be statistically at par with P₄₅, during both the years. Notably both P₆₀ and P₄₅ treatments were statistically better than the control. The reason for increased available soil P with the P fertilization treatments

is the lesser solubility of P in the soil. Thus, when P fertilization is applied to the soil, amount of available soil increases and later when the P fixation started in the soil, the available soil P begins to decrease. Khan *et al.* (2007) reported similar observations of increase in available soil P with higher doses of P fertilizers. The PSB inoculation treatments and the interaction effect between P fertilization and PSB inoculation treatments were found to be non-significant.

The present investigation amplified the scope of phosphorous levels and phosphate solubilizing bacteria in direct seeded rice (DSR)-wheat cropping system. Improved growth and yield parameters of DSR and wheat were observed with application of 60 kg P_2O_5 /ha (P_{60}), which was comparable with P_{45} (45 kg P_2O_5 /ha) but statistically better than P_{30} (30 kg P_2O_5 /ha) and control. The P_{60} level of phosphorous lead to significantly higher grain yield of both wheat and DSR followed by P_{45} , P_{30} and control. Although, no significant influence of PSB inoculations were observed in improving growth and yield components of any of the crop in the DSR-wheat system, but single and mixed inoculation of both the PSB strains (*Bacillus* spp. and PSB IARI) improved the yield components and grain yield comparing with non-inoculated taken as control. Significantly higher grain yield of wheat crop was observed with seed and soil inoculation of *Bacillus* spp. than non-inoculated control treatment during 2016-17. In 2015-16, 1.9 q/ha higher wheat yield and in 2016 and 2017, 4.1 and 5.5 q/ha higher DSR yield, were recorded with seed and soil inoculation of *Bacillus* spp. than control. Thus, PSB inoculation needs immediate attention for improving productivity of DSR-wheat system.

Authors' contribution

Conceptualization of research work and designing of experiments (JSD); Execution of field/lab experiments and data collection (JSD, MSS); Analysis of data and interpretation (JSD, MSS, RS); Preparation of manuscript (JSD, MSS, RS)

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