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Response of Phosphate Solubilizing Microorganism on Quality of Wheat (*Triticum Aestivum* L.) Plant Grown Conventionally in Temperate Climate

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Abstract

The present study was conducted to assess the effect of inoculation on wheat (*Triticum aestivum* L.) with the bacterium *Pseudomonas fluorescens* sp. SSC in Doon valley by conventional method. Treatments involved SSC inoculation with phosphorus fertilization. The results revealed that application of phosphate solubilising bacteria in combination with half dose (50%) of superphosphate significantly increases the yield (36%) as well as P status in soil and plant over control. It significantly increases the total uptake of P content in plant by 33%. The conventional treatment without bacterial inoculation has resulted in very low P uptake in plants. Thus, it is suggested that application of phosphate solubilizer SSC is able to mobilize more P to the plants and improves plant yield. Thus, results of the study indicate that inoculation of wheat with *P. fluorescens* SSC alone improves yield and quality parameters allowing input of reduced dosage of phosphorus fertilizer

Keywords: Pseudomonas fluorescens SSC; PSM; Yield; P-uptake; Wheat

Introduction

Wheat contributes about 35% to the total food grain basket of the country. Despite having great potential, India's average productivity of wheat is undergoing stagnation. Phosphorus is an important macronutrient required for the development, growth and yield of wheat and of every crop [1]. But, compared to other nutrient phosphorus is least mobile in moist soil conditions. Though every soil has abundant phosphorus, be inorganic or organic form, still it is a prime limiting factor in plant growth. On applying superphosphate to soil and watering, within short period of time it gets converted into insoluble phosphate, though the span varies from soil to soil. The use of phosphate solubilising bacteria capable of association in rhizosphere with agriculture crops can promote phosphate availability to plant. As the adoption of minimum tillage and zero tillage practices can help in restoration of rhizosphere microorganisms, higher microbial biomass carbon and higher microbial community necessary to maintain the soil quality.

In view of this, attempt has been made to solubilise the insoluble phosphate by using phosphate solubilising bacteria (PSB). These bacteria secrete various organic acids merely to enhance phosphate availability. Since, soil is a complex matter, our understanding of the mechanism of this process is not clear. Hence, carrying out field experiment on soil is the best way available to us so as to have optimum yield with minimum PSB and superphosphate.

Material and Methods

Experimental site and soil preparation

The field trial was conducted in Uttaranchal state to determine the effect of inoculation of phosphate solubilizer in Wheat (*Triticum aestivum*), at agricultural farm located at Bhandari bagh, Dehradun. The soil type was sandy loam. The chemical characteristics of the soil of the site are given in Table 1. Soil pH and electrical conductivity were measured (1:2.5 soil to water ratio) using a digital pH and EC meter. Available phosphorus (Olsen's P) in the soil was determined colorimetrically on spectrophotometer at 882 nm by extraction with sodium bicarbonate for 30 min. Nitrogen was determined by modified Kjeldahl distillation method, Organic carbon was estimated colorimetrically. Microbial isolation was done by enrichment culture technique followed by serial dilution and plating on King's B media.

Bacterial inoculum and planting germplasm

PSB inoculum, Pseudomonas fluorescens SSC produced through in

| pH (1:2.5 H ₂ O) | EC (dS/m) | Organic C (%) | PSM density (cfu/g soil) | Olsen P (ppm) | Total N (ppm) | Exchangeable K (ppm) |
|--------------------------------|--------------|------------------|--------------------------------|------------------|------------------|-------------------------|
| 7.6 | | 2.1 | | 2.0 | 146.0 | 46.0 |

 Table 1: Biochemical characteristics (0–30 depth) of experimental soil at the time of initiation of field trial.



Figure 1: Determination of economic (grain) yield, stover yield and biological yield per hectare after harvest of wheat plants var. PBW-373 in different treatments and control.

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vitro method was isolated from soil of Doon valley and was certified by IMTECH, Chandigarh. It was used for inoculation. The phosphate solubilising ability was determined by halo-zone test performed on Pikovskaya's agar medium containing tricalcium phosphate (TCP) [2].

Seeds of wheat cultivar variety PBW-373, obtained from National Seed Corporation, Dehradun, were used.

Experimental setup and design

In field, seedbed was prepared with tractor-drawn cultivar followed by planking, 4 days before sowing. The treatments consist of five fertilizer and inoculation treatments each in flat/conventional rows over standard treatment. The experiment was laid out manually in randomized block design (RBD) with 6 replicates having a block size of $1 \times 1 \text{ m}^2$.

A basal dressing of nitrogen@120 kgha⁻¹ and potassium@60kgha⁻¹ were applied. Phosphorus (P) was applied in the form of Diammonium phosphate as a source@80 kgha⁻¹ in graded dose in combination with PSM. Potassium was supplied in the form of Murate of Potash (MOP) and Nitrogen by urea before sowing. Farm yard manure@10tonsha⁻¹ was applied. Seeds were sown in rows@120kgha⁻¹. Bacterial inoculation was done by seed encapsulation by mixing them in slurry with grains, half an hour before sowing.

The treatments were planned to study the effective combination. These were PSB + $P_{100\%}$ (T_1); PSB + $P_{67\%}$ (T_2); PSB + $P_{50\%}$ (T_3); PSB + $P_{34\%}$ (T_4); PSB + $P_{25\%}$ (T_5), over control (T_0) $P_{100\%}$. To study the direct effects of the identified strain on plant growth, one plant system was selected.

After care, harvest and study

The crop was irrigated five times covering all the critical stages

of growth and harvested after 140 days of sowing. Number of tillers (n), ear length (cm), number of spike lets and grains per ear(n), dry shoots weight (dried at 70°C for 70 hrs), harvest index, shoot height, grain weight per spike and 100-seeds weight were recorded. P analysis in soil, plant and grain was done colorimetrically. The grain yield of plants inoculated with microbe and externally applied with phosphorus was calculated mathematically for fertilizer savings by microbial application. Yield was recorded in kgha⁻¹.

Statistical analysis

The data collected were analyzed statistically using ANOVA and significance test was done at the rate 5% (p<0.05) [2].

Results

In field, observations were recorded in treatments inoculated with bacteria *Pseudomonas fluorescens* SSC and inorganic fertilizer in wheat, grown conventionally (farmers practice). The analysis of variance indicates that all plant growth agronomic parameters of wheat grown in rows responded positively to bacterial inoculation.

Data regarding yield (Table 1 and Figure 1) showed that PSM SSC inoculation with organic manure and 50% superphosphate significantly influences the yield. Grain yield was influenced upto 36% (1087 kgha⁻¹) over control. PSM addition significantly affects the stover yield and biological yield of wheat by 65% (280583kgha⁻¹) and 56% (389363kgha⁻¹) as compared to other treatments and control. Pertaining to tillers number per plant showed an increasing trend with PSM. The data revealed that treatments influence the plant height by increasing the ear head length by 23%, number of spikelets by 38.7% and grains by 25% number per ear in crop plants over control. The 100-grain weight was found maximum in T₃ (23.3%) and minimum in T₅ (6%) over control. Thus, increase in biological yield over economic (grain)

| Treatments | Tillers no/ plant (n) | Ear length (cms) | Spikelets no ear¹ (n1) | Grain no ear⁻¹ (n2) | Weight/100 grains (g) | Grain yield (kg ha⁻¹) | Stover yield (kg ha ⁻¹) | Biological yield (kg ha ^{.1}) | Harvest index (%) |
|--|--------------------------|---------------------|---------------------------|--------------------------|--------------------------|--------------------------|--|--|----------------------|
| P _{100%} + <i>Pf</i> SSC (T ₁) | 8 | *15.05 (9.5) | **90.00 (20) | 22.00 ^{ns} (10) | **3.50 | 101583 (27) | 241500 (42) | 343083 (37.4) | 29.60 |
| P _{67%} + <i>Pf</i> SSC (T ₂) | 7 | *15.28 (11.6) | *89.00 | *23.00 (10.5) | 3.10 ns | 95833 (20) | 237666 (40) | 333500 (33.5) | 28.70 |
| P _{50%} + <i>Pf</i> SSC (T ₃) | 9 | **16.60 (23) | **104.00 (38.7) | **25.00 (25) | **3.70 (23.3) | **108780 (36) | **280583 (65) | **389363 (56) | 27.90 |
| P _{34%} + <i>Pf</i> SSC (T ₄) | 7.2 | 14.60 ^{ns} | *88.00 | 22.00 ^{ns} (10) | 3.24 ^{ns} | 90000 (12.7) | 205200 (21) | 295200 (18) | 30.40 |
| P _{25%} + <i>Pf</i> SSC (T ₅) | 6 | 13.80 ^{ns} | **83.00 (10.7) | 22.00 ^{ns} (10) | 3.18 ^{ns} (6) | 82483 (3) | 185166 (9) | 287650 (7) | 30.80 |
| P _{100%} (T ₀) | 5.3 | 13.70 | 75.00 | 20.00 | 3.00 | 79790 | 16986 | 24965 | 31.90 |
| CD @ 5% | - | 1.74 | 6.70 | 2.65 | - | 178.24 | 653.23 | 829.88 | 1.93 |

highly significant @ 5% level of CD; *significant @ 5% over control; data given is the average of 6 replicates; Value in parenthesis shows % increase over control, (ns = non significant; CD = critical difference).

Table 2: Tillers number, spike length and number, spikelets number, grain number and 100-grains weight and yield parameters in wheat crop PBW-373 in different treatments and control.

| Treatments | Soil Av. P ₂ O ₅ (ppm) | Plant (Stover) Total P ₂ O ₅ (ppm) | Grain Total P ₂ O ₅ (ppm) | Total P ₂ O ₅ uptake in Plant (mg plant ⁻¹) |
|--|--|--|---|---|
| P _{100%} + Pf SSC (T ₁) | *2.00 (100) | **183.33 (9.99) | **466.67 (11.99) | *4.78 (26) |
| P _{67%} + Pf SSC (T ₂) | *2.00 (100) | **183.33 (9.99) | **450.00 (7.99) | *4.52 (19.5) |
| P _{50%} + Pf SSC (T ₃) | **3.00 (200) | **200.00 (19.99) | **483.33 (13.79) | **5.03 (33) |
| P _{34%} + Pf SSC (T ₄) | 1.50 ^{ns} (50) | 166.67 ^{ns} | **450.00 (7.99) | 4.14 ^{ns} (9.5) |
| P _{25%} + Pf SSC (T ₅) | 1.50 ^{ns} (50) | **183.33 (9.99) | **433.33 (3.99) | 4.13 ^{ns} (9.3) |
| P _{100%} (T ₀) | 1.00 | 166.67 | 416.67 | 3.78 |
| CD @ 5% | 1.11 | 6.29 | 12.06 | 0.75 |

** highly significant @ 5% level of CD; *significant @ 5% over control; data given is the average of 6 replicates; Value in parenthesis shows % increase over control, (ns = non significant; CD = critical difference).

Table 3: Phosphate status in soil, plant and grain and its uptake in plant in different treatments.

yield indicates that in low yielding environment wheat characters can be improved with low-input cultural conditions characterized by low soil fertility and irrigation. The harvest index shows decreasing trend with SSC application. Least was recorded with 50% P application over control having highest harvest index (HI). Thus, none of the treatments influenced the partitioning of dry matter (Harvest index) in wheat. The post-harvest studies revealed a positive influence of PSB *Pseudomonas fluorescens* SSC on P contents. This showed that yield of wheat is a function of integrated effects of its individual yield component that increases.

In Table 2, the available P content in soil, total P in plant and grain variants with PSB shows an increase upto 200% (3ppm), 19.9% (200ppm) and 13.79% (483.33ppm) significantly over control. It also increases the total P- uptake in plant by 33% (5.03mg plant⁻¹).

Thus, PSB application increases the P-uptake in plant. Finally, it can be said that application of 50% superphosphate combined with PSB *Pseudomonas fluorescens* SSC alone leads to increase in yield and P status of wheat plant, thus allowing input of reduced dosage (50%) of phosphorus fertilizer in agriculture soil of Doon valley. But, the success of bacterization program ultimately depends on aspects such as costeffective ratio, widespread applicability of specific strain, development of practical delivery systems and sustained positive results.

Discussion

The yield observation resembles the findings of Gholami et al. [4] that application of PSM Pseudomonas and Azospirillum increases the yield in grains per plant by 44% over control. This was in agreement to Dwivedi et al. [5] observation that presowing inoculation of wheat grains with PSM led to increase in yield over non-inoculated treatments. The increase in ear head length, number of spikelets and grains resembles the earlier findings of Algawadi [6] while working with sorghum involving the co-inoculation of Trichoderma harzianum and Pseudomonas striata. The increase in yield is in agreement with the earlier work done [7,8,9] and reported an increase in biological yield of sorghum, maize and rice, respectively. Harvest index shows the trend as studied earlier by Afzal et al. [10], and Carlier et al. [11]. Earlier study refers [12] that P content increases significantly in wheat plant with Bacillus meliloti combined with P as compared to uninoculated plants growing in control conditions and the availability of P content in soil and its uptake by plants increases with PSB application [13].

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