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Effect of single super phosphate, rock phosphate and phosphorus solubilising bacteria on phosphorus availability and its uptake by paddy in an acid soil

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Abstract

A pot experiment was conducted to examine the influence of Single Super Phosphate (SSP), Rock Phosphate (RP) and Phosphorus Solubilising Bacteria (PSB) on availability of phosphorus and its uptake by paddy (CAU-R1) in an acid soil. In comparison to the untreated control, all instances of phosphorus-treated soil exhibited notably higher levels of available phosphorus and its uptake at different stages of crop growth. Comparing with the untreated control all phosphorus treated soil gave significantly higher available P and its uptake at different stages of crop growth. Significantly higher available P was observed in soil treated with 50% SSP + 50% RP + PSB. Relatively more phosphorus uptake was recorded in paddy grown in 50% SSP + 50% RP + PSB followed by 25% SSP + 75% RP + PSB. Application of SSP and RP at 50:50 in combination with PSB maintains a constant pool of phosphorus for availability and agronomic effectiveness. PSB enhances the efficiency of the applied SSP and RP thereby increasing phosphorus availability to the crop which can ultimately boost the yield of paddy in the acid soil.

Keywords: Paddy, phosphorus solubilizing bacteria, single super phosphate, rock phosphate, nutrient uptake

1. Introduction

Phosphorus, one of the three primary macronutrient required for plant growth, plays a crucial role in various metabolic processes, including energy transfer, photosynthesis, and the synthesis of nucleic acids and proteins (Roch *et al.*, 2019) ^[27]. The general phosphorus content in soil measures approximately 0.05% (by weight), and only a mere 0.1% of this is accessible for plant uptake. Phosphorus availability in soil is often limited due to its strong fixation and immobilization reactions, leading to suboptimal phosphorus uptake by crops (Richardson *et al.*, 2011) ^[26]. Most of it is inaccessible to plants due to fixation by Al and Fe, or Ca and Mg which is either insoluble or insufficiently absorbed by plants (Murphy and Sims, 2012) ^[20].

To mitigate the challenges associated with phosphorus deficiency, farmers commonly apply phosphorus fertilizers to enhance nutrient availability and promote plant growth. Among these fertilizers, single super phosphate (SSP) and diammonium phosphate (DAP) are widely used due to their varying release rates and accessibility to plants (Azeem *et al.*, 2018) ^[3]. They provide readily available phosphorus to the plants. Along with the high costs associated with importing fertilizers from outside, the indiscriminate use of phosphate fertilizers is also detrimental. The following effects can be mentioned: excessive phosphorus absorption leading to phosphorus toxicity, which raises the phosphorus concentration in plant tissues and breaks nutritional balance; toxicity of boron; decreased copper absorption; immobilisation of iron in the soil; and prevent absorption of iron by roots (Jupp, *et al.*, 2021 and Renneson *et al.*, 2016) ^[14, 25]. Additionally, the prolonged use of phosphorus fertilizers results in eutrophication, water contamination, and soil acidification (Chaney, 2012) ^[6].

In recent years, there has been growing interest in exploring alternative and sustainable approaches to enhance phosphorus availability in soil and improve phosphorus uptake by plants. Utilizing affordable and readily available sources of phosphate is one of the alternate solutions. Rock phosphate cannot be use directly due to the low absorbability in most soils (Walan *et al.*, 2014) ^[30]. Phosphate-solubilizing microorganisms (PSMs) constitute a beneficial group of microbes capable of transforming insoluble inorganic and organic phosphorus compounds into soluble varieties. These beneficial microorganisms have the capacity to solubilize inorganic phosphorus, thereby increasing the

pool of available phosphorus for plant uptake (Khan *et al.*, 2007) ^[15]. The PSB solubilize the insoluble phosphate forms by generating siderophores, various organic acids, carboxyl and hydroxyl groups, and by chelating them with the bound phosphates and available calcium (Sharma *et al.*, 2013) ^[29]. The primary method for making phosphorus (P) available in soil is to lower soil pH by releasing protons or organic acids, followed by mineralization via the creation of acid phosphatases. (Alori *et al.*, 2017) ^[2]. The utilization of PSB has the potential to cut down P-fertilizer application by 50%, while maintaining plant growth and yield without significant decline (Jilani *et al.*, 2007) ^[13]. Among many others, PSB (*Bacillus megatherium*) are one of the most common and significant (Rawat *et al.*, 2021)^[24].

Rice (*Oryza sativa*) serve as a staple food crop for a substantial portion of the world's population, relies greatly on soil phosphorus availability for its productivity. Therefore, understanding the combined effect of phosphorus fertilizers (SSP and RP) and Plant Growth-Promoting Bacteria on phosphorus uptake and its availability is crucial for sustainable agriculture and efficient phosphorus management (Adnan *et al.*, 2022) ^[1]. The symbiotic relationship between PSB and plants has a positive impact on plant nutrient acquisition and resilience to environmental stresses, leading to enhanced crop productivity and reduced dependency on chemical fertilizers. (Richardson *et al.*, 2011) ^[26].

Considering the points mentioned above, a study was undertaken in the acidic soil of Manipur, India, to explore the impact of rock phosphate and single super phosphate, with or without the presence of phosphorus-solubilizing bacteria, on phosphorus availability and uptake by paddy (variety CAU-R1).

2. Materials and Methods

An acid soil having the characteristics as outlined in Table 1 were use from the research field of the College of Agriculture, CAU, Imphal. Uppermost soil layer (0-20 cm depth) were collected as slices, following the methodology laid out by Jackson (1973)^[12]. These composite soils were crushed in a wooden mortar and pestle and then sieved through a 2 mm screen after being fully air dried in the shade. Polythene bags were used to collect soil. Paddy variety CAU-R1 (Tamphaphou) was taken as test crop. Each plastic pot was filled with 5 kg of soil that had been already air-dried. Two phosphorus sources, namely single super phosphate (SSP) and rock phosphate (RP), were administered to each pot individually or in combination, adhering to the prescribed phosphorus dosage. i.e. 40 kg P₂O₅ ha⁻¹ for the paddy (variety CAU-R1) except control. Udaipur Rock Phosphate was used for the research. Urea and muriate of potash (MOP) were applied as basal dressing at the rate of 60kg N ha⁻¹ and 30kg N ha⁻¹, respectively to each pot before sowing to all the pots equally. Phosphorus solubilizing bacteria used in the study was Bacillus megatherium which was procured from the Plant Pathology department of College of Agriculture, CAU, Imphal. PSB was applied as seed treatment by soaking the seed in the PSB solution (10ml/litre of water) for 15 minutes. While some of the seeds were untreated. After inoculation, the paddy seeds were air-dried in the shade and promptly sown. Within each individual pot, a set of three to four paddy seeds were planted. Once germinated, a single seedling was selected and reetained for the entire duration of the experiment. Throughout the study, each pot was consistently

kept in a submerged state, with the water level consistently maintained at 5-10 cm above the soil surface. Regular management was carried out for rice growth. Altogether there were 11 treatment combinations replicated thrice. The experimental setup followed a completely randomized block design (CRD) which was replicated three times. Treatments: T₁= Control, T₂= 100% RD of P₂O₅ from SSP, T₃= 100% RD of P_2O_5 from RP, T_4 = 75% RD of P_2O_5 from SSP + 25% RD of P₂O₅ from RP, T₅= 50% RD of P₂O₅ from SSP + 50% RD of P₂O₅ from RP, T₆= 25% RD of P₂O₅ from SSP + 75% RD of P₂O₅ from RP, T₇=100% RD of P₂O₅ from SSP + PSB, T₈=100% RD of P₂O₅ from RP + PSB, T₉=75% RD of P₂O₅ from SSP + 25% RD of P_2O_5 from RP + PSB, $T_{10}=50\%$ RD of P₂O₅ from SSP + 50% RD of P₂O₅ from RP + PSB, $T_{11}=25\%$ RD of P_2O_5 from SSP + 75% RD of P_2O_5 from RP + PSB.

Rhizosphere soil and plant samples were collected by destructive sampling from the pot at 25th, 50th, 75th, 100th day after sowing (DAS) and at harvest. The initial soil samples underwent a process of air drying, after which they were finely ground into a powder and sifted through a 2.0 mm sieve. This preparations are use for analysis of fundamental soil properties, including soil texture, organic carbon content, pH, EC, cation exchange capacity (CEC), available nitrogen, phosphorus, and potassium. All assessments were conducted following standard procedures. The determination of available phosphorus content was done through spectrophotometric means using the Bray and Kurtz No. 1 method. Additionally, measurements were taken for both the fresh weight and dry matter yield of the plants. To determine the plant's phosphorus concentration, samples underwent digestion using a di-acid mixture of nitric acid and perchloric acid in a 4:1 ratio. The resulting digested plant materials were then subjected to analysis using the Vanadomolybdo phosphoric yellow color technique, as detailed by Jackson (1973) ^[12]. Phosphorus uptake was calculated based on the information concerning P concentration and dry matter yield, employing the following formula:

P uptake in plant (mg plant⁻¹)= Phosphorus concentration in plant sample (mg kg⁻¹) X calculated dry matter yield of plant sample (g plant⁻¹) X 1/1000

The study was conducted using a completely randomized design (CRD). To analyse the effects of the treatments, the experimental data underwent statistical analysis through the analysis of variance technique (ANOVA). The significance of different effects was assessed at a probability level of 5% (Gomez and Gomez (1984)^[10].

3. Results and Discussion

3.1 Available-P

The changes in available phosphorus (P) content in paddy soil, resulting from the application of single superphosphate and rock phosphate, in the presence or absence of phosphorus-solubilizing bacteria, have been documented in Table 2. The findings indicated that available phosphorus (P) peaked on the 50th day and subsequently decreased until the harvest stage, regardless of the various treatments applied. This showed the release of phosphorus into available form (Laskar *et al.*, 1990 and Goswami and Baroove, 1998) ^[16, 11]. The decrease might be due to fixation into different forms (Gerke, 1992) ^[9] or phosphorus uptake by paddy (Goswami and Baroove, 1998; Muraoka *et al.*, 2002; Xiong *et al.*, 2002; Poleshi *et al.*, 2008; Banerjee and Pramanik, 2009 and Lan *et* al., 2012) [11, 19, 32, 23, 4, 17]. All phosphorus fertilized soils showed significantly higher available-P accumulation over control. This aligns with the outcomes reported by Singh et al. (2007) ^[28]; Wang et al. (2011) ^[31] and Nayak et al. (2015) ^[21]. Comparing between single application of SSP and RP significantly higher available P was accumulated more in100% SSP treated soil. This indicated that RP releases available P at slower rate (Fotyma et al., 2002) [8]. Critical study of the data pointed out that significantly higher available-P was built up more in soil added with T_{10} (50%) SSP + 50% RP + PSB) followed by T_9 (75% SSP + 25% RP + PSB) on 25th, 75th and 100th day. However, on 50th day and at harvest, available-P accumulation in soil applied with T_{10} (50% SSP + 50% RP + PSB) was statistically at par with T₉ (75% SSP + 25% RP + PSB). Further study revealed that irrespective of different P sources, application of PSB accumulates significantly higher available P concentration than the corresponding treatment without PSB at 25th, 50th, 75th and 100 days after sowing. Reports on increasing the availability of phosphorus due to PSB application were given earlier by Deshpande et al. (2015)^[7] and Rawat et al. (2021) ^[24]. Phosphate solubilizing bacteria plays an important role in dissolving both fertilizer phosphorus (P) and bound P in the soil and phosphate solubilization is mainly due to the reaction between excreted organic acids with phosphate binders such as Al, Fe and Ca or Mg to form stable organic chelates to free the bound phosphate ion (Khan et al., 2007)^[15].

3.2 Phosphorus uptake

Data on changes in P-uptake by paddy are presented in Table 3. The findings revealed a consistent increase in phosphorus uptake by paddy plants up to the point of harvest, regardless of the treatments applied. Throughout its growth cycle, rice consistently absorbed phosphorus nutrients. Among the different treatments, rice plants exhibited the highest phosphorus nutrient uptake from the tillering stage to the elongation stage. As the dry matter content increased, the amount of phosphorus uptake also showed a gradual rise (Liu and Zhu, 1996) ^[18]. Detailed analysis indicated that regardless of the various treatments and sampling stages, paddy plants cultivated in phosphorus-fertilized soil exhibited significantly higher phosphorus uptake compared to the untreated control group. Similar findings were also reported by Poleshi et al. (2008) ^[23]; Goswami and Baroova (1998) ^[11]; Xiong et al. (2002) [32]; Muraoka et al. (2002) [19]; and Banerjee and Pramanik (2009)^[4]. Data further showed comparatively higher phosphorus uptake by paddy grown in soil added with T_{10} (50% SSP + 50% RP + PSB) which is at par with T_9 (75% SSP + 25% RP + PSB) and $T_{11} (25\% SSP + 75\% RP + PSB)$ on 50th and 75th day, respectively. However, on 100th day and at harvest comparative higher phosphorus uptake was observed in $T_{10}\ (50\%\ SSP\ +\ 50\%\ RP\ +\ PSB)$ added soil followed by T₁₁ (25% SSP + 75% RP + PSB). Bhardwaj et al. (1996) ^[5] also noted an elevated phosphorus uptake in paddy with the combined application of single superphosphate (SSP) and rock phosphate (RP). Critical study of the data pointed out that comparing between P treatments with or without PSB, statistically higher P uptake was found in PSB treated ones than the corresponding treatment without PSB. This is consistent with the conclusions drawn by Deshpande et al. (2015) ^[7]. Studies have shown that the application of PSB strains led to a higher solubilization of phosphorus from the soil, which in turn significantly enhanced plant uptake in aerobic rice (Othman and Panhwar, 2014)^[22].

Table 1: General	l characteristics	of the soil	used in	the experiment
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Soil Parameters	Results			
Soil Texture:				
Sand (%)	28.20			
Silt (%)	22.50			
Clay (%)	49.30			
pH (1:2.5 soil: water ratio)	5.2			
EC (1:2.5 soil: water ratio, dSm ⁻¹)	0.31			
CEC $[\text{cmol}(p^+) \text{ kg}^{-1}]$	14.92			
Organic carbon (%)	1.65			
Available nitrogen (kg ha ⁻¹)	263.42			
Available phosphorus (kg ha ⁻¹)	22.76			
Available potassium (kg ha ⁻¹)	211.37			

 Table 2: Changes in available-P (mg kg⁻¹) content in paddy soil

 applied with rock phosphate, single super phosphate and phosphorus

 solubilizing bacteria

Treatments	Days After Sowing				
Treatments	25	50	75	100	Harvest
T1	24.55	27.81	20.33	14.10	7.86
T2	31.71	35.37	23.38	19.37	12.73
T3	28.50	32.89	21.79	18.65	11.18
T4	30.49	34.56	25.07	19.92	13.21
T5	33.57	36.52	23.97	21.01	12.23
T6	29.81	33.74	23.18	17.17	12.18
T7	34.75	38.30	25.66	21.49	14.28
T8	31.42	35.54	25.12	19.88	10.98
Т9	35.39	40.16	26.71	21.58	13.41
T10	36.92	41.60	30.18	24.64	14.22
T11	33.43	38.72	26.56	20.65	11.89
S.E.d(±)	0.69	0.69	0.90	0.72	0.61
CD(p=0.05)	1.43	1.43	1.88	1.51	1.28

 Table 3: Phosphorus uptake (mg plant⁻¹) by paddy plant grown in soil applied with rock phosphate, single super phosphate and phosphorus solubilizing bacteria

Turstanta	Days After Sowing				
1 reatments	25	50	75	100	Harvest
T1	12.53	29.52	46.34	56.23	55.83
T2	14.64	39.99	67.95	66.61	81.54
T3	19.10	39.88	69.25	71.08	83.23
T4	18.06	49.54	73.89	77.77	83.26
T5	19.92	41.93	75.14	86.01	92.21
T6	21.30	49.13	77.72	81.89	92.53
T7	18.77	47.39	79.58	88.49	103.45
T8	23.33	55.21	83.01	88.91	116.60
T9	22.55	58.51	83.43	106.16	117.76
T10	24.84	58.80	87.21	117.28	134.98
T11	22.30	56.14	84.80	110.11	121.63
S.E.d(±)	0.68	0.83	1.76	1.15	1.22
CD(p=0.05)	1.41	1.72	3.65	2.38	2.54

4. Conclusions

The research findings presented in this study shed light on the significant impact of phosphorus fertilizers and PSB on soil phosphorus availability and phosphorus uptake. The results demonstrated that the combined application of single super phosphate (SSP) and rock phosphate (RP) along with Phosphorus solubilizing bacteria (PSB), can substantially increase the pool of available phosphorus in the soil and enhance phosphorus uptake by paddy. From the study, it can be concluded that application of rock phosphate and SSP at 50:50 in combination with PSB maintains a constant pool of P

for availability and higher P uptake by paddy in acid soil of Manipur.

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