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Title: Mineralization of soluble P fertilizers and insoluble rock phosphate in response to phosphate solubilizing bacteria and poultry manure and their effect on the growth and P utilization efficiency of Chilli (*Capsicum annuum* L.)

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Short Title: Efficiency of Rock Phosphate with PSB

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Mineralization of soluble P fertilizers and insoluble rock phosphate in response to phosphate solubilizing bacteria and poultry manure and their effect on the growth and P utilization efficiency of Chilli (*Capsicum annuum* L.)

Abstract

The ability of soil microorganisms and organic manures to convert insoluble phosphorus (P) to an accessible form offers a biological rescue system for improving P utilization efficiency in soil-plant systems. Our objective was to examine the P mineralization potential of two soluble P fertilizers (SPF) i.e. single super phosphate (SSP) and di-ammonium phosphate (DAP) and insoluble rock phosphate (RP) with and without phosphate solubilizing bacteria (PSB) and poultry manure (PM) and their subsequent effect on the growth, yield and P-utilization efficiency (PUE) of chill (*Capsicum annuum* L.). An incubation study was carried-out on a loam slightly alkaline soil with 12 treatments including T_0 : control; T_1 : RP; T_2 : SSP; T_3 : DAP; T_4 : PM; T_5 : ½RP+½SSP; T_6 : ½RP+½DAP; T_7 : ½RP+½PM; T_8 : RP+PSB; T_9 : ½RP+½SSP+PSB; T_{10} : ½RP+½DAP+PSB; T_{11} : ½RP+½PM+PSB. Phosphorus mineralization was measured by analyzing extractable P from the amended soil incubated under controlled condition at 25 ^{0}C for 0, 5, 15, 25, 35, 60 days period. A complementary greenhouse experiment was conducted in pots with chilli (*Capsicum annuum* L.) as a test crop. Growth, yield, P-uptake and PUE of the chilli was determined during the study. Results indicated that P mineralization in soil amended with RP was 6.0–11.5 mg kg^{-1}, while both soluble P fertilizers resulted in 68-73 mg P kg^{-1} at day 0, which decreased by 79-82% at the end of incubation study. Integrated use of PSB and PM with RP in T_{11} stimulated P mineralization by releasing a maximum of 25 mg P kg^{-1} that was maintained at high levels without any loss. Use of PSB decreased soil pH. In the greenhouse experiment, RP alone or RP+PSB did not have a significant impact on plant growth. However, combined use of RP, PM and PSB in T_{11} resulted in similar growth, yield and P-uptake of chilli as DAP. The PUE of applied P varied from 4 to 29% and was higher in the treatments that included PSB. We conclude that use of PSB and PM with insoluble RP or with soluble P fertilizers could be a promising approach to enhance P availability from both low-grade RP and SPF for crop production in intensive cropping systems.

1 Introduction

Phosphorus (P) is the second important key plant nutrient which affects the overall growth of plants by influencing the key metabolic processes such as cell division and development, energy transport (ATP, ADP), signal transduction, macromolecular biosynthesis, photosynthesis and respiration (Shenoy and Kalagudi, 2005; Khan et al., 2009, Khan et al., 2014). Soils contain very little total P of 0.02–0.5% (w/w) (Fernandez et al., 2007), of which only 0.1% is available to plants (Zou et al., 1992). Thus, P needs to be applied to soils as soluble P fertilizers, a smaller part (1%) is utilized by plants and remainder (~99%) is rapidly converted into insoluble complexes (Mehta et al., 2014) due to precipitation reactions with Al^{3+} and Fe^{3+} in acidic, and Ca^{2+} in calcareous soils (Khan et al., 2009). These metal ion complexes precipitate about 80% of added P fertilizer. Hence, the recovery efficiency of P is not more than 20% of applied P in the world soils (Qureshi et al., 2012). Considering the low recovery of applied and native P and the high cost of chemical phosphatic fertilizers besides increasing concern of environmental degradation (Aziz et al., 2006; Khan et al., 2014), it is important to find viable solutions to increase P fertilizer use efficiency. Two management options can be effective: i) increase the recovery and solubility of applied P fertilizers and ii) to replace the expensive chemical P fertilizers with novel, cheaper, more ecologically friendly but nevertheless efficient P sources, such as use of indigenous rock phosphates (RPs).

Interest in the use of RPs as alternative P sources has been increased in recent times due to their relative low cost and utilization potentials (Zapata and Zaharah, 2002; Akande et al., 2010). It has been suggested that the production of P fertilizer from RP is estimated to peak within the next 30 years because of the rising costs of synthetic fertilizers presently available in the market (Cordell et al., 2009; Beardsley, 2011; Ekelöf et al., 2014). Application of RPs directly to the soils has yielded some positive results in acidic soils but the efficacy of such material is almost negligible in neutral and alkaline soils (Begum et al., 2004). However, there are reports that Syrian RP was an effective P fertilizer for rape plants (*Brassica napus* L.) in alkaline soil (pH=7.72; Habib et al., 1999) and for maize grown in an acidic Lilysoil (pH=3.95; Alloush and Clark, 2001). Therefore, efforts have been made to find suitable ways to improve the solubility and efficiency of indigenous RPs.

Numerous studies have been conducted to evaluate the efficiency of different amendments to increase the availability and solubility of P from native and applied sources including RP. Among these, organic amendments including animal manures, plant residues, and green manures (Alloush, 2003; Toor, 2009; Aria et al., 2010; Adesanwo et al., 2012), composts (Nishanth and Biswas, 2008; Wickramatilake et al. 2010; Saleem et al., 2013), and bacterial inoculation (Panhwar et al., 2011; Gupta et al. 2011) are considered beneficial for improving the P efficiency. In addition, combined application of water soluble P fertilizers with RP is another option to increase the efficiency of RP-P. Mashori et al. (2013) used maize as a test crop in a pot experiment to examine the relative performance of RP, SSP, RP+SSP with and without FYM. They reported that RP+SSP (25+75%) with FM (10 t ha^{-1}) (RP+SSP+FYM) increased maize growth, dry matter, leaf P content and P uptake followed by the treatment receiving RP+SSP (50+50%).

Soil microorganisms have generally been found effective in making P available to the plants from both inorganic and organic sources by solubilizing and mineralizing complex P compounds (Wani et al., 2007; Khan et al., 2014). In particular, P-solubilizing bacteria (PSB) are reported to play a significant role in increasing P efficiency of both native and applied P and improving the growth and yield of various crops (Khan et al., 2009). It is generally accepted that the mechanism of P solubilization by PSB is associated with the release of low molecular weight organic acids (Goldstein, 1995; Kim et al., 1997), which through their hydroxyl and carboxyl groups chelate the cations bound to phosphate, thereby converting it into soluble forms (Kpomblekou and Tabatabai, 2003; Chen et al., 2006).

Similarly, application of organic manures with phosphatic fertilizers is considered another possible means of mobilizing P because of the acidic environment generated during decomposition of the manures (Nishanth and Biswas, 2008). These organic manures increase the microorganisms, release acids in the root rhizosphere and may help to solubilize P and to increase P availability to the plants (Fankem et al. 2006; Hu et al., 2006). In addition, combined use of RP, soluble P fertilizers and bacterial inoculation is also considered an option that may increase the efficiency of both RP and soluble P fertilizers. Experimentations on this option are not common however, recently it has been reported that 50% of triple super phosphate (TSP) could be substituted with RP when P-solubilizing bacterial inoculants *Enterobactor gegovie*, *Bacillus pumilus*, and *Bacillus subtilis* were applied with RP to wetland rice both under pot and field conditions (Rajapaksha et al., 2011).

Keeping in view the considerable expense involved in importing raw material for manufacturing P fertilizers or P fertilizers directly imported, it is imperative to explore the possibility of the utilization of indigenous RPs and the ways to increase the efficiency of other P fertilizers. Effects of PSB or organic manures on the efficiency of both soluble and insoluble P fertilizers with regard to plant growth and yield have been studied and are a topic of interest these days. However, the effect of these combinations on P release capacity (mineralization) of both soluble and insoluble P sources especially RPs has been given little attention. Therefore, the present study was conducted to examine the effect of poultry manure (PM) and PSB with soluble P fertilizers (SSP and di-ammonium phosphate, DAP) and insoluble rock phosphate (RP) on P mineralization capacity and their subsequent effect on growth, yields, P-uptake and P utilization efficiency (PUE) of chilli (*Capsicum annuum* L.) grown in greenhouse.

2 Materials and methods

2.1. Soil sampling/collection

Surface bulk soil (0–15 cm) from a field under long-term wheat – maize management system in the Faculty of Agriculture, The University of Poonch, Rawalakot Azad Jammu and Kashmir, Pakistan was collected during spring 2013. The soil used in the experiment was classified as a Humic Lithic Eutrudepts (Inceptosols). The field fresh soil was passed through a 2 mm sieve to eliminate coarse rock and plant material, thoroughly mixed to ensure uniformity and stored at 4^0C prior to use (not more than two weeks' time). A sub–sample of about 500 g was taken, air dried and passed through 2-mm sieve and used for the determination of physical and chemical characteristics (Table 1). Soil texture was determined by the hydrometer method. Soil pH was determined in a 1:2.5 (w/v) soil/water suspension. Soil organic carbon was determined by oxidizing organic matter in soil samples with $K_2Cr_2O_7$ in concentrated sulphuric acid followed by titration with ferrous-ammonium sulphate (Nelson and Sommers, 1982). Total N was determined by Kjeldahl distillation and titration method (Bremner and Mulvaney, 1982). Available P from soil samples was determined according to Soil and Plant Analysis Laboratory Manual (Ryan et al., 2001) using AB-DTPA method modified by Soltanpour and Workman (1979). Exchangeable K was determined using a flame photometer following soil extraction with 1 N ammonium acetate ($COOCH_3NH_4$) (Simard, 1993). The bulk density (BD) was determined from undisturbed soil cores taken from the upper horizon (0–15 cm) at about five locations from the field. Bulk density of the soil was calculated on a volume basis (Blake and Hartge, 1986).

2.2 Collection of added amendments/materials

The different amendments used in this study were rock phosphate (RP), single super phosphate (SSP), di-ammonium phosphate (DAP), poultry manure (PM) and P-solubilizing bacteria (PSB). Rock phosphate was collected from the Land Resources Research Institute (LRRI), NARC Islamabad, Pakistan. Major reserves of this RP are found in lagarban region of Hazara division positioned in the North East of Pakistan (Mashori et al., 2013). According to Memon (2005), the RP contains an average of 25.8% P_2O_5 along with 6% MgO. Di ammonium phosphate and SSP were purchased from the local market, while PM was collected from local farms located near by the university campus. A composite sample of well-dried PM was taken, crushed into smaller particles by hand pressing, homogenized, and passed through a 1–mm sieve before use. Total N in PM was determined by the Kjeldahl method of digestion and distillation (Bremner and Mulvaney, 1982). The P content was determined by the vanadomolybdate yellow color using spectrophotometer. Total N and P in PM were 2.53% and 1.64%, respectively. The bio-power inoculant of PSB was provided by the National Institute of Biology and Genetic Engineering (NIBGE), Faisalabad, Pakistan. The inoculant used in this study was a commercialized product containing K-1 (*Pseudomonas stutzeri*) as nitrogen fixer, ER-20 (*Azospirillum brasilense*) as IAA producer and Ca-18 (*Agrobacterium tumefaciens*) as phosphate solubilizer.

2.3 Experimental Procedures and details–incubation study

The fresh soil samples stored in the refrigerator were taken and transferred into a glass jar. Soil samples were pre-incubated at 25 ^{0}C for 1-week prior to actual incubation to stabilize the microbial activity. A known weight of soil (30 g oven dry weight basis) was taken and transferred into 100 mL capacity jars. Moisture content of soil was adjusted to 60% of iwater holding capacity (WHC) by adding deionized water. There were 12 treatments including control i.e. (unfertilized control); RP, SSP; DAP; PM; ½RP+½SSP; ½RP+½DAP; ½RP+½PM; RP+PSB; ½RP+½SSP+PSB; ½RP+½DAP+PSB; ½RP+½PM+PSB, six incubation periods: 0, 5, 15, 25, 35, 60 days (after adding amendments) and three replications. A total of 216 treatment combinations (experimental units) were used at the start of the experiment.

Phosphorus from all the treatments/sources was applied on an equivalent basis i.e. at the rate of 90 mg P kg^{-1} soil as this is a recommended optimum P application rate for chilli production in the region. Nitrogen was also added to each jar (including control) at the rate of 100 mg N kg^{-1} as urea. The amount of N added as urea was adjusted after taking into account the amount supplied by DAP and PM. Following the addition of all amendments, soil was thoroughly mixed and the weight of each jar was recorded. Jars were covered with parafilm which was perforated with a needle to ensure natural gas exchange. All the amended jars were kept in an incubator at 25± 2 ^{0}C for a total of 60 days. Jars in the incubator were arranged according to the completely randomized design. Soil moisture was checked/adjusted after every 2 days by weighing the jars the required amount of distilled water was added when the loss was greater than 0.05 g. During this process, care was taken not to disturb the soil either through stirring or shaking.

2.4 Soil extraction and analysis

Samples of all treatments incubated for different time intervals were analyzed for changes in soil available P and pH. Triplicate samples from each treatment was taken from the incubator at 0, 5, 15, 25, 35, 60 days and analyzed for available P by AB–DTP extraction method (Soltanpour and Workman, 1979). Soil (20 g) was weighed in 125 mL Erlenmeyer flasks and 40 mL of extraction solution was added (1:2). The soil available P was measured by ammonium molybdate (Murphy and Riley, 1962) using spectrophotometer. At each sampling time, the remaining 10 g soil from each jar was taken and used for measuring the changes in pH in response to different amendments. The soil pH was determined using a glass electrode in 1:2.5 (v/v) soil/water suspension.

2.5 Experimental procedures and details – greenhouse experiment

To complement the incubation study, a greenhouse experiment was conducted in pots with chilli (*Capsicum annuum* L.) as a test crop. Seedling nursery of chilli was grown by making nursery beds in the greenhouse during the last week of April 2013. Chilli seeds of variety 'Pusa Jawla' were sown separately on ridges. All the necessary culture practices were carried-out when needed. About 12 kg soil (passed through a 4-mm sieve) was placed in the cleaned earthen pots of 38 cm height and 18 cm width. There were 12 treatments including a control i.e. (unfertilized control); RP, SSP; DAP; PM; ½RP+½SSP; ½RP+½DAP; ½RP+½PM; RP+PSB; ½RP+½SSP+PSB; ½ RP+½DAP+PSB; ½ RP+½PM+PSB (same as used in incubation study) with four replications to form a total of 48 treatment combinations. Pots were arranged according to a completely randomized design. Addition of different amendments was made according to the methods/procedures followed in the incubation study. However, PSB was grown in LB broth to lag phase, containing about 10^8 $CFUml^{-1}$, applied to respective treatments by dipping roots of chilli plants in inoculum up to 20 minutes. On attaining 5-8 leave stage, four healthy and vigorous plants from the nursery were transplanted into each pot. All pots were equally irrigated when needed. The soil was moistened with water and maintained at 58% water-filled pore space throughout the study.

Plant sampling was done at two stages of development i.e. one at vegetative stage (just before flowering) for measuring growth traits of plant including shoot length, root length, shoot dry weight, root dry weight, and shoot P contents, the second one at physiological maturity stage for measuring growth, shoot P contents and yield traits i.e. fruit length, number of fruits per plant, number of seeds per fruit, fruit yield and fruit P contents.

For the determination plant P content, vegetative tissue of a plant (shoot + leaves) and fruits were washed, cleaned, and then oven dried at 70 ^{0}C for 48 hours. The oven-dried samples were ground to pass through a 1-mesh sieve in a Micro Wiley Mill. The total P was determined after digestion in a triple acid mixture ($HClO_4$, H_2SO_4, and HNO_3 in the ratio of 1:3:9). Total P in the acid digest was determined by the vanadomolybdate phosphoric yellow color method (Olsen and Sommers, 1982). The P uptake and P utilization efficiency was computed according to the methods reported earlier (Abbasi et al., 2010).

At the end of the experiment i.e., after the final crop harvest, soil samples from each pot were taken to examine changes in soil properties. A composite sample from each pot was collected and air dried for 2 to 3 days. The samples were ground and sieved to pass through a 2-mm mesh to remove rocks and large organic residues if present. Soil organic matter, total N, available P, K and pH of soil from each treatment were determined according to methods described earlier in Table 1.

2.6 Statistical analysis

All data from incubation experiment were statistically analyzed by multifactorial analysis of variance (ANOVA) using the software package Statistix 8.1. Least significant differences (LSD) are given to indicate significant variations between means of either treatments or time intervals. Confidence values (*P*) are given in the text for the significance between treatments, time interval and their interactions. Data from the greenhouse experiment was analyzed by one way analysis of variance and LSD is given to indicate significance variations among different treatments. A probability level of $P \leq 0.05$ was considered significant for both experiments.

3 Results

3.1 P release capacity (mineralization) of added amendments

Phosphorus release capacity (mineralization) of soil amended with RP varied between 6.0–11.5 mg kg^{-1} significantly ($P \leq 0.05$) higher than the control but lower than the remaining treatments (Table 2). Application of PSB with RP in RP+PSB did not show any remarkable effect on P mineralization except that a significant increase was noticed on 25 and 60 days of incubation. Soil amended with the soluble P fertilizers i.e. SSP and DAP displayed the highest P mineralization of 73.3 and 68.5 mg kg^{-1} immediately after application (day 0). However, initial P released significant ($P \leq 0.05$) decreased with subsequent incubations and at the end only 14 mg P kg^{-1} was left in the mineral P pool. The P released from the PM amended soil was progressively increased with time and the highest P concentration of 20.2 mg kg^{-1} was recorded at day 35 compared to 10.4 mg kg^{-1} at day 0. However, this increasing trend changed at day 60 where P contents declined to a background level i.e. 9.6 mg kg^{-1}. Rock phosphate when combined with soluble P fertilizers (SSP, DAP) did not show any significant impact on P mineralization. However, P mineralization of the ½RP+½DAP+PSB and ½RP+½SSP+PSB treatments was significantly higher than their sole application throughout the incubation. Similarly, the P mineralization under ½RP+½PM+PSB exhibited an increasing trend with subsequent incubation periods (showing no losses), a trend normally not common for phosphatic fertilizers.

The overall effect of different amendments on P mineralization (averaged across incubation timings) is presented in Figure 1. Results indicated that by applying 90 mg P kg^{-1} from different P sources, RP was able to release only about 8 mg kg^{-1} compared to 5 mg P kg^{-1} in the control. Both soluble P fertilizers i.e. SSP and DAP displayed the highest P release capacity of about 30 mg kg^{-1}. The P mineralization tendency of soil amended with soluble P fertilizers+insoluble RP did not show any increasing effect. However, RP when combined with PM in ½RP+½PM released significantly higher P compared to RP treatment (80%) and equivalent to that recorded under PM treatment. Effect of PSB on P release capacity of different P amendments was significant ($P \leq 0.05$). The efficiency of RP was increased by 17% when PSB was applied with RP (RP+PSB, T_8), 12% increase with ½RP+½SSP+PSB (T_9) compared to ½RP+½SSP (T_5), 18% increase with ½RP+½DAP+PSB (T_{10}) compared to ½RP+½DAP (T_6), and 28% increase with ½RP+½PM+PSB (T_{11}) compared to ½RP+½PM(T_7).

3.2 Effect of different amendments on changes in soil pH

Effect of different P amendments and their combinations on changes in soil pH over 60 days incubation is presented in Table 3. Soil amended with DAP, PM and SSP alone or with different combinations showed the maximum pH and among all, PM and DAP had the highest pH. However, except RP+PSB, pH of all the added amendments tended to decline with time. The pH of both DAP and PM significantly decreased at the end (day

60) and the reduction in pH compared to day 0 was 8%. Among different amendments RP showed the lowest pH.

Averaged across different amendments, the data presented in Figure 2 indicated that a combination of SSP, DAP and PM with RP significantly increased RP pH from 7.62 to 7.89, 7.88 and 7.83, respectively. However, application of PSB decreased soil pH. Average pH under the treatments RP, ½RP+½SSP, ½RP+½DAP, ½RP+½PM was 7.80 while application of PSB with these four amendments tended to result in a decline of pH to 7.72. The maximum reduction in pH of about 15 units was recorded in the treatment where PSB was applied with PM.

3.3 Growth and yield characteristics of chilli

Different P treatments when applied alone or used in different combinations significantly ($p \leq 0.05$) increased chilli growth characteristics compared to the control i.e. shoot length (7-53%), root length (22-113%), shoot dry weight (SDW, 8-156%), and root dry weight (RDW, 12-108%) (Table 4). Among different P amendments, growth characteristics were maximum in the treatments under full DAP or DAP, SSP and PM with PSB. RP alone had little effect on plant growth but the response of RP+PSB over RP was: no effect on shoot length, 54% increase in root length, 50% increase in SDW and 8% increase in RDW. Application of PSB with DAP, SSP and PM displayed a significant increase in growth characteristics over treatments without PSB. The relative increase in shoot length, root length, SDW and RDW due to PSB over the treatments without PSB (as a group) was 20, 14, 51 and 32%, respectively.

Yield and yield components responses of chilli to applied P treatments are presented in Table 4. Significant differences in fruit length (18-56%), number of fruits per plant (45-226%) fruit yield per plant (10-194%) and number of seeds per fruit (13-50%) were observed between the control (no-P) and the rest of the P treatments. Significant differences in yield components were also recorded among the sources of P, with DAP (full) and ½RP+½PM+PSB producing the largest yields. Application of RP alone induced a significant increase in yields (over the control). However, the magnitude of increase was remarkably higher when PSB was combined with RP. The relative increase in fruit length, no. of fruits, fruit yield and number of seed due to RP+PSB was 18, 34, 14 and 16%, respectively compared to the RP alone. Between the two synthetic P fertilizers used (SSP, DAP), DAP showed superiority over SSP while PM also exhibited a comparative yields to DAP and SSP.

Integrated use of RP with SSP, DAP and PM (50:50) was not comparative to their full dose. However, combined used of these amendments with PSB resulted in yields significantly higher than their application without PSB and equivalent to or higher than the yields recorded under full P fertilizer treatments. For example, fruit length, no. of fruits, and fruit yield from the ½RP+½SSP+PSB, ½RP+½DAP+PSB, ½RP+½PM+PSB treatments (as a group) was significantly higher (16, 44, and 40%, respectively) than their application without PSB. The highest fruit yields (10.4 g plant^{-1}) and the highest no. of fruits per plant (21.2) were recorded from the ½RP+½PM+PSB treatment equivalent to that recorded from the full DAP (10.0 g and 19.9) but significantly higher than that from full SSP (7.2 g and 15.3).

3.4 P content, P-uptake and P utilization efficiency

The P content of plant biomass and fruits of chilli treated with different P sources and combinations was significantly ($P \leq 0.05$) higher compared to P content of the control (Table 5). Soil amended with DAP resulted in the highest P content of shoot (1.33 mg plant^{-1}) and fruit (1.57 mg plant^{-1}) as compared to SSP and other P amendments. However, fruit P content recorded from the PM and the ½RP+½PM+PSB treatment (1.54 and 1.51 mg plant^{-1}) were statistically equivalent (at par) to that recorded under DAP. P content of shoot and fruit under RP was significantly higher than the control (6 and 77%) and application of PSB with RP (RP+PSB) further increased shoot and fruit P by 6 and 5%, respectively compared to RP alone.

Application of phosphatic fertilizers had a significant effect ($P \leq 0.05$) on P-uptake of plant biomass and fruit of chilli compared to the control treatment (Table 5). The values ranged between 4.3–15.3 mg plant^{-1} for shoot and 4.4–15.7 mg plant^{-1} for fruit compared to 3.7 and 1.8 mg plant^{-1} in the control, respectively. Among different P sources and combinations, DAP exhibited the highest P-uptake while the PM and ½RP+½PM+PSB treatments showed values (for fruit P-uptake) at par (statistically equivalent) with DAP.

The total P-uptake (shoot + fruit) in the control was 5.5 mg plant^{-1} that significantly increased to 8.7–31.3 mg plant^{-1} following the application of different P sources. The DAP and ½RP+½PM+PSB treatments exhibited the highest total P-uptake and the difference between the two was non-significant. The RP treatment alone resulted in a significant increase in P-uptake (8.7 mg plant^{-1}) compared to the control (5.5 mg plant^{-1}). The effectiveness of P fertilizers with regard to plant P-uptake was in the order: DAP > PM > SSP > RP. The total P-uptake under PM was significantly higher than the SSP (31%) but lower than the DAP (20%).

Application of PSB with different P sources resulted in a significant ($P \leq 0.05$) increase in P-uptake i.e. 20% with RP, 29% with ½RP+½SSP, 56% with ½RP+½DAP and 132% with ½RP+½PM.

The P-utilization efficiency (PUE) of added P sources and their combinations ranged from 4% by RP to a maximum of 29% with DAP (Fig. 3). The PUE of SSP and PM was 14 and 23%, respectively showing higher PUE by PM compared to SSP. The PUE of RP was only 4% that increased to 6–8% when RP was combined with either PSB or SSP, DAP or PM. Results indicated a significant improvement in PUE when PSB was combined with P amendments. For example, the PUE of the ½RP+½SSP+PSB, ½RP+½DAP+PSB, ½RP+½PM+PSB treatments was 14, 19 and 27% compared to 8, 7 and 7% from the ½RP+½SSP, ½RP+½DAP, and ½RP+½PM treatments, respectively showing about 2-4fold increase in PUE due to PSB. The response of PUE to PSB was more prominent when PSB was combined with PM compared to its combination with DAP or SSP.

4 Discussion

4.1 P release capacity of added amendments

In order to determine the P release capacity (mineralization) of soluble and insoluble P fertilizers and their response to PM and PSB, an incubation study of 60 days was conducted under controlled laboratory conditions. The P release capacity of different amendments and their combinations varied with source and timings. Soluble P fertilizers i.e. SSP and DAP displayed the highest mineralization compared to the insoluble RP and organic PM. In most of the cases (except PM and combined treatment of ½RP+½PM+PSB), there was a general trend of a rapid mineralization in the first few days of incubation followed by a gradual decrease and a sharp decline thereafter. The P mineralization trend (over time) observed in this study was in accordance with the previous studies where P mineralization of different P sources significantly decreased/declined with time (Begum et al., 2004, Toor, 2009; Toor and Haggard, 2009). This decreasing trend may be ascribed due to rapid conversion of available P into insoluble complexes (Mehta et al., 2014) by entering into the immobile pools through precipitation reaction with highly reactive Ca^{2+} ions (Khan et al.,2009). The soil under investigation was slightly alkaline (pH 7.57), non-clacrious ($CaCO_3$ 0.68%) and having Ca content of 58.7 mg kg^{-1}. The soil Ca concentration was low but still its presence may contributed towards P fixation or precipitation. In addition, soil belong to Chinasi soil series and parent material is residum-colluvium from shales. Therefore, it is likely that kaolinite may be a dominant clay mineral present in soil composition that adsorb high $H_2PO_4^-$. The other possibility may be the fixation of some of the applied or native P on the surface of the clay particles in the presence of 24% clay content of the soil used in the study. As organic matter plays an important role in P solubilization through the acidifying and chelation mechanisms. The low organic matter in our soil may also be an important factor for overall low mineralization trend of P observed in this study.

Addition of RP alone released a maximum of 6% of the total P applied showing that mineralization capacity of RP-P even under favorable environmental conditions is low and the fertilizer value of this RP (alone) is quite negligible. These values were substantially lower than those reported for North Carolina and Syrian RP applied to an acid Lily soil showing P dissolutions of about 27% after 126 days of incubation. However, the observed values are in the range reported for Indian RP i.e. 6-8 mg kg^{-1} applied under alkaline conditions (pH 8.5) (Begum et al., 2004). Similarly, application of RP alone to slightly alkaline soil (pH 7.9) at Faisalabad Pakistan did not show any significant effect on bioavailable P contents of the soil (Saleem et al., 2013). These reports suggested that RP works best in acidic soils while showed poor efficiency in neutral and alkaline soils. Under acidic conditions, organic acid anions with oxygen containing $OH^-$and $COOH^-$groups, have the ability to form stable complexes with cations such as Ca_2^+, Fe_2^+, Fe_3^+ and Al_3^+, that are commonly bound with phosphate (Jones, 1998). By complexing with cations on the mineral surface, organic acid anions loosen cation-oxygen bonds of the mineral structure and catalyze the release of cations to solution (Kpomblekou and Tabatabai, 1994). This is the major reason that why RP is more effective under acidic conditions.

Effect of PSB on P release capacity of different P amendments was significant ($P \leq 0.05$). Application of PSB with RP in RP+PSB (T_8) exhibited overall 17% higher mineralization than RP alone showing a solubilizing effect of PSB on RP. Jha et al. (2013) isolated ten PSB strains and tested them for mineral phosphate solubilization activity of RP and stated that all these strains could solubilize only 0.02–2.6% of the total RP-P applied. In addition, *Aspergillus niger* (a fungus), used in the industrial production of citric acid, has been recognized as one of the most effective organisms for RP solubilization (Abd-Alla and Omar, 2001). These results suggested that i) PSB increased P solubilization of added P fertilizers either from soluble or insoluble source and ii) the relative efficiency of PSB for releasing P was higher with PM compared to soluble or insoluble P fertilizers. Khan and Sharif (2012) conducted an incubation study in soil amended with PM, PM+RP and PM+RP+EM (EM, effective microorganisms) and reported that the extractable P was significantly higher in the treatments PL+RP+EM, and PL+RP compared to PL only. Reddy et al. (2002) compared the efficiency of three isolates on the solubilization of RP and reported that all the isolates increased RP-P release efficiency by solubilize the tested RPs. Similar effects of bio- and organic fertilizers on RP availability and P fertilizers efficiency had also been reported in soils incubated for different incubation periods (Aria et al., 2010; Alzoubi and Gaibore, 2012). The mechanisms involved in the potential of PSB to solubilize P complexes or insoluble phosphates are well known and has been attributed to the processes of acidification, chelation, exchange reactions, and the production of organic acids (Chen et al., 2006; Ekin, 2010).

The mineralization of RP-P was unaffected when RP was combined with soluble P fertilizers (SSP, DAP) demonstrating that soluble P fertilizers had no solubilizing effect on RP. In contrast to our results, Begum et al. (2004) found a substantial improvement in extractable-P status when RP was combined with SSP and MAP (mono-ammonium phosphate). However, RP when combined with PM in ½RP+½PM released significantly higher P compared to the RP alone (80%) and was equivalent to that recorded under full PM treatment showing that the additional P released from RP was associated with PM. Toor (2009) found a substantial increase in soil solution P following the application of PM with P fertilizers because of the release of organic acids during decomposition of the manure and production of carbon dioxide during organic matter decomposition that may increase the solubility of Ca^{2+} and Mg^{2+} phosphates.

The PSB tented to decline soil pH showing acidifying effect. The maximum reduction in pH of about 15 units was recorded in the treatment where PSB was applied with PM. Effect of PSB on soil pH at different time intervals indicated that in some cases, addition of PSB temporarily increased soil pH more than other treatments. The general decrease in pH during the experiment could have arisen from a "move back to equilibrium" as well as due to increase in microbial activity. Our results were in accordance with the previous observations of Aira et al. (2010) and Khan and Sharif (2012) who reported a significant decrease in soil pH after applying PSB.

4.2 Growth, yield, P-uptake and P utilization of chilli

RP alone had little effect on plant growth but the response of RP+PSB over RP was: no effect on shoot length, 54% increase in root length, 50% increase in SDW and 8% increase in RDW. The difference between the two treatments is attributed to the effect of PSB on releasing P either from RP or from native soil P thereby increased plant growth. Among four main P sources used (SSP, DAP, RP and PM), DAP showed superiority over SSP and PM because of the highest P release capacity shown in the incubation study. However, the efficiency of SSP for growth and yield characteristics of chilli was significantly lower than the DAP and PM for most of the parameters studied. The P release capacity of SSP was higher than the PM throughout the incubation while the growth and yield attributes was lower. The possible reasons for this discrepancy is not understood however, in addition to supply P to plants the additional beneficial effects of PM on soil physiochemical characteristics, root proliferation and plant nutrient uptake may affect the growth and yield of plants grown in PM amended soil. The results of the present study indicated that application of RPs directly to soils had shown positive effects on root dry weight and yield components of chilli but the efficacy of RP for most of the growth characteristics was almost negligible.

Application of PSB with RP, SSP, DAP and PM or their combinations displayed a remarkable improvement in the growth and yield of chilli. The treatment which received ½RP+½PM+PSB produced growth and yield comparable to that recorded from the full DAP showing that this mixed treatment may able to save almost 50% of chemical P fertilizer. The higher response of plant growth to PSB might be due to mobilization of available P by the native soil microflora, or increased PSB activity in the rhizosphere following PSB application and consequently by enhanced P solubilization which enhanced growth and yield of plants (Ekin, 2010). Combined application of PSB and PM with P fertilizers is considered an important management strategy for mobilizing P where inert P is expected to be converted into plant available forms because of the acidic environment prevailing during decomposition of organic manures (Nishanth and Biswas, 2008) and then additional beneficial effects of PSB to the processes of acidification, chelation, exchange reactions, and the production of organic acids (Chen et al., 2006; Ekin, 2010). These combined effects increased the efficiency of applied materials thereby increased the growth and yield of the plant as observed in the present study. Our results are in accordance to the previous studies conducted on the use of organic materials and PSB for increasing the efficiency of applied P fertilizers and their subsequent effect on the growth and yields of plants (Biswas and Narayanasamy, 2006; Nishanth and Biswas, 2008; Abbasi et al., 2013).

The effectiveness of P fertilizers with regard to plant P-uptake was in the order: DAP > PM > SSP > RP. The total P-uptake under PM was significantly higher than the SSP (31%) but lower than the DAP (20%). Application of PSB with different P sources resulted in a significant ($P \leq 0.05$) increase in P-uptake i.e. 20% with RP, 29% with ½RP+½SSP, 56% with ½RP+½DAP and 132% with ½RP+½PM. These results indicated that use of PSB with PM had a dominating effect on increasing plant P-uptake compared to the application of other P sources. The overall PSB effect (group effect) displayed that total P-uptake under the treatments supplemented with PSB was 23.4 mg plant^{-1} compared to 13.2 mg plant^{-1} under the treatments without PSB showing a relative increase of 77% compared to the treatments without PSB.

Results of our incubation and pot experiment indicated that the total P uptake by the plants in response to the different amendments was significantly correlated with P mineralization ($r^2 = 0.64$) (determined at the end of the experiment at day 60) showing that P-uptake by plants is associated with the mineralization capacity of added P amendments. Similarly, the effect of added amendments on increasing root mass may also have affected the P-uptake as significant correlations were found between these two parameters ($r^2 = 0.71$). The increasing effect of P mineralization and plant root mass/density on P-uptake due to PSB and organic amendments had also been reported earlier (Lorion, 2004; Nishanth and Biswas, 2008; Abbasi et al., 2013). Wickramatilake et al. (2010) investigated P release capacity of RP treated with compost prepared from PM, cattle manure (CM), sewage sludge (SS), or P-adjusted sawdust (PSD) and reported that the uptake of P from RP by plants is enhanced by compost, especially PM or CM compost, the increase was four- to five-fold compared with no compost addition.

Results of this study showed that PUE of chemical P fertilizers commonly used in most parts of the world i.e. SSP and DAP was low i.e. 14 and 29%, respectively. However, this recovery of applied P is in accordance with the recovery efficiency of P generally reported (20–25%) (Qureshi et al., 2012). The organic P sources i.e. PM displayed higher PUE (23%) compared to SSP (14%) although the P mineralization capacity of SSP was significantly higher than PM. This favorable effect may be attributed to i) the increased P uptake by plants through enlarged proliferation of roots as the root mass of plants under PM was 17% higher than the root mass recorded under SSP ii) reduction in the activity of Ca^{2+}, Al^{3+} and Fe^{3+} ions by root exuded organic anions as reported earlier (Toor,2009).

The PUE of RP and RP+PSB was just 4 and 7%, respectively indicating that RP alone was not able to generate any positive impact as a P fertilizer. However, the PUE of RP, SSP and DAP was remarkably increased when these sources were combined either with PM or PSB. Among different combinations, ½RP+½PM+PSB showed the significant contribution by increasing PUE to 27% equivalent to that recorded under full DAP treatment. This finding highlighted the importance of RP as a P source when combined with organic and microbial amendments. The increased PUE may have resulted in increased dry matter yield (DMY), fruit yield and greater P accumulation as significant correlations existed between PUE and DMY (r^2= 0.93), fruit yield (r^2 = 0.97), PUE and shoot and fruit P concentrations (r^2= 0.86. r^2= 0.93), and PUE and shoot and fruit P uptake (r^2= 0.97).

The role of organic amendments or PSB in improving P utilization from applied P fertilizers has been reported earlier by several researchers (Begum et al., 2004; Toor, 2009, Abbasi et al., 2013). This positive effect is attributed to the fact that release of organic acids from these amendments in the root rhizosphere can reduce fixation of applied P, induce greater P availability in the soil, and form phosphor-humic complexes that are easily assimilated by plants (Toor, 2009). These mechanisms can result in greater amounts of applied P in available forms to be used by plants.

5 Conclusions

Results of our incubation experiment indicate that chemical P fertilizers used in the study i.e. SSP and DAP released the highest P at the start of the experiment but this mineral P significantly decreased with subsequent incubation periods. At the end of the experiment (at day 60) about 80% of P initially present had disappeared from the system showing that the P recovery in the soil mineral pool was 20% of the total P in the applied P fertilizers. RP alone or RP+PSB released a maximum of only about 12 mg P kg^{-1} demonstrating that application of RP directly to the soil having neutral pH did not show any positive effect on overall P mineralization. However, use of PSB and PM with RP in ½RP+½PM+PSB treatment released a substantial amount of available P (25 mg kg^{-1}) that remained at high levels (without any loss) until the end of incubation (day 60) showing that combination of PSB and PM with RP may be a feasible option for releasing P for a longer period. This correlates well with the fact that two-thirds of the total P demand of most crops is met during the first one-third of their growth period. When these amendments were applied to chilli under greenhouse conditions, DAP exhibited the highest growth, yield and P-uptake. RP was able to increase the yield of many plant components compared to the control but was not as effective as SSP, DAP and PM. Combinations of RP with either SSP or DAP in 50:50 proportion did not show any significant effect on P mineralization and subsequent plant growth and P-uptake. However, application of ½RP+½PM+PSB showed a remarkable effect and induced growth, yields, P-uptake comparable to that recorded under the full DAP treatment. The P utilization efficiency of chilli supplemented with ½RP+½PM+PSB was not statistically different from the full DAP treatment (27% and 29%). This combination (½RP+½PM+PSB) hold a lot of promise as an efficient alternative to conventional P fertilizers especially its effectiveness for the utilization of RP. However, the results need to be confirmed under field conditions and the economic feasibility of its application needs to be quantified.

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Table 1. The initial physical and chemical characteristics of soil used in the incubation study

Soil properties	values
Bulk density (g cm^{-3})	1.32
Sand (g kg^{-1})	433.9
Silt (g kg^{-1})	326.0
Clay (g kg^{-1})	240.1
Textural class	loam
Soil pH (1:2.5H_2O)	7.57
Organic matter (g kg^{-1})	10. 3
Organic carbon (g kg^{-1})	5.64
Total N (g kg^{-1})	0.53
NH^+_4-N (mg kg^{-1})	8.85
NO^-_3-N (mg kg^{-1})	7.21
Available P (mg kg^{-1})	4.70
Available K (mg kg^{-1})	98.5
Calcium Ca (mg kg^{-1})	58.7
Magnesium Mg (mg kg^{-1})	15.5
Iron Fe, (mg kg^{-1})	17.8
Manganese Mn (mg kg^{-1})	6.2
Zinc Zn (mg kg^{-1})	8.4
Copper Cu (mg kg^{-1})	3.79
$CaCO_3$ content (%)	0.68
Cation exchange capacity (CEC) $cmol^{(+)}$ kg^{-1}soil	11.9

Table 2. Phosphorus release capacity i.e. mineralization potential of soluble P fertilizers and insoluble rock phosphate in response to phosphate solubilizing bacteria and poultry manure applied to a soil incubated under controlled laboratory conditions at 25 °C over 60 days periods.

Treatments	Days after amendments application						
	0	5	15	25	35	60	LSD ($P \leq 0.05$)
Extractable (available) P (mg kg^{-1} soil)							
Control	4.7	4.8	5.3	5.7	6.5	4.5	0.44
RP	6.0	7.7	9.9	6.2	11.5	6.2	1.02
SSP	73.3	30.5	21.6	18.8	21.7	13.5	2.25
DAP	68.4	29.4	23.0	19.5	20.5	14.1	2.37
PM	10.4	13.1	18.8	17.7	20.2	9.6	1.16
½RP+½SSP	42.9	21.0	14.6	21.0	5.8	6.9	2.34
½RP+½DAP	43.3	17.3	25.2	13.6	7.9	6.2	2.21
½RP+½PM	11.8	12.8	15.3	13.5	23.0	8.9	3.32
RP+PSB	6.1	6.3	10.4	11.5	11.4	9.8	1.13
½RP+½SSP+PSB	38.2	18.8	18.8	16.0	22.7	11.2	1.80
½RP+½DAP+PSB	44.6	20.9	16.9	16.0	23.1	13.0	2.30
½RP+½PM+PSB	12.7	12.4	16.8	17.5	25.2	24.2	2.21
LSD ($P \leq 0.05$)	1.23	2.11	1.45	1.11	1.21	1.15	

*RP =rock phosphate; SSP = single super phosphate; DAP = di-ammonium phosphate; PM = poultry manure; PSB = phosphate solubilizing bacteria; full dose of P from different sources was applied at the rate of 90 mg P kg^{-1} soil.

Table 3. Changes in pH of the soil supplemented with soluble P fertilizers and insoluble rock phosphate along with phosphate solubilizing bacteria and poultry manure and incubated under controlled laboratory conditions at 25 ^{0}C over 60 days periods.

Treatments	Days after amendments application						
	0	5	15	25	35	60	LSD (P≤0.05)
pH							
Control	7.57	7.74	7.82	7.68	7.6	7.29	0.11
RP	7.57	7.65	7.87	7.76	7.5	7.39	0.08
SSP	7.93	7.91	7.85	7.76	7.86	7.43	0.16
DAP	8.00	7.94	8.00	7.98	7.81	7.34	0.06
PM	8.10	7.93	8.07	8.10	7.96	7.49	0.13
½RP+½SSP	7.90	7.91	7.89	8.07	7.99	7.56	0.08
½RP+½DAP	7.92	7.96	8.01	8.03	7.71	7.62	0.09
½RP+½PM	7.89	7.93	7.78	8.03	7.68	7.66	0.10
RP+PSB	7.52	7.59	7.46	7.47	7.69	7.63	0.09
½RP+½SSP+PSB	7.93	7.91	7.91	7.65	7.65	7.58	0.09
½RP+½DAP+PSB	7.95	7.92	7.93	7.77	7.69	7.63	0.08
½RP+½PM+PSB	7.95	7.87	7.75	7.66	7.59	7.54	0.07
LSD (P≤0.05)	0.13	0.08	0.16	0.10	0.10	0.07	

*RP =rock phosphate; SSP = single super phosphate; DAP = di-ammonium phosphate; PM = poultry manure; PSB = phosphate solubilizing bacteria; full dose of P from different sources was applied at the rate of 90 mg P kg^{-1} soil.

Table 4. Effect of soluble P fertilizers and insoluble rock phosphate applied alone or in combination with phosphate solubilizing bacteria and poultry manure on the growth and yield characteristics of chilli (*Capsicum annuum* L.) grown in pots under greenhouse conditions at Rawalakot Azad Jammu & Kashmir.

Treatments	Shoot length (cm)	Root length (cm)	Shoot dry wt. (g plant^{-1})	Root dry wt. (g plant^{-1})	Fruit length (cm)	No of seeds fruit^{-1}	No. of fruits plant^{-1}	Fruit yield (g plant^{-1})
Control	30.0	8.8	4.8	1.13	6.1	33.7	6.7	3.5
RP	32.0	10.7	5.2	1.26	7.2	38.0	8.7	4.9
SSP	39.0	17.4	8.4	1.33	7.8	50.0	15.3	7.2
DAP	43.3	15.6	11.8	1.76	9.3	49.0	19.9	10.0
PM	31.3	14.2	10.9	1.56	8.5	42.7	13.7	9.9
½RP+½SSP	33.3	14.5	6.4	1.37	7.5	41.3	11.7	5.8
½RP+½DAP	34.2	17.5	7.9	1.42	7.7	48.7	13.0	6.6
½RP+½PM	32.2	16.2	6.9	1.36	7.6	50.0	13.3	6.4
RP+PSB	30.3	16.4	7.8	1.35	8.5	44.0	10.6	5.6
½RP+½SSP+PSB	32.3	16.7	9.5	1.40	8.4	40.3	14.9	7.2
½RP+½DAP+PSB	43.7	15.7	10.2	1.73	8.5	42.3	18.5	8.7
½RP+½PM+PSB	45.8	18.7	12.3	2.35	9.5	50.3	21.2	10.4
LSD ($P \leq 0.05$)	3.7	1.73	1.81	0.09	0.74	4.10	1.7	0.8

*RP =rock phosphate; SSP = single super phosphate; DAP = di-ammonium phosphate; PM = poultry manure; PSB = phosphate solubilizing bacteria; full dose of P from different sources was applied at the rate of 90 mg P kg^{-1} soil.

Table 5. Effect of soluble P fertilizers and insoluble rock phosphate applied alone or in combination with phosphate solubilizing bacteria and poultry manure on P content and P-uptake of chilli (*Capsicum annuum* L.) grown in pots under greenhouse conditions at Rawalakot Azad Jammu & Kashmir.

Treatments	Shoot P (mg g^{-1})	Fruit P (mg g^{-1})	Shoot P-uptake (mg $plant^{-1}$)	Fruit P-uptake (mg $plant^{-1}$)	Total P-uptake (mg $plant^{-1}$)
Control	0.78	0.50	3.7	1.8	5.5
RP	0.83	0.89	4.3	4.4	8.7
SSP	1.03	1.30	8.7	9.3	18.0
DAP	1.33	1.57	15.3	15.7	31.3
PM	1.01	1.54	11.0	15.2	26.2
½RP+½SSP	0.89	1.12	5.7	6.5	12.2
½RP+½DAP	0.92	1.08	7.3	7.1	14.4
½RP+½PM	0.90	1.06	6.2	6.8	13.0
RP+PSB	0.88	0.94	6.9	5.3	12.1
½RP+½SSP+PSB	0.98	1.17	9.3	8.4	17.7
½RP+½DAP+PSB	1.10	1.28	11.2	11.2	22.4
½RP+½PM+PSB	1.17	1.51	11.4	15.5	30.1
LSD ($P \leq 0.05$)	0.11	0.13	1.81	1.95	2.11

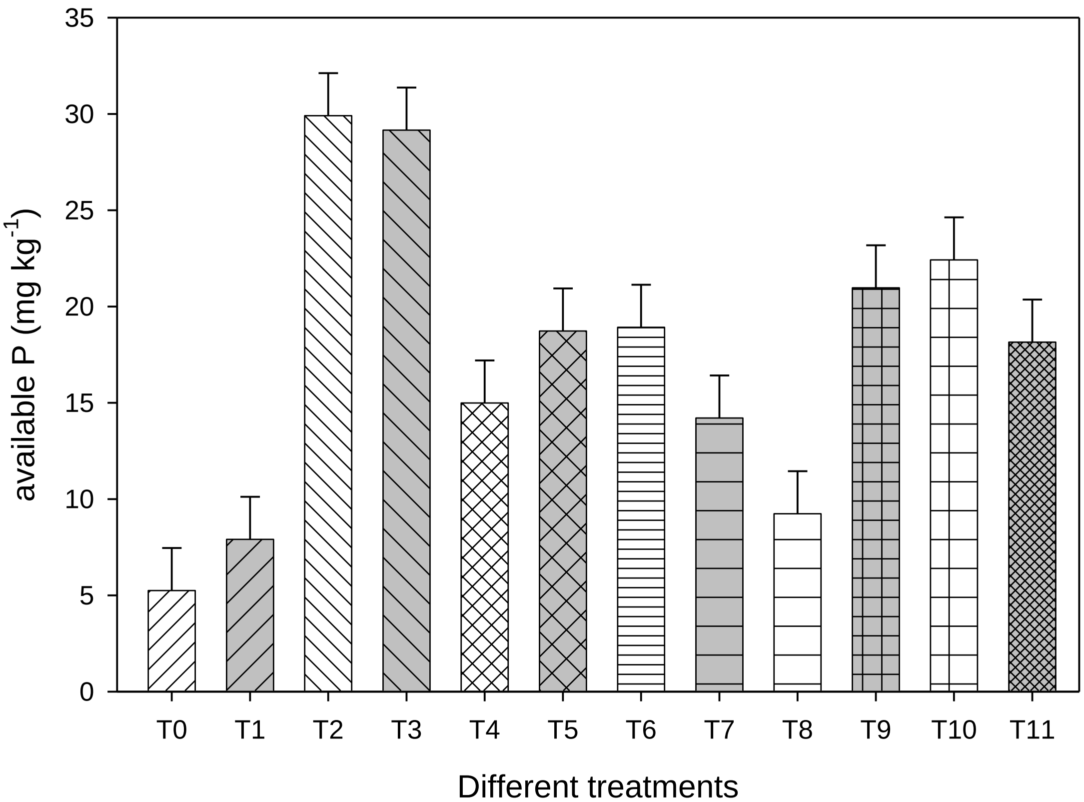
* RP= Rock Phosphate; SSP= Single Super Phosphate; DAP= Di ammonium Phosphate; PM= Poultry Manure; PSB= Phosphate Solubilizing Bacteria; full dose of P from different sources was applied at the rate of 90 mg P kg^{-1} soil. The treatments represents

FIGURE CAPTIONS

Figure 1. The P release capacity of different P sources applied alone or in combination with PSB and PM (average over incubation periods) to a soil incubated under controlled laboratory conditions at 25^{0}C. The legend at x-axis represent T_0 = control; T_1 = RP; T_2 = SSP; T_3 = DAP; T_4 = PM full; T_5 = ½RP+½SSP; T_6 = ½RP+½DAP;T_7 = ½RP+½PM; T_8 = RP+PSB; T_9 = ½RP+½SSP+PSB; T_{10} = ½RP+½DAP+PSB; T_{11} = ½RP+½PM+PSB. Full dose of P from different sources was applied at the rate of 90 mg P kg^{-1} soil.

Figure 2. Changes in pH of the soil (average over incubation periods) supplemented with different P sources applied alone or in combination with PSB and PM and incubated at controlled laboratory conditions at 25^{0}C. The legend at x-axis represent T_0 = control; T_1 = RP; T_2 = SSP; T_3 = DAP; T_4 = PM full; T_5 = ½RP+½SSP; T_6 = ½RP+½DAP;T_7 = ½RP+½PM; T_8 = RP+PSB; T_9 = ½RP+½SSP+PSB; T_{10} = ½RP+½DAP+PSB; T_{11} = ½RP+½PM+PSB. Full dose of P from different sources was applied at the rate of 90 mg P kg^{-1} soil.

Figure 3. P utilization efficiency of chilli grown under greenhouse conditions following the application of different P sources applied alone or in combination with PSB and PM. The legend at x-axis represent T_1 = RP full; T_2 = SSP; T_3 = DAP; T_4 = PM; T_5 = ½RP+½SSP; T_6 = ½RP+½DAP;T_7 = ½RP+½PM; T_8 = RP+PSB; T_9 = ½RP+½SSP+PSB; T_{10} = ½RP+½DAP+PSB; T_{11} = ½RP+½PM+PSB. Full dose of P from different sources was applied at the rate of 90 mg P kg^{-1} soil.



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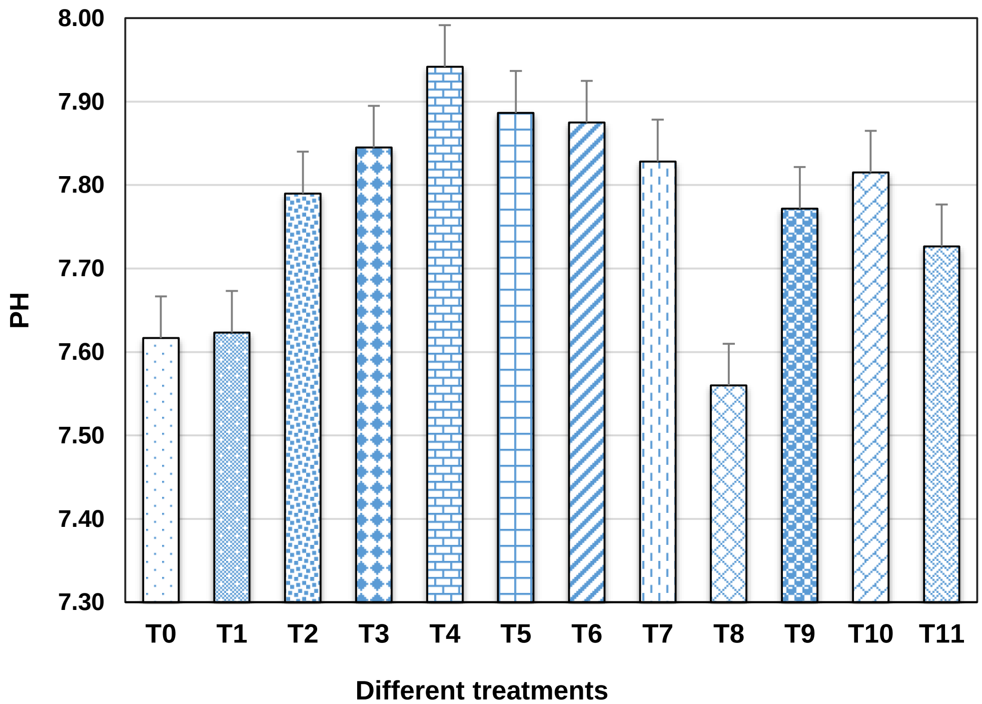
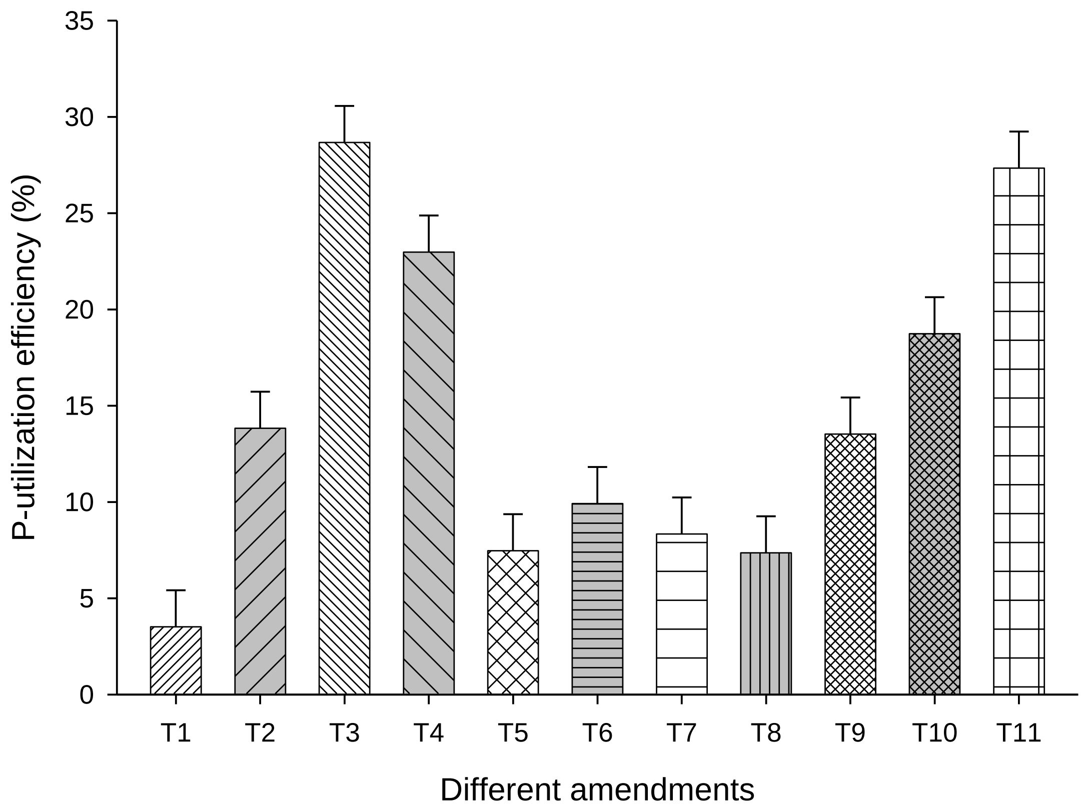


Figure 2. Changes in pH of the soil (average over incubation periods) supplemented with different P sources applied alone or in combination with PSB and PM and incubated at controlled laboratory conditions at 25^{0}C. The legend at x-axis represent T_0 = control; T_1 = RP; T_2 = SSP; T_3 = DAP; T_4 = PM; T_5 = ½RP+½SSP; T_6 = ½RP+½DAP;T_7 = ½RP+½PM; T_8 = RP+PSB; T_9 = ½RP+½SSP+PSB; T_{10} = ½RP+½DAP+PSB; T_{11} = ½RP+½PM+PSB. Full dose of P from different sources was applied at the rate of 90 mg P kg^{-1} soil.



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