

## Enhancing Extractable Phosphorus from Phosphate Rock in Some Soils by Amending With: Organic Acids, Sulfur, Farmyard Manure, Ammonium Thiosulfate and Phosphate Solubilizing Bacteria

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### ABSTRACT

Phosphorus is an important plant nutrient and therefore has a major impact on the productivity of agriculture. Phosphate rock is spread as natural mined rock in many locations around the globe and in Egypt's western desert. Depending on the source of the mine, the significance of phosphate rock is linked to its elevated phosphorous element content from 16 to 25 percent of P<sub>2</sub>O<sub>5</sub>. Many approaches and techniques have been followed for the treatments of phosphate rock to get benefits of its content from extractable phosphorous. A set of laboratory experiments were conducted to investigate phosphate-dissolution ability from phosphate rock (PR) in clayey and sandy soils as a function of incubation time. The different amendments used in this study oxalic acid (OA), citric acid (CA), sulfur (S<sup>0</sup>), farmyard manure (FYM), ammonium thiosulfate (ATS) and phosphate solubilizing bacteria (PSB) and incubated at 25°C for 14, 28, 42, 56 and 70 days. The obtained results indicated that the availability of P from phosphate rock was increased by increasing incubation time up to 70 days. The useful data gained refers to the fact that blending organic acids, sulfur, ATS, FYM or PSB with phosphate rock considerably significantly enhanced extractable P over PR alone. In example, incubation PR with oxalic acid increased extractable P in clay and sandy soil by 3.49 and 5.2 times that of the treatment PR alone, respectively. Moreover, pH of all the added amendments in clayey and sandy soils tended to decline with time, except control, PR or SSP. In comparison with PR alone, the pH obtained from PR with oxalic, citric, sulfur, ATS, FYM and PSB decreased significantly at the end of incubation.

**Keywords:** Phosphate rock, Elemental sulfur, Farmyard manure, Citric acid, Oxalic acid, Phosphate solubilizing bacteria, Ammonium thiosulfate.

### Introduction

Phosphorus (P) is the second most common growth-limiting macronutrient responsible for lower crop production. It is associated with many metabolic components and enzymatic reactions within the plant. Unavailability of P to plants during early growth stages results in reduced root growth and seed formation (Ahmad *et al.*, 2018). The availability of P to plants depends on its concentration in soil solution which subsequently depends on several factors such as soil mineralogy, pH and concentration of soluble Ca, Fe and Al, organic matter and microbial activities. Soils contain very little of P (0.02–0.5%, which only 0.1% is available to plants (Fernandez *et al.*, 2007). Thus, P needs to be applied to soils as soluble P fertilizers; a small part (1 %) is utilized by plants and the remainder (~99 %) is rapidly converted into insoluble complexes due to precipitation reactions with Al<sup>3+</sup> and Fe<sup>3+</sup> in acidic soil and Ca<sup>2+</sup> in calcareous soils. These metal ion complexes precipitate about 80% of the added P fertilizer. The retrieval effectiveness of P in world soils therefore does not exceed 20% of the P used (Mehta *et al.*, 2014).

Considering the low recovery of applied and native P and the high cost of chemical phosphatic fertilizers in addition to an increasing concern about environmental degradation (Khan *et al.*, 2014). It

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is essential to discover feasible alternatives to improve the effectiveness of P fertilizer use. In recent times, interest in using PRs as alternative P sources has increased due to their relatively low cost and potential for utilization (Akande *et al.*, 2010). In arid and semi-arid regions, the application of PR is not common because of high pH, low organic matter as well as less solubility of P in these soils.

Numerous investigations on soil pH, particle size of PR and concentrations of Ca and P in soil solution have examined the solution of PR in soils and its subsequent effect on soil availability (He *et al.*, 2005). Many traditional techniques are used such as thermal alteration or partial acidulation require substantial capital investments in order to increase the solubility of P from PR, however alternative approaches are also required to increase the availability of P from PR (Khasawneh and Doll 1979). In order to improve the accessibility and solubility of P from indigenous and applied sources including PR, numerous studies have been performed to assess the effectiveness of various modifications. Among these, organic amendments, including animal manure, plant residues, green manure, composts and bacterial inoculation are considered beneficial for improving the P efficiency (Saleem *et al.*, 2013). Furthermore, combining water-soluble P fertilizers with PR is another option to increase the efficiency of PR.

Mashori *et al.* (2013) used maize as a test crop to examine the comparative efficiency of PR in a pot experiment, single superphosphate (SSP) and PR with SSP with and without farmyard manure (FYM). They reported that PR with SSP (25+75 %) with FYM (10t ha<sup>-1</sup>) (PR with SSP with FYM) increased maize growth, and P uptake; the next highest increase was seen in the treatment receiving PR with SSP (50+50 %).

Soil microorganisms have generally been found to make P available to plants from both inorganic and organic sources through the solubilization and mineralization of complex P compounds (Khan *et al.*, 2014). In particular, P-solubilizing bacteria (PSB) are reported to play a significant role in increasing the 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 (Kim *et al.*, 1997), which through their hydroxyl and carboxyl groups chelate the cations bound to phosphate, thereby converting it into soluble forms (Chen *et al.*, 2006). Similarly, the use of manure with phosphatic fertilizers is considered to be another possible means of mobilizing P due to the acidic environment generated during manure decomposition. (Nishanth and Biswas, 2008). The various types of manure increase the microorganisms, release acids in the root rhizosphere and can increase the availability of P (Fankem *et al.* 2006). In addition, the combined use of PR, soluble P fertilizers and bacterial inoculation is also considered an option that may increase the efficiency of both PR and soluble P fertilizers. Experimentations on this option are not common, however; recently it has been reported that 50% of triple superphosphate (TSP) could be substituted with PR when P-solubilizing bacterial inoculants *Enterobacter gergovie*, *Bacillus pumilus* and *Bacillus subtilis* were applied with PR to wetland rice both under pot and field conditions (Rajapaksha *et al.*, 2011).

The beneficial effect of mixing PR with elemental sulfur (S<sup>0</sup>) has been practiced using the logic that *Thiobacillus* sp. would oxidize S to H<sub>2</sub>SO<sub>4</sub>, which would then react with PR to release P. The effects of S and gypsum additions on P availability from PRs were investigated in a fine sandy soil by Neller (1956). The most common low-molecular weight organic acids (LMWOAs) identified in soils are oxalic, succinic, tartaric, fumaric, malic, citric, sinapic, caffeic, syringic, salicylic, gallic, *p*-coumaric, gentisic, protocatechuic, vanillic, *p*-hydroxybenzoic and ferulic (Kaurichev *et al.*, 1963). These organic acids exhibit chelation and complexing properties. By forming soluble complexes with polyvalent cations from rocks and minerals, organic acids play an important role in dissolution, transportation and concentration of elements in the earth's surface (Huang and Keller, 1972) as well as in soil formation and plant nutrition. Kpombrekou-A and Tabatabai (1994) studied the ability of 19 LMWOAs to release P from PRs. Their results showed that reactions involved in the P release process are not only pH-dependent but also related to the structural characteristics of the LMWOAs. Although information is available on alternative ways to increase P availability in PRs using various additives, there is only limited amount of information on the use of LMWOAs to enhance P availability in PRs. In the present study the effect of oxalic acid, citric acid, sulfur, farmyard manure, ammonium thiosulfate and phosphate solubilizing bacteria on available phosphorus of phosphate rock was investigated in order to find the best combination of examined factors.

## Materials and Methods

### Soil sampling and collection

Two surface soil samples were used (0–20 cm) from El-Nubaria, Cairo- Alexandria desert road and El-Kanater farm, qaliobia Governorate. Each soil was air dried, crushed to pass through a 2mm-sieve. The analysis of soils were reported in (Table 1), soil texture was determined by the hydrometer method. The pH was determined by a glass electrode (soil–water ratio, 1:2.5). Total soluble salts were measured in soil paste extracts using EC-meter. Total carbonates were estimated by gas metrically using Collin's calcimeter and calculated as equivalent  $\text{CaCO}_3$  (Nelson, 1982). Total N was determined by the Kjeldahl method of digestion and distillation (Bremner and Mulvaney, 1982). Organic carbon by the method of Piper (1950). The available P was measured by ammonium molybdate (Olsen and Dean (1965).

**Table 1:** Physical and chemical properties of the investigated soil samples.

Location	pH (1:2.5)	EC (1:5) $\text{dSm}^{-1}$	$\text{CaCO}_3$ %	O.M. %	Texture	Total N (%)	Available P (ppm)	Available K (ppm)
El-Nubaria	7.5	0.27	1.61	0.9	Sandy	0.04	7.3	38.2
El-Kanater	7.9	0.37	3.2	1.6	Clayey	0.06	15.6	250

### Collection of added amendments and materials

The different amendments used in this study were oxalic acid , citric acid , sulfur, farmyard manure , ammonium thiosulfate and phosphate solublizing bacteria. The organic acids (OA), (CA) were used at unify dose 60 mmol/ L, (FYM) applied at 3g per 200g soil, sulfur 99% ( $\text{S}^0$ ) at 0.06g per 200g soil, (ATS) 34% S at 8ml per 200g soil, single super phosphate (SSP) 1.6g per 200gm soil and phosphate solublizing bacteria (PSB) (mix of *Bukholderia cepacia* and *Acintobactersp*) 0.5ml per 1g PR mixed with 200gm soil in 500ml plastic pots. The air-dried sample of phosphate rock was finely grinded to pass a 100-mesh sieve (150 $\mu\text{m}$ ). The basic properties of phosphate rock are shown in Table (2), total P of the sample was determined by the digestion solution ( $\text{H}_2\text{SO}_4/\text{H}_2\text{O}_2$ ) as described by Murphy and Riely (1962). Ammonium thiosulfate (12%  $\text{NH}_4\text{-N}$  and 34% S), SSP (15.5%  $\text{P}_2\text{O}_5$ ) and  $\text{S}^0$  were purchased from the local market, while FYM was collected from local farms. A composite sample of well-dried FYM was taken, crushed into smaller particles by hand pressing, homogenized and passed through a 1mm sieve before use. The bio-power inoculants of PSB were provided by the Microbiology Department National Research Center, Ciza, Egypt. Chemical properties of farmyard manure (FYM) are shown in Table (3).

**Table 2:** The origin and composition of the phosphate rock sample

Location	Resource Name	Total $\text{P}_2\text{O}_5$ (%)	Available P (ppm)	pH (1:5)	EC (1:5)	$\text{CaCO}_3$ %	Major minerals
Western Desert	Abu Tartor	24	0.62	7.0	2.4	12.2	Fluroapatite- Apatite Calcite- Francolite- Quartz Low

**Table 3:** Some chemical properties of farmyard manure.

O.C %	O.M. %	C/N ratio	pH (1:10)	EC $\text{dSm}^{-1}$ (1:10)	Total nutrients (%)		
					P	K	N
19.5	33.2	25.2	7.2	5.81	0.35	1.2	1.60

### Experimental procedures

The effect of amendments on the release of inorganic P from phosphate rock; soils was carried out as follows. A 200g of each soil sample (2-mm mesh) was taken and transferred into 200mL capacity plastic pots (15 cm in diameter). There were 9 treatments including the soil without amendment (control), PR, SSP, PR with OA, PR with CA, PR with  $\text{S}^0$ , PR with FYM, PR with ATS and PR with PSB; five incubation times (14, 28,42 ,56 and 70 days) with three replicates. All the plastic pots were kept in an incubator at 25C<sup>0</sup> and arranged according to a completely randomized

design. The moisture content of soil sample was adjusted to field capacity by adding tap water and checked every 2 days by weighing the pots.

### Soil extraction and analysis

The extractable P from soil by  $\text{NaHCO}_3$  according to the method described by Olsen *et al.* (1973) was used for all treatment samples. The soil-available P was measured by ammonium molybdate (Olsen and Dean (1965)) using a spectrophotometer after each incubation times (14, 28, 42, 56 and 70 days). Also, the changes in the soil pH values for all treatments were determined using pH meter with glass electrode (soil–water ratio, 1:2.5) as described by Jackson (1973).

### Statistical analysis

One way ANOVA test and Least Significant Difference Test (LSD) were performed on the incubation experiment data set. The differences between treatment means were tested at 5% for all the sets of data analyzed.

## Results and Discussion

### Effect of different amendments on the release of P from PRs added to soils

The extractable P released from PR or PR mixed with organic acids, sulfur, ATS, FYM and PSB in clayey and sandy soils in relation to incubation time is shown in Table 4, 5 and Figs. 1(A,B) and 2(A,B). The extractable P from phosphate rock was increased by increasing incubation time. The rate of increment average over incubation periods reached 27.0, 33.0, 43.0, 54.7 and 59.9 ppm in clayey soil and 22.6, 32.2, 43.3, 55.1 and 55.6 ppm in sandy soil after 14, 28, 42, 56 and 70 days, respectively (Table 4, 5). Such results were confirmed by Agbenin and Tissen (1995). They found that P adsorption occurs rapidly in the first time and followed by a slow adsorption processes, reaching the soil to equilibrium after 50 days. In general, that the highest extractable P of incubation were found for both the clayey and sandy soils treated with ATS.

The behavior of rock phosphates in the two soils indicated slight solubilization of extractable P as compared to the control. The extractable P significantly ( $p < 0.05$ ) varied at all incubation intervals to applied treatments (Figs. 1A and 2A). The application of PR with organic acids, sulfur, ATS, FYM or PSB significantly enhanced extractable P over control showing contribution toward P build up in two soils with time (Figs. 1B and 2B). The extractable P from the phosphate rock with FYM treatment increased in both soils compared to the control and the percentage increase represented about ½ from the percentage increase of exactable P from the superphosphate. PR with FYM increased extractable P in clayey and sandy soil by 1.75 and 4.1 times that of the treatment PR alone, respectively. The extractable P progressively increased with increase in incubation time and at 70 days the extractable P attained a range of 17.2 to 49.7 ppm in clayey soil and 14.3 to 60.8 ppm in sandy soil (Table 4 and 5). This increase in P with passage of time showed the release of P from the source like PR, desorption of adsorbed P on soil colloids, dissolution of precipitated P with changing in soil properties associated with continuous watering and incubation and mineralization of immobilized or organic P with time. The positive influence of organic matter on PR dissolution has long been recognized (Dean, 1963).

**Table 4:** Effect of different treatments on phosphorus availability ( $\text{mg kg}^{-1}$ ) in clayey soil

Treatments	14 d	28 d	42 d	56 d	70 d	Mean
Control	15.9	16.1	15.8	16.3	16.0	16.0
SSP	63.1	76.5	67.2	66.0	57.6	66.1
PR	16.3	18.8	19.1	19.7	19.2	18.6
PR + Oxalic	32.5	45.8	67.3	88.4	91.2	65.0
PR + Citric	20.0	26.2	30.3	40.0	39.3	31.2
PR + Sulfur	17.3	20.1	23.3	36.0	47.3	28.8
PR+ ATS	43.8	55.6	95.7	134.5	140.9	94.1
PR+ FYM	17.2	19.5	34.7	42.3	49.7	32.7
PR+ PSB	16.5	18.1	33.2	49.3	51.9	33.8
Mean	27.0	33.0	43.0	54.7	59.9	
LSD 5%	Treatments (T) = 5.45		Periods (P) = 3.75		T × P = 10.70	

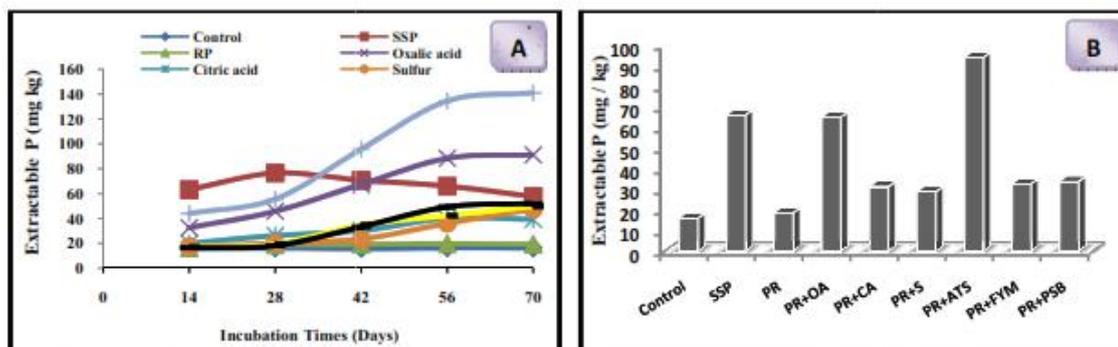


Fig. 1: Effect of incubation time (A) and different treatments (B) on phosphorus availability ( $\text{mg kg}^{-1}$ ) in clayey soil

Table 5: Effect of different treatments on phosphorus availability ( $\text{mg kg}^{-1}$ ) in sandy soil

Treatments	14 d	28 d	42 d	56 d	70 d	Mean
Control	7.4	7.2	7.6	7.7	7.1	7.4
SSP	81.9	85.9	89.2	87.7	85.2	86.0
PR	8.0	8.2	9.2	9.9	10.1	9.1
PR+ Oxalic	21.3	35.1	48.5	67.9	62.7	47.1
PR+ Citric	13.2	23.2	35.2	55.4	48.8	35.2
PR+ Sulfur	10.4	24.9	36.6	53.3	56.2	36.3
PR+ ATS	31.1	53.2	84.9	101.3	99.8	74.1
PR+ FYM	14.3	22.9	35.1	52.6	60.8	37.1
PR+ PSB	15.6	29.8	43.8	60.5	69.9	43.9
Mean	22.6	32.2	43.3	55.1	55.6	
LSD 5 %	Treatments (T) = 3.68		Periods (P) = 2.43		T × P = 7.32	

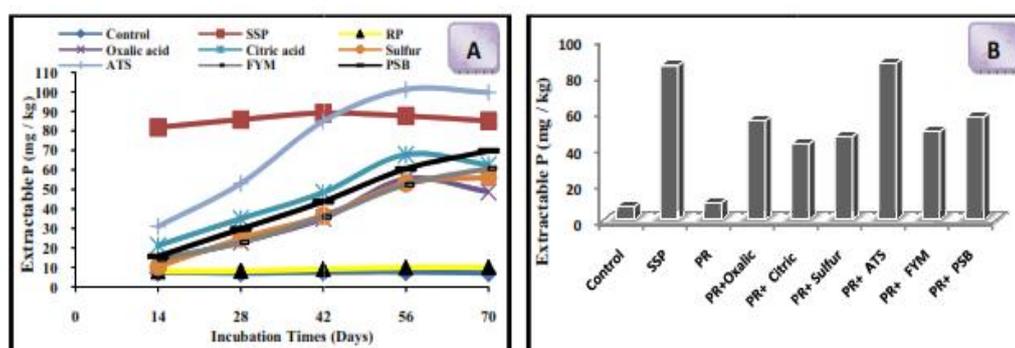


Fig. 2: Effect of incubation times (A) and different treatments (B) on phosphorus availability ( $\text{mg kg}^{-1}$ ) in sandy soil.

This seems to arise from the (i) high cation exchange capacity of organic matter and (ii) organic acids produced as a result of microbial and chemical transformations of organic debris. The cation exchange capacity of mineral soils, depending on their clay content, may range from a few to 50 or 60  $\text{cmol kg}^{-1}$ , whereas that of organic matter may exceed 200  $\text{cmol kg}^{-1}$  (Helling *et al.*, 1964). Thus organic matter can enhance PR dissolution by enhancing the Ca buffer capacity of soils. Amberger, 1991 found that composting phosphate rock materials with straw or other organic waste materials resulted in phosphate mobilization. They further indicated that the driving force for phosphate rock solubilization is the pH decrease and the chelating effect of simple organic acids as citric and malic and complex acids as humic and fulvic acids on  $\text{Ca}^{2+}$  ions from insoluble phosphate forms. Sahin *et al.* (2014) reported that humic substances in interaction with P in the soil could decrease the P fixation and increase the P uptake of plants. Humic acid served as a buffer at a broad pH interval and several macro and microelements could be taken by the plants since the soil was neutralized.

Odongo *et al.* (2007) reported that phosphorus from phosphate rock could be mobilized by using composting manure. They also reported that wheat straw attributed to phosphorus release from phosphate rock as a result of mineralized organic compounds produced during composting chelated with Ca from phosphate rock to increase available P. This is in agreement with Kpombrekou and Tabatabai (2003) who found that phosphorus release was negatively correlated with the equilibrium pH and positively correlated with Ca released from the phosphate rock. Evolved CO<sub>2</sub> during the process of organic manures decomposition results in formation of carbonic acid, which dissolves phosphate rock (Biswas and Narayanasamy, 2006).

Inoculation of phosphate rock with PSB showed a significant effect at  $p < 0.05$  on mean of extractable P (Tables 4, 5 and Fig.1B and 2B). The mean of extractable P from the phosphate rock with PSB treatment increased in both soils compared to the control and the percentage increase represented about ½ from the percentage increase of extractable P from the superphosphate. PR with PSB increased extractable P in clay and sandy soil by 1.82 and 4.82 times that of the treatment PR alone, respectively. After 14 days of incubation, extractable P increased remarkably with PSB in clayey and sandy soils. The highest concentration of extractable P was obtained at 70 days of incubation. After 70 days of incubation, the concentration of extractable P was 3.8 and 9.4 times higher than control treatment in clayey and sandy soil, respectively.

Jha *et al.* (2013) isolated 10 PSB strains and tested them for mineral phosphate solubilization activity of PR and stated that all these strains could solubilize only 0.02–2.6% of the total PR P applied. In addition, *Aspergillus niger* (a fungus), used in the industrial production of citric acid and has been recognized as one of the most effective organisms for PR solubilization (Abd-Alla and Omar, 2001). These results suggest that (i) PSB increased P solubilization from phosphate rock compared to the control. Reddy *et al.* (2002) compared the efficiency of three isolates on the solubilization of PR and reported that all the isolates increased P release efficiency by solubilizing the tested PRs. Similar effects of bio- and organic fertilizers on PR availability and P fertilizer efficiency had also been reported in soils incubated for different incubation times (Alzoubi and Gaibore, 2012). The mechanisms involved in the potential of PSB to solubilize P complexes or insoluble phosphates are well known and have been attributed to the processes of acidification, chelation, exchange reactions and the production of organic acids (Chen *et al.*, 2006).

Table 4 and 5 show the results of sulfur application with phosphate rock on extractable P. When phosphate rock amended with sulfur and extractable P increased significantly. The most significant effect of sulfur on extractable P was observed until the 70<sup>th</sup> days of incubation. After 70 days of incubation, the concentration of extractable P was 3.0 and 7.6 times higher than control treatment in clayey and sandy soils, respectively.

PR with S<sup>0</sup> increased extractable P in clayey and sandy soils by 1.45 and 4.0 times that of the treatment PR alone, respectively. Evans *et al.* (2006) reported that co-treatment of phosphate rock with sulfur was necessary to increase phosphorus which was generally associated with increased plant dry matter. They also reported that the amount of required sulfur was less in acidic soils. Ghani *et al.* (1994) reported that treatments without elemental sulfur or *Thiobacillus* had little effect on the dissolution of phosphate rock. The combination of elemental sulfur and *Thiobacillus* increased extractable phosphorus to nine times greater than that of untreated phosphate rock.

Data in Figure.1B and 2B show that the mean of extractable P from phosphate rock was significantly increased in clayey and sandy soils with organic acids (oxalic or citric acid). This may be due to the favorable effect of such acids in increasing the solubility of P from PR. Treatment PR with oxalic or citric acid increased extractable P in clayey soil by 3.49 and 1.67 and sandy soil by 5.2 and 3.87 times that of the treatment PR alone, respectively (Table 4 and 5). In this respect (Marschner, 1990) pointed out that, plant excrete organic acids such as citric, oxalic and tartaric acid in root zone to increase phosphorus solubility and availability in rhizosphere. The oxalic acid was superior to citric acid. PR with oxalic acid that produced the highest values of extractable P (mg kg<sup>-1</sup>) not only at 70 days but the consistently and significantly higher values P with such combination at all incubation interval suggested that this could be the optimum combination. These results are corroborated by the findings of Cajuste *et al.* (1996) who reported that increasing the loading of organic anions (oxalate and citrate) decreased the maximum P adsorption capacity of the soils and increased the P release. It seemed that in the current study, the effect of oxalic acid on P mobilization was greater than the effects produced by citric acid. The stability constants of Ca<sup>2+</sup> with citrate and oxalate have been

found to be 4.85 and 3.19, respectively. It may help to explain the difference in the effect of organic acids on P mobilization. On the same lines, Khademi *et al.* (2009) reported that oxalate is slightly more effective than citrate in mobilizing P that could be attributed to different acid strength and the number of carboxyl groups. Among these organic acids, oxalic acid (H<sub>2</sub>C<sub>2</sub>O<sub>4</sub>) is a relatively strong acid. Oxalate has a tendency to be precipitated readily in the presence of Ca<sup>2+</sup> and reduce the content of active carbonates in calcareous soils. Gang *et al.* (2012) further concluded that the pattern of P mobilization by the addition of organic acids was highly soil type dependent and was controlled by intrinsic P status such as amounts and distribution of P fraction in the soil. Citric acid (C<sub>6</sub>H<sub>8</sub>O<sub>7</sub>) is weak organic acids with three carboxyl groups.

Table 4 and 5 show the results of ATS application with phosphate rock on extractable P. When phosphate rock amended with ATS, extractable P in clayey and sandy soils increased by 5.06 and 8.14 times that of the treatment PR alone, respectively, extractable P increased significantly. The most significant effect of ATS on extractable P was observed until the 70 days of incubation. After 70 days of incubation, the concentration of extractable P was 8.9 and 13.7 times higher than control treatment in clay and sandy soil, respectively. ATS [(NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>] is used as a source of nitrogen (N) for crop production by farmers in the sub-Saharan region of Africa. The fertilizer has an acidifying effect on soils due to the nitrification process in warm soils, where microbes will rapidly begin to convert ammonium to nitrate in the process of nitrification [2NH<sub>4</sub><sup>++</sup> 3O<sub>2</sub> → 2NO<sub>2</sub><sup>-</sup> + 2H<sub>2</sub>O + 4H<sup>+</sup>]. During this microbial reaction, H<sup>+</sup> is released, which ultimately decreases soil pH after repeated use (IPNI, 2016).

### Effect of different amendments on soil pH

The change of acidity in the soil depends on various soil and plant factors, such as soil buffering capacity, soil moisture level and aeration CO<sub>2</sub> production by plant roots and microorganisms, microbial acid production, root exudation of carboxylates, plant genotype and nutritional status of the plant (Marschner 1995).

The effect of different P amendments with phosphate rock on changes in soil pH over 70 days incubation is presented in Table 6,7 and Figs (3(A, B) and 4(A, B)). Soil with PR or soil without amendment (control) showed the maximum pH. The pH of all the added amendments in clayey and sandy soils tended to decline with time, except control, PR or SSP. The pH obtained from PR with oxalic, citric, sulfur, ATS, FYM and PSB significantly decreased at the end (70 days).

The soil pH decrease relative to day 14 was 3.4, 2.5, 5, 6.7, 3.8 and 3.8 percent in clayey soil and 2.8, 4.1, 8.1, 8.5, 5.4 and 5.5 percent in sandy soil, respectively. Averaged across different amendments, the data presented in Table 6 and 7 indicated that the mixture of oxalic, citric, sulfur, ATS, FYM or PSB with PR decreased pH considerably in clayey soil from 7.9 to 7.4, 7.7, 7.7, 7.2, 7.7 and 7.6, respectively.

A mixture of oxalic, citric, sulfur, ATS, FYM and PSB with PR reduced pH considerably from 7.5 to 7.0, 7.1, 7.1, 6.7, 7.2 and 7.1, respectively, in sandy soil. Effect of time on soil pH shows that at 14, 28, 42 and 56 and 70 days, the pH of soil was decreased as compared to the pH seen at the beginning of the experiment. Among these five times, the highest decrease of soil pH was observed at end of experiment (70 days) in both clayey and sandy soil.

**Table 6:** Effect of different treatments on pH clayey soil

Treatments	14 d	28 d	42 d	56 d	70 d	Mean
Control	7.9	7.9	7.9	7.9	7.9	7.9
SSP	7.2	7.1	7.1	7.2	7.3	7.2
PR	7.9	7.9	7.9	7.9	7.9	7.9
PR+ Oxalic	7.6	7.5	7.4	7.3	7.3	7.4
PR+ Citric	7.8	7.8	7.7	7.6	7.6	7.7
PR+ Sulfur	7.9	7.8	7.7	7.6	7.5	7.7
PR+ ATS	7.5	7.4	7.2	7.0	7.0	7.2
PR+ FYM	7.8	7.7	7.7	7.6	7.5	7.7
PR+ PSB	7.8	7.7	7.6	7.5	7.5	7.6
Mean	7.7	7.6	7.6	7.5	7.5	
LSD 5 %	Treatments (T) = 0.085		Periods (P) = 0.063		T × P = 0.23	

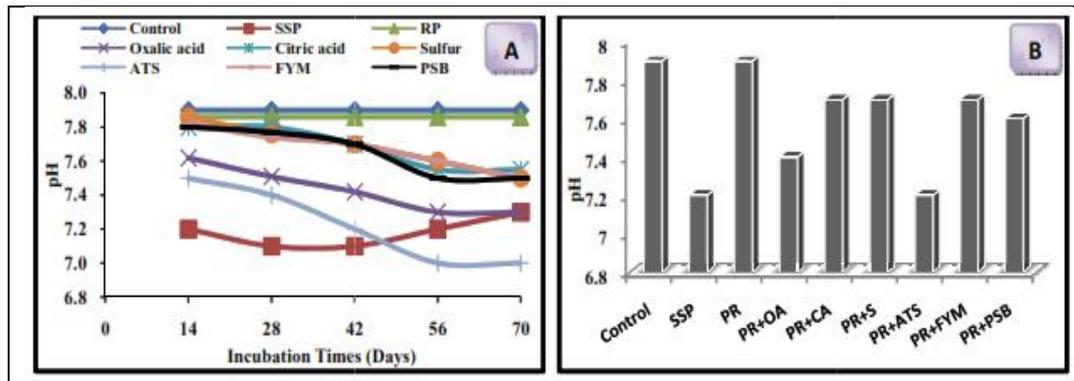


Fig. 3: Effect of incubation time (A) and different treatments (B) on pH in clayey soil

Table 7: Effect of different treatments on pH in sandy soil

Treatments	14 d	28 d	42 d	56 d	70 d	Mean
Control	7.5	7.5	7.5	7.5	7.5	7.5
SSP	6.7	6.6	6.6	6.6	6.6	6.6
PR	7.4	7.4	7.4	7.4	7.4	7.4
PR+ Oxalic	7.2	7.1	7.0	6.8	6.9	7.0
PR+ Citric	7.3	7.2	7.1	6.9	7.0	7.1
PR+ Sulfur	7.4	7.3	7.0	6.8	6.8	7.1
PR+ ATS	7.1	6.9	6.7	6.5	6.5	6.7
PR+ FYM	7.4	7.3	7.2	7.1	7.0	7.2
PR+ PSB	7.3	7.2	7.1	7.0	6.9	7.1
Mean	7.3	7.2	7.1	7.0	7.0	
LSD 5 %	Treatments (T) = 0.071		Periods (P) = 0.054		T × P = 0.18	

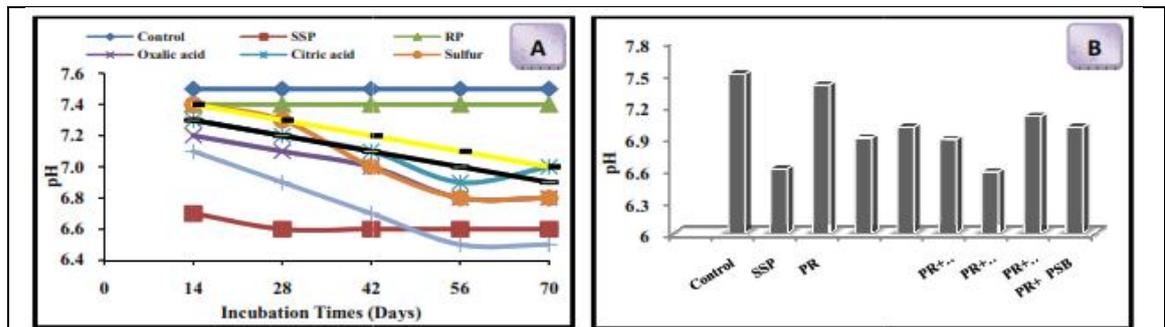


Fig. 4: Effect of incubation time (A) and different treatments (B) on pH in sandy soil

### Conclusion

From the above mentioned results it can be conclude that the amount of extractable P from phosphate rock could be increased by increasing incubation time of rock in both clayey and sandy soils. The applications of organic acids, farmyard manure, ammonium thiosulfate, phosphate solubilizing bacteria or/and elemental sulfur could be successfully used for increasing P-extractable from rock phosphate. Comparatively more P was solubilized from PR through mixing with ammonium thiosulfate and oxalic acid.

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