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Bio-Organo-Phos: A Sustainable Approach for Managing Phosphorus Deficiency in Agricultural Soils

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Abstract

Sustainable agriculture is essential for a positive relationship between supply and demand of food for the growing world population. This relationship was found to be affected by many environmental factors, including biotic and abiotic. From the point of view of crop nutrition, sustainability in the supply of essential nutrients particularly phosphorus is vital. Due to the energy crisis, the fluctuation in the prices of chemical fertilizers, environmental concerns, and cessation in the supply of high quality rock phosphate (RP) are hindering the use of chemical phosphatic fertilizers for sustainable crop production. Therefore, there is great need for a sustainable solution to this problem. It could be solved by employing a strategy to use native low quality RP. It is only possible by composting of organic material in the presence of RP and phosphate solubilizing microorganisms. During composting, most of organic P is mineralized. Due to release of organic acids, P availability to crop plants increases. In this chapter, the importance of economical and sustainable sources of P and comparative efficacy of the use of organic fertilizer containing RP for legumes is critically reviewed.

Keywords: Phosphorus, rock phosphate, sustainable crop nutrition, PSMs, legumes

1. Introduction

Agriculture plays a crucial role in the economic development and poverty alleviation in developing countries. Also, agricultural sustainability is vital for a sustainable agriculture. There must be a positive link between its supply and demand. Unfortunately, this link has been disturbed by many factors from which the deficiency of the nutrients especially that of phosphorus (P) has been a major one [1]. Moreover, its application to the soil in developing countries has been hindered due to alarming increase in its price.

Adequate amount of phosphorus (P) is critical for normal plant growth and development as it is a vital component of plant energy system and genetic material in the form of adenosine triphosphate-ATP and deoxyribonucleic acid-DNA, respectively [2]. Moreover, it also is involved in many plant processes like photosynthesis, carbon metabolism, membrane formation, energy generation, nucleic acid synthesis, glycolysis, respiration, activation and inactivation of enzymes, and nitrogen fixation [3]. Root architecture, seed development, and crop maturity is also affected by P deficient conditions [4].

Leguminous crops engaged in symbiotic nitrogen (N_2) fixation generally require high amount of P due to high energy requirement which is mainly contributed through ATP [5]. Under P-sufficient conditions, nodules have a higher P concentration (up to 1.5% of the total plant P) as compared to that of the shoots and roots [6]. It is also needed for signal transduction, membrane biosynthesis, and nodule development and function [7]. Many studies concluded that P is the most limiting nutrient in many soil types for the production of crops especially the nitrogen fixing leguminous crops [8–10]. The direct and positive role of P in nodulation of red clover [11], peas [12], white clover [13], *Medicago truncatula* L. [14], and soybeans [15] has been reported. Under P deficient conditions, reduced root growth and photosynthetic carbohydrate supply to the nodules occurs [6, 16], which results in reduced nodule growth and function and ultimately reduced symbiotic N_2 fixation [17]. It has been found that N and P control the nodule growth and modulate the symbiotic processes of the legume and *Rhizobium* [11]. Phosphorus has its basic function in plant energy system and reduction of N_2 to NH_3 requires 16 ATP molecules. The availability of soil P is optimum at pH ranging between 6.5 and 7.0 for plant absorption, hence creating a soil environment that is more favorable for N_2 -fixers like *Rhizobium*.

Rock phosphate (RP) is an economical source of P to the crop plants and 200–300 billion tons of RP are available throughout the world. For example in Pakistan, 20–30 million tons of different grade phosphate rock has been documented. The major issue with RP is having a low available P. Research work is needed to find out a way to enable farmers to utilize this RP as P-fertilizers to meet fertilizer demand and cope with the prices. A huge share of foreign exchange is utilized in importing phosphatic fertilizers which is not feasible for countries, especially the developing ones. Another constraint with chemical phosphatic fertilizers is that these are prepared from high quality RP which may be depleted by the year 2050 [7]. These problems have increased the necessity to find other measures and approaches so as to exploit indigenous RP resources in bioavailable form without compromising on yield.

From direct application of RP as a P source to the soils, it has been clearly found that this approach is feasible for the acidic soils having low pH and direct application of RP to the alkaline soils [18]. Another approach which could help solubilize the fixed P in RP could be to use the bio-inoculants, which through the release of organic acids (acetate, lactate, oxalate, tartarate, succinate, citrate, gluconate, ketogluconate, glycolate, etc.) reduce the pH of the micro-environment prevailing around these microbes [1, 19–21]. The plant growth promoting rhizobacteria (PGPR) include all the bacteria found in the rhizosphere which directly or indirectly enhance plant growth. Phosphate solubilizing microorganisms (PSMs) are the PGPR and can be utilized for enhancing the availability of P which has a direct effect on nodulation.

There are many reports about the use of RP along with PSMs as an alternative cheaper source of P [22, 23].

Scientists around the world have documented positive effects of organic fertilizers in improving the physical properties of soils, thereby increasing the availability of nutrients especially the least mobile ones like P [24–29]. The combined application of RP-EC with PSMs could be helpful in improving the nodulation, growth, and yield of crop plants. There are few reports about their combined application. Shahzad and his co-workers [30] found that combined application of rhizobacteria and P-enriched compost resulted in an increased growth and yield parameters compared to uninoculated control without compost. So PGPB along with SSP-enriched compost was found highly effective in improving growth, yield, and nodulation of chickpea as compared to their application alone. Saleem *et al.* [29] conducted a series of field and pot experiments to check the effectiveness of RP, compost, and PSMs in increasing the growth and yield of wheat. RP-enriched compost was used based on 25 and 50% P of the crop requirement and the balance amount of P was applied through chemical phosphatic fertilizers. From the results, it was found that application of 50% P from RP-EC with PSMs and 50% from the chemical fertilizers maximally increased most of the growth and yield parameters compared to the controlled (100% P from chemical Fertilizers).

Recently, we have conducted a series of pot and field experiments in comparing P nutrition of legumes through bio-organo-phos (a mixture of PSMs, RP, and compost) and found a significant improvement in growth, nodulation, and yield compared to the recommended chemical fertilizers [31]. This chapter will give an overall research progress in finding out the economical and sustainable source of P for the crops especially for the legumes and their future perspectives. It also addresses the pros and cons with their future perspectives and research needs.

2. Importance of phosphorus for symbiotic nitrogen fixation in legumes

Phosphorus is the most limiting nutrient for the production of crops especially the nitrogen fixing leguminous crops for adequate growth and nodulation. There are many reports about its direct and positive effect on nodulation of red clover, peas, white clover, *Lupinus L.*, soybeans and many more [11–15]. Indirect effect on plant growth has also been reported, thereby increasing the nodulation and stimulating the nitrogenase activity [32]. Reduced root growth and photosynthetic carbohydrate supply to the nodules occurs under P deficient conditions [6, 16] which results in reduced nodule growth and function and ultimately reduced symbiotic N₂ fixation [17]. It has been found that N and P control the nodule growth and modulate the symbiotic processes of the legume and Rhizobium [11, 33]. Various morphological and physiological changes occur under P deficient conditions including increase in the root/shoot ratio, changes in root architecture [4, 34], development of root hairs [35], induction of the high-affinity phosphate transporter [36], and synthesis and exudation of the organic acids, phosphatases and ribonucleases (RNases) [37–39]. It has been reported that crop yield is seriously affected under P limited conditions especially during early stage of growth [40].

Phosphorus (P), being the second most important macronutrient after nitrogen, has a critical role in biological nitrogen fixation (BNF). It has been a renowned fact that legumes require more phosphorus (P) compared to non-leguminous crops as they perform the process of BNF through nodulation which is a characteristic property of legumes. Nodulation occurs in almost all the legumes. However, physiology and efficiency of nodules to conduct the process of nitrogen fixation is species specific. So for the process of BNF, P serves as an ultimate source of energy in the form of ATP [5, 41, 42]. It is also needed for signal transduction, membrane biosynthesis, and nodule development and function [8]. Moreover, nodules under P-sufficient conditions have a higher P concentration (up to 1.5% of the total plant P) as compared to shoots and roots [6].

There are many reports about the influence of P on nodulation; ultimately the amount of N fixed by the plants e.g. nitrogen contents of the legume, *Crotalaria micans*, were increased about 4 folds due to increased nitrogen fixation with the application of P at 90 kg ha⁻¹ [43]. Similarly Israel [15], studied the effect of P on accumulation of nitrogen in soybean (*Glycine max* L.). It was found that the concentration of nitrogen was increased with increasing the supply of P that was suggested to be due to increase in the symbiotic nitrogen fixation as P serves as an energy source. Plant dry matter, nodule number, and mass was also increased with increasing P supply. Enzyme activity of nitrogenase was also enhanced with increasing P. Earlier legumes are well renowned as P exhaustive crop plants due to the formation of nodules for symbiotic nitrogen fixation. In another experiment on different legumes (soybean, clover), similar effect of P on growth and nodulation parameters was recorded [44].

Another soil and sand culture experiment to study the interaction of N and P and their effect on growth, nodulation, nitrogen fixation, activity of nitrate reductase, and on the accumulation of nitrogenous compounds (ureides, amino acids, nitrate) in the xylem sap of common bean was conducted [3]. Both the soil and sand culture experiment showed that with increasing levels of N, nodulation parameters such as nodule number and mass, nitrogenase activity, and xylem ureides were decreased, while the concentration of asparagine in the xylem sap increases. Symbiotic nitrogen fixation was only increased at low N concentration with increasing P application. Similarly it was also found that the effect of N on the inhibition of nodulation including nodule number and biomass was systemic, while high dose of P had a systemic stimulatory effect on nodulation parameters as mentioned above. The systemic effect was confirmed by its direct effect on nodulation and not on the plant growth overall. There is still lack of information on whether there is effect of N and P on both the nodulation and nitrogen fixation or not and needs to be explored in future studies [32].

Similarly, another Leonard jar experiment was conducted to study the impact of P (0–2 mM P) on growth, symbiotic nitrogen fixation, N and C metabolism, as well as on the concentration of ATP, N and P contents of common bean (*Phaseolus vulgaris*). With the application of medium to high-P, not only the nodulation (nodule biomass: 4-fold) and growth parameters but also the P contents of the harvested plants were increased at the onset of the flowering. In the case of total soluble sugar and amino acid contents of leaf, root, and nodules of the plant, these were decreased with increasing the level of P application. Moreover, an increase of 20-fold in nodule-ARA and 70-fold in ARA per plant was observed with the application of 1.5 mM P [45]. Another

split root experiment found that P has a specific effect on the nodulation not generally on the overall growth of the plant. More P was required in the earlier stage of nodule initiation and growth of the plant. It was also found that P could suppress the effect of N on the inhibition of symbiotic nitrogen fixation [32]. Many other researchers have reported the effect of P on nodulation, growth, and nitrogen fixation in many crops like clover, soybean, red clover, etc. [11, 15, 44, 46].

3. Phosphorus problems in the soils

Plants depend almost exclusively on the availability of nutrients from the soil as these are fixed in the soil. As far as P is concerned, its availability in the form of PO_4^{3-} , HPO_4^{2-} , and H_2PO_4^- depends on the soil pH and is a significant determinant of plant growth [41]. There are two forms of P present in the soil: the organic (20–85%), which is the large proportion, and the inorganic. Phosphates (PO_4^{3-} , HPO_4^{2-} , and H_2PO_4^-) are the most available forms of inorganic P in the soil. While the organic portion of P, phytin and its derivatives constitute about 50%, lecithin and glycerol-phosphate are present in minor fraction, and these organic compounds could serve as a good source of P if mineralized [47].

Normally, the amount of total P present in the soil is about 0.05% (w/w) but the available fraction of P to the plants is very small (Reference). To overcome this problem and to maximize the production of crops per unit area, farmers have to apply costly inorganic synthetic phosphatic fertilizers, but their availability to the crop plants hardly reaches 20% [48]. This has led to the addition of 2–4 times more than the required amount of phosphatic fertilizers and has led to the maximization of cost to benefit ratio of growing crop plants per unit area [49]. Another issue with the phosphatic fertilizers, most available form of P, is that their cost has gone sky high and unaffordable for the poor farmers of the world especially of the developing countries like Pakistan. Moreover, it becomes an environmentally ill practice if we consider the production of chemical phosphatic fertilizers, which include the use of sulfuric acid [50]. Moreover, environmental problems such as eutrophication has also emerged which has led to the destruction of the habitat of the aquatic life and has caused environmental degradation. In conclusion, these problems have pessimistically posed a problem not only on the environment but also on the economics of growing crops. There are two reasons, which are mostly quoted for acidic and basic/alkaline soils depending on the soil pH. In acidic soils such as oxisols, inceptisols, and utisols, the added P quickly reacts with Fe and Al oxides as these are the dominant cation oxides in these types of soils and make it unavailable to the plants. In case of alkaline soils, Ca and Mg salts are abundantly present, so these cations react with phosphatic compounds thus making them unavailable to the plants [41].

4. Expected depletion of RP

The high pH of the soils from the Savannah zones of Nigeria resulted in increased adsorption of P when varying quantities of phosphate were added in these soils. The suggested respon-

sible factor causing this fixation was the high activity of hydroxy-aluminum at high pH which has strong attraction with phosphate compared to hydroxyl. This attraction was found enough to displace hydroxyl from the hydroxy-aluminum-phosphate attraction. Due to this, increase in phosphate buffer capacity and the amount of phosphate required to attain the desired level of P in the equilibrium solution was noted [51]. In order to study the mechanisms behind the fixation of P in the soils, another study was conducted which showed that P adsorption by the amphoteric soil surface decrease with increasing pH from 4.0–7.0. But in soils high in exchangeable Al, increasing pH results in the formation of highly reactive adsorbing surfaces for P as Al-ions precipitate and insoluble polyhydroxy-Al cation species. So if acidic soils are reacted with lime without intervening air drying, this will result in the adsorption of more P in the soils. Alternatively, it was found that the application of lime to the acidic soils after intervening air-drying results in decreasing the P adsorption in the soil as clear through isotherm studies [52].

As mentioned earlier, precipitation, and adsorption of P with the soil colloids are the main responsible mechanisms/reactions for the removal of P from the soil solution. The former is induced by the presence of Ca^{2+} ion in the soil solution while latter depends on the chemical properties of the soil colloids. There is high probability of having the precipitates of calcium phosphate in the soils rich in exchangeable cations. It has also been found that calcareous soils were poor in plant available P compared to the limed acid soils [53].

If we look at the manufacturing process of phosphatic fertilizers, we would found that the major factor responsible for their high prices is the use of very high energy in their manufacturing process. Coming to the initial step of the process is the raw material that is used for the manufacture of most of the phosphatic fertilizers, the phosphate rock or RP that is a naturally occurring mineral source of P. One strategy could be the use of this raw material, the cheaper source of P, directly in the field and getting the benefit of phosphatic fertilizers. To cope with this alarming situation, there is a need to find alternative cheaper sources of P, so that we could have a sustainable agriculture to feed out the ever growing population of the world.

5. Rock phosphate (an indigenous and economical source of P)

Phosphate rock or the RP is a non-renewable alternative natural source of P. It serves as raw material for the so-called chemical phosphatic fertilizers. It has been found on every continent of the world, however, quality may differ depending on the percentage of P present in it. Distribution of RP on the earth is given in Figure 1 [54].

In literature, there have been many reports showing effectiveness of directly applied RP being less expensive compared to the chemical phosphatic fertilizers like SSP or TSP. It has also been found to be effective for the perennial crops [55]. It has been directly applied in the soil of Nigeria in place of very expensive phosphatic fertilizers [50, 56]. The demand for the direct application of RP is increasing day by day as it would help reduce pollution and the burden on manufacturing industry for the high demand of chemical phosphatic fertilizers, and this would ultimately serve as a cheaper source of P. It has been found that RP results in increasing

Figure 1. Rock phosphate (RP) reserves around the world.

the relative agronomic efficiencies of the crops grown on P deficient soils. It has sedimentary origin and its direct application might be feasible due to the presence of somewhat open, roughly consolidated aggregates of microcrystals with large surface area [57].

In a study, the direct application ortho rock phosphate (ORP) as a P source increased the crop yield and was comparatively found to be more effective than chemical phosphatic fertilizers [58]. Another greenhouse experiment was conducted to assess the relative effectiveness of directly applying Togo and Egypt RP sources and these sources were compared with TSP, using Lucerne as a test crop. From the results, it was found that the application levels of 60 and 43 kg P ha⁻¹ were the effective levels of Togo and Egypt RP, respectively. At these levels, the Egyptian and Togo RP were 92 and 64% as effective as that of TSP, respectively [59]. The direct application of RP to vertisols, oxisols, and ultisols, having pH less than 7, has reported to yield similar results as that caused by TSP. Moreover, the initial and residual effects of RP on volcanic ash soils like that of vertisols, oxisols, and ultisols are less compared to the TSP. It has also been found that soils with pH >7, like in andepts, should be taken with care when applying RP directly to the soils as these soils have greater tendency of P sorption [59–61].

It has been found that most of the phosphate rocks or RPs yield better results in the acidic soils like that of oxisols and ultisols compared to alkaline and neutral soils like andepts with high pH, high P sorption capacity, low cation exchange capacity (CEC), low rainfall, low organic matter, low microbial activity, etc. Moreover, RP has significant proportion of isomorphic substitution in the crystal lattice and variable proportion of impurities and accessory minerals. Thus, it has been found to be more beneficial if applied in acidic soil conditions as compared to the neutral and alkaline conditions [57].

One strategy for the direct application of RP on andepts soils might be the application of partially acidulated RP by using 20% H_2SO_4 . It has been reported to increase the plant response from applied RP from 0–16% to 59–77% relative agronomic efficiency (RAE) on Andepts [61]. However, the application of inorganic acid like 20% H_2SO_4 could have detrimental effects on the soil microbiota. Moreover, some other procedures like mixing with elemental sulfur, partial acidulation with an acid, thermal alteration, combination with chemical phosphatic fertilizers, preparation of RP enriched compost, and dry compaction with water-soluble chemical phosphatic fertilizers [62–64] have been reported to increase the efficiency of RP in increasing the availability of P for the crops. However, these procedures are labor intensive, costly, and inappropriate to be practiced at large scale. Due to these problems, there is a growing interest in manipulation certain biological procedure like the application of PSMs. Composting have been proposed, which are discussed in the next sections separately.

6. Integrated application of RP and PSMs

It is well renowned that RP has no substitute as a source of P however; minimum processing is required for its direct application to the soils especially the non-acidic soils. It has been found that RP could be an effective source of P after four years of its application in soils with pH 5.5–6.0 [65]. Moreover, RP is not economically feasible for the soils with high adsorption capacities, low CEC, high pH, low rainfall, low organic matter content, and low microbial activity [66]. For these reasons, various strategies like that of the application of PSMs have been proposed in order to increase the solubilization of P from RP [67].

PGPR are the bacteria present in the rhizosphere (the area near plant roots up to where the effect of roots and its exudates is found), which helps directly or indirectly to the growth and yield attributes of the plants. Many species of PGPR have been identified like PSMs, 1-aminocyclopropane-1-carboxylate deaminase (ACC-deaminase) producing bacteria depending on the mechanism of action employed [68]. As mentioned in the above section, phosphate solubilizing bacteria (PSBs) and/or plant growth-promoting rhizobacteria not only improves the physicochemical properties of the soils, but also help in the solubilization of RP, which leads to increased availability of P to the crop plants. Certain mechanisms employed by these PGPR and PSMs have been identified regarding the solubilization of RP. This includes: 1) production of certain organic acids formic, acetic, propionic, lactic, gluconic, fumaric, and succinic which results in lowering the pH of microclimate in the rhizosphere, 2) synthesis of chelating compounds which help in easy provision of nutrients to the crop plants and many more [19, 21]. Other mechanisms reported in literature include the production of phytohormones, siderophores, nutrient assimilation, protection of seeds from pathogens through antagonistic action, competition for the nutrients and space, induction of systemic resistance and emission of volatile organic compounds [69–76]. Moreover, the presence of microbiota in the soil is an indicator of good soil health. In this way, the application of these bio-inoculants not only helps in the solubilization of fixed P in RP but also reduce the amount of costly prepared chemical phosphatic fertilizers being applied to the soil, thereby providing a cheaper and sustainable source of P for the plants [77–78].

Previously, it has been found that under biotic and abiotic stress like that of nutrients; there is increased production of ethylene that is a well renowned stress hormone and causes senescence, abscission, and chlorosis in the environment where it is produced. So as we have phosphorus stress due to the low recovery efficiency of phosphatic fertilizers as mentioned earlier, we have to adopt certain strategy that can reduce the amount of ethylene in the rhizosphere of the plants. Studies aimed on finding the biosynthetic pathway of ethylene have found that ACC is the precursor of ethylene.

One strategy could be to use the microorganisms that can use its precursor the ACC as a nutrient source and reduce the amount of ethylene produced. Scientists have isolated the so-called PGPR with ACC-deaminase activity. These rhizobacteria contain an enzyme ACC-deaminase that can convert ACC, the precursor of ethylene, into ammonia and α -ketobutyrate, thereby reducing the ethylene stress and ultimately increase the plant growth especially through the proliferation of root growth with increased surface area to explore more soil volume [80]. Many studies have confirmed the efficacy of these PGPR in increasing the root growth of the crop plants, and hence improved yield through increased absorption of nutrients [71–72, 74–76]. In literature, there are many reports about the use of RP along with PSBs as an alternative cheap source of P for costly chemical fertilizers [22–23]. Zaidi and Khan [80] have suggested a synergistic interaction between PSBs and nitrogen fixers like *Azotobacter chroococcum* that helped in the better utilization of poorly soluble RP. Organic acids produced by different PSBs, are mainly responsible for this solubilization as reported earlier [20, 82]. The degree of P-solubilization of RP by the application of PSMs and their impact on nodulation, growth, and yield of mung bean was studied on an acidic and alkaline soil. The results clearly showed an increased nodulation, growth, and yield attributes of mung bean over control. Moreover, it was suggested that optimum results regarding nodulation, growth, and yield of mung bean could be because of PSMs applied along with an initial dose of chemical phosphatic fertilizer [83].

From the above discussion, it could be imperative to use both types of microorganisms with the ability to solubilize P from the RP, and decrease the level of ethylene through the enzyme ACC-deaminase. In soil microbiology, we usually use a term co-inoculation which involves the application of microbes with more than two traits. In plant sciences, it would be the application of microbes with phosphorus solubilizing activity and ACC-deaminase activity i.e. the application of PSBs and PGPR with P-solubilizing and ACC-deaminase activity.

This strategy has been opted under pot and field conditions and many success stories have shown its effectiveness. However, this strategy has been studied for increasing the availability of P from the so-called chemical phosphatic fertilizers, which are becoming a burden for the farmers to use them due to high cost as already mentioned in the previous section, and not from the RP. Similarly, there are certain problems associated with biofertilizers like shelf life and lack/limited knowledge about their mechanism of action. Poor handling and lack of availability in remote areas are also one of the problems due to poor extension work to disseminate their beneficial effects to the crops in remote areas. These issues could be solved by employing biotechnological approaches to produce certain genetically modified organisms

(GMOs) which could sustain the harsh environmental conditions and thereby improved shelf life.

7. Integrated application of RP and compost

As mentioned earlier, the direct application of RP has been useful only in soils with acidic pH due to poor solubility in alkaline and neutral soils [85]. However, a limited number of climatic and soil situation are available in which direct application of RP would sufficiently provide nutrients for the fast growing crops to feed the fast growing population of the world. So the scientists are on the way to find out alternate strategies to increase the solubility of directly applied RP. One such strategy is the use of bio-inoculants, i.e. the use of PSMs and PGPRs and as explained in the earlier section. The other strategy is the mixing of RP with a well rotten product of crop residues and daily waste materials through a process known as composting. Many researchers have reported increased availability of P to the plants by increasing the solubility of RP in the soil through composting [86–89]. The mechanisms behind this solubilization includes the release of organic acids during the decomposition of organic residues which helps solubilize RP by lowering the pH, and by the conversion of inorganic P in RP into organic P which might become available to the plants after mineralization in the soil [89–91].

The processing of RP through the processes like that of composting is essential before applying to the alkaline soils found in Pakistan. In Pakistan, most of the phosphatic fertilizers are imported, as most of the reserves of available RP are of poor quality, which hinders its use for the preparation of phosphatic fertilizers. So as mentioned earlier, the possible strategy to cope with this situation would be to utilize the capacity of composting in increasing the solubility of raw RP. It is one of the most efficacious strategies for the recycling of organic waste materials which helps in boosting the level of organic matter, thereby playing an important role in productivity and sustainability of the soil [92]. Many researchers have authenticated the role of composting in ameliorating the soil fertility, structure, and plant growth [84, 89, 93–95]. They have found that organic acids (humic, fulvic acids, etc.) are released during this process which helps solubilize the fixed or unavailable P present in RP, thereby increasing the availability of P to the plants.

For composting process, a variety of waste material like cow dung, rice husk, poultry waste, fruit peels, mango stones, and many others could be used with RP, which does not only improve the physicochemical properties of the soil and a good source of nutrients [26], but also helps in cleaning our polluted environment. During this process, a variety of organic acids are produced, which helps reduce the pH, and ultimately results in better solubilization of P from the RP [89]. Composting process is very old and has been used for many centuries but up until now, very few or no studies have considered this process to be utilized for increasing the availability of P from the raw source of P, the RP. In a field experiment at the Agricultural Research Farm of NWFP Agricultural University, Peshawar, Pakistan, the effect of different levels of the combined application of chemical phosphatic fertilizer (0, 30, 60, 90 kg P₂O₅ ha⁻¹) and that of the compost (0, 5, 10, 15 Mg ha⁻¹) was investigated for improving nodulation,

growth, and yield of chickpea. There was no significant interaction effect of compost and phosphatic fertilizer that was suggested to be due to the P-deficiency of the experimental site. However, there was a significant effect of P on the nodulation and yield parameters studied, and 60 kg ha⁻¹ P₂O₅ from the phosphatic fertilizers was found to be optimum [96].

The quantity of available P varies with the nature of organic residues being used in the composting process and its rate of decomposition [84]. They found an increased citric acid solubility of RP by the composting of unreactive Mussoorie RP, chopped grasses, and tree leaves. Moreover, when this product was applied to the crop on equivalent total P basis, the grain and straw yield of the test crop, Guar, were similar to that of the application of SSP. Similar results were obtained using pigeon pea as test crop. The reason behind increased P availability from the phosphocompost might be due to the conversion of unavailable P in RP to water soluble P, thus increased efficiency of the dissolved P for the plant [97].

To investigate the impact of two RPs (nutriphos guano powder and Indian potash limited) with and without the composts (compost mulch and compost residues) on soil P pools and P uptake, a glasshouse experiment was conducted using wheat as test crop for 75 days in a loamy sand soil [98]. The composts were applied as a thick layer of 2.5 cm on the soil surface and nutriphos guano powder and Indian potash limited were applied at the rate of 35 and 26 mg P kg⁻¹, respectively. There was a control with an un-amended soil and amended soil with a soluble source of P (KH₂PO₄) at the rate of 50 mg P kg⁻¹ soil (inorganic P fertilizer applied soil). The results showed that there was no significant effect of the treatments on the total organic carbon concentrations and pH of the soil. However, the soil respiration was significantly higher in case of compost applied soil with and without RP compared to un-amended and inorganic P fertilizer applied soil. The plant growth was increased by 30–50% in the case of soils applied with compost alone or with RP sources. In case of the concentration of NaHCO₃-P and microbial P after 75 days of the applied treatments, an increase of 30% was noted in the case of the combination of compost with either RP compared to compost or RP applied alone, which suggested that compost helps in the mobilization of P present in RP. A significant change in labile pools of P was observed in case of the combined application of compost and RP compared to RP applied alone. Moreover, there was no significant effect on the plant growth and P uptake of wheat in case of combined application of compost and RP compared to compost applied alone, which shows that compost contains sufficient amounts of nutrients for the plants.

The organic matter in the soils or soil organic matter (SOM) is a good indicator of the productivity and quality of the soil as it mediates the physicochemical and biological properties of the soil. Many studies have authenticated the beneficial effects on the physicochemical, e.g. improves structure and water holding capacity of soils, decreases P fixation, increases CEC and buffer capacity of soils to resist pH change, and biological properties of the soil, e.g. provides energy to a variety of soil fauna and flora. For example, P sorption capacity of the soil was reduced through the addition of the SOM, which resulted in the alteration of chemical properties of the soil, e.g. complex formation of P compounds on the reaction sites [85]. The organic matter in the soil also serves as a reservoir of micro- and macronutrients.

The SOM exists as partially decomposed residual layer of plants and animals, microorganisms, and humus. The humus, a stable compound of carbon, comprises about 50–75% of the total

soil carbon. It has been found that the humus content of the soil are increased by the addition of organic fertilizers, and it also enhances the microbial activities in the soil [100]. It has also been speculated that farming methods aiming at increasing the organic matter in the soil would ultimately reduce the requirement of P application [100].

As mentioned earlier, SOM also serves as a basic source of mineral nutrients in the soil, e.g. nitrogen, sulfur, and phosphorus. About 95% of the total N and S, and up to 75% of P in the surface soil is in organic forms [101, 102]. The organic P in the soil exists in various forms of which phytic acid is the most important one. It is stored in the plant seeds to accomplish early establishment of the seedlings from the germinating seeds. Other organic compounds include mono and di-esters, phospholipids, nucleotides, sugar phosphate, phosphoproteins, and phosphonates [103]. The release of P from its organic compounds is not a very simple process and depends on many factors, like the relative stability of the organic substances and their chemical composition, climatic conditions, physicochemical properties of the soil, cropping scheme, and their interaction with mineral fertilizers [104–106]. Organic fertilization through compost application is a common practice for sustainable agricultural production and P cycling [107]. However, the amount or concentration and type of P compounds depend on the source of the material being used for composting [108]. Long term application of organic fertilizers in the form of composts results in boosting the organic P in the soil [109–111]. The conversion of this organic P into inorganic P or the mineralization depends on the type of compound being mineralized, e.g. orthophosphate di-esters are quickly mineralized compared to orthophosphate monoesters [112, 113]. Mineralization helps in increasing the total available P, which has been suggested to be due to reduction in the P adsorption and increased rates of microbial enzyme activities, which boost up the biologically mediated turnover of organic P into inorganic P [114].

It has been well established that the addition of organic fertilizers not only increases the organic matter and nutrient status of the soil, but also reduces the amount of costly chemical phosphatic fertilizers to be added into the soil, thereby, promoting healthier and sustainable environment of the soil [115]. It has also been documented that sustainable production from the continuous cropping system through the application of recommended levels of chemical fertilizers may not be possible in future [116]. The integrated use of chemical and organic fertilizers would be a sustainable strategy and has achieved substantial attention throughout the world. Chemical and organic fertilizers both supplement each other's efficiency for nutritional deficiencies and would ultimately decrease the exclusive dependence on chemical fertilizers [117].

Another field experiment was conducted to study the effect of different coir dust based composts on dry matter production and nutrient uptake of maize. It was found that the C:N ratio of the soil samples taken after 120 days of crop growth was decreased, and more biomass was produced with the application of organic fertilizer [118]. Another pot experiment using four different combinations of organic wastes including horse manure and bedding, sewage sludge along with clarifier solids from pulp mill, mink farm wastes, and municipal solid waste (MSW) was conducted to compare the capacity of different organic wastes to improve the growth and yield of tomato, and to assess the phytotoxicity of these organic wastes in radish and cress (*Lepidium sativum*) seedlings. It was concluded that paper mill waste, applied alone

without the application of organic fertilizer, causes toxic effect in vegetables, i.e. radish and cress [119].

During composting, organic acids like humic and fulvic acids are released, which decreases the pH of the material being composted [120], and increases the solubility of fixed P in the soils with high pH. These composts act like plant growth regulators when applied to the soil. The effect of these humic acids isolated from the cattle vermicompost was tested on the earliest stages of lateral root and on the plasma membrane H⁺-ATPase activity in an experiment using maize as a test crop. From the results, it was clearly found that the humic acids significantly increased the overall root growth of maize seedling in conjunction with lateral root emergence. Also a significant stimulatory effect on the activity of H⁺-ATPase was observed, which implies that the humic acids enhance the expression of this enzyme. Moreover, exchangeable auxin groups in the macrostructures of humic acid-containing compost, as revealed through structural analysis, shows that the hormonal activity is enhanced by the application of organic wastes containing humic acids [121]. In a pot experiment, to confirm the production and impact of organic acids on the growth and yield of crop plants during the composting process, the comparison of the impact of natural and synthetic humates like that of humic acids found in composts and exogenously applied synthetic humates, e.g. potassium humate on the growth of chicory plants and behavior of soil microbial population was studied. The results confirmed that during composting, there is production of organic acids, and these acids have a stimulatory effect on the growth and yield of crop plants and also increases the microbial population of beneficial microorganisms as observed in this study [122].

The impact of commercially produced vermicomposts produced from the mixture of cattle manures and food and paper wastes was tested in a two year field study using pepper (*Capsicum annuum* L.) as a test crop. It was suggested that increased growth and yield of pepper resulted due to the production of humic material and plant growth hormones by the increased microbial biomass present in the compost [123]. Similarly, they also found that the composted material significantly increase the soil microbial biomass and their dehydrogenase activities. Similar results were observed by Manna *et al.*, [124] who tested the effect of the application of compost and chemical fertilizers on the growth and yield of chickpea and wheat. They found that the compost application not only significantly enhance water soluble, citrate soluble, and total P in the soil, which resulted in an increased growth and yield parameters of chickpea and wheat, but also increase the microbial biomass and their enzyme activities.

The impact of different rates of application of vermicompost on the physicochemical properties of the soil was investigated in a field experiment using tomato (*Lycopersicum esculentum* L.) as test crop. The compost was applied into the upper 15 cm layer of the soil at the rates of 0, 5, 10, 15 Mg ha⁻¹. From the results, it was found that the application rate of 15 Mg ha⁻¹ significantly enhanced the electrical conductivity, total organic carbon, total N, P, K, Ca, Zn and Mn, as compared to that of the control. Overall, it was concluded that vermicompost can significantly improve the physicochemical properties of the soil [125].

As mentioned earlier, the effect of every compost/vermicompost depends on the chemical nature of that compost. It has been found that with the application of vermicomposts, there was an improvement in the biological properties, which resulted in an increased yield of the

rice, while the impact of cow dung in improving the biological properties of the soil was more pronounced as compared to that of the green forage [126]. Similar results were observed regarding the biological properties and fertility status of the soil [127, 128].

From the above discussion, it has been clear that different composts have a well renowned effect on improving the fertility status, physicochemical, and biological properties of the soil which ultimately results in improving the growth and yield of crop plants. Intensive agriculture has become essential to feed an ever-increasing population of the world. Similarly, burning of farm waste not only increases the environmental pollution due to the emission of CO_2 but also causes the wastage of nutrients and very precious organic matter. So depending solely on organic fertilizers would not be a wise strategy, instead the combined use of organic and an economical source of P like that of RP would serve better compared to the use of organic or chemical fertilizers alone [50]. Moreover, the composting process helps in the recycling and stabilization of organic wastes, which reduces their contribution to the environmental pollution, and this stable product can increase the plant production [129]. In this area of research, the interest has been increased and new strategies are underway to make a valuable product. These include partially acidulating RP with natural or synthetic organic acids as through composting, decreasing the particle size [130] and through the addition of bio-inoculants [131].

The properly employed process of composting converts the organic wastes into a stable and mature product of carbon, i.e. humus [132], while improperly composted organic wastes lead to the immobilization of plant nutrients and cause phytotoxicity [127, 133, 134]. During composting, heat is produced which helps in the destruction of pathogens as well [135]. The properties of composts depend not only on the chemical nature of organic wastes being composted but also the make-up of the organic material during composting [136]. In other words, stability and maturity indices are good indicators of the worth of composted material. However, it is very difficult to measure these indices and still no standards have been devised yet [137–140]. In general, the following parameters including C:N ratio, carbon contents (water soluble), CEC, humus contents, and the evolution of carbon dioxide from the finished composted material have been used to evaluate the stability and maturity of the composted material [141, 142]. For the measurement of the phytotoxicity of the matured composted material, germination index is used [143].

Many studies have proved that if we could find a natural and non-polluting way of increasing the solubilization of RP, its use could serve as a valuable substitutional source of chemical phosphatic fertilizers [144]. There have been many reports about the preparation of RP-enriched compost and their ability to increase the total P in the soil compared to the straw compost alone without RP, but the quantity of water soluble P is decreased in the RP-enriched compost compared to the straw compost or RP alone due to the dilution effect of RP being mixed with a large amount of composted material reaction of soluble P with CaCO_3 present in RP [64, 83]. On the other hand the RP-enriched compost has considerably more citrate soluble P compared to the straw compost, which is due to the production of organic acids like citric, oxalic, tartaric, 2-ketogluconic, acetic, malic, and succinic acids, etc., which enhance the dissolution of RP-P [64, 145]. These organic acids are in anionic form and produced during the

degradation of complex organic compounds [146]. A lot of CO₂ is produced during composting which results in the formation of carbonic acid, ultimately increasing the solubility of P in RP decreasing the pH [64, 147]. The RP-enriched compost has low microbial biomass carbon compared to the straw compost due to the dilution of carbon over a large biomass of compost material. The levels of organic P are also higher in case of RP-enriched compost compared to the straw compost as clear from the higher amount of alkaline phosphatases activities and acids [64]. The level of citrate soluble and organic P increases with increasing the addition of RP but up to a certain level and then decrease. So overall, during this organic matter decomposition, the available P and calcium contents are increased to the plants [147].

An experiment was conducted to compare the impact of pill millipede (*Arthrosphaera magna*) compost and farm yard manure, using black gram (*Phaseolus mungo*) and finger millet (*Eleusine coracana*) as test crop. From the results, it was found that the former compost resulted in better growth and yield of both the crops as compared to the farm yard manure, which was suggested due to the provision of plant nutrients present in both the types of composts [25]. Similarly the combined effect of mimosa compost and phosphatic fertilizers was more pronounced compared to the application of mimosa compost and phosphatic fertilizer alone [28]. Another field experiment found that RP-enriched compost performed better in case of yield and nutrients concentration in cow pea [24].

However, the extent of P solubilization depends on many factors like that of the ratio between the RP and compost, time and rate of application to the soils, which has not been done yet. It has been reported that P from this RP-enriched compost is available even at high pH of 8.5 or more [81], so it would ultimately serve as an economical and environment friendly way to reduce the use of chemical phosphatic fertilizers.

8. Integrated application of RP, PSMs, and compost (bio-organo-phos)

The PGPR include all the bacteria found in the rhizosphere, which directly or indirectly enhance plant growth. These may be involved in enhancing the availability of nutrient directly or through other indirect mechanisms like lowering the pH of rhizosphere. PSMs are the PGPR and can be utilized for enhancing the availability of P, which has a direct effect on nodulation. The combined application of RP-EC with PSMs could be helpful in improving the nodulation and the plant growth. There are few reports about their combined application. Shahzad and his co-workers [30] studied the effect of integrated use of plant growth promoting bacteria, and compost enriched with single super phosphate (SSP) for improving growth, yield, and nodulation of chickpea. Their results revealed that combined application of rhizobacteria and P-enriched compost resulted in an increase of 84, 97, and 79% in fresh biomass, number of pods plant⁻¹ and grain yield, respectively compared to uninoculated control without compost. So PGPB along with SSP-enriched compost was found highly effective in improving growth, yield, and nodulation of chickpea as compared to their application alone.

A field experiment was conducted to study the impact of four different agro-industrial wastes inoculated with PSMs (*Aspergillus niger* and *Phanerochaete chrysosporium*) in increasing the

availability of P from RP. From the results it was found that with the application of *Aspergillus niger*, 59.7, 42.6, and 36.4% of the total P present in the RP was released in the case of the application of sugar beet wastes, olive cake, and olive mill wastewaters, respectively which was suggested to be due to the secretion of organic acids. Overall, the growth and yield of the plants was increased with the combined application of RP, *Aspergillus niger* and different agro-industrial wastes, i.e. sugar beet wastes, olive cake, olive mill wastewaters, and dry olive cake [27]. Series of field and pot experiments were conducted in our lab to check the combined effect of RP, compost and PSMs in increasing the growth and yield of wheat. From the results, it was found that the combined application of RP-enriched compost with PSMs could serve as an alternate source of P for increasing the growth and yield of many crops, thereby leading to sustainable agriculture and cleaner environment [29]. Recently, we have conducted a series of pot and field experiments and their results have clearly shown better improvement in growth, nodulation, and yield of lentil compared to conventional use of chemical phosphatic fertilizers [31].

9. Conclusions and perspectives

For sustainable agriculture, there must be a positive link between the nutrients applied to the soil and crop uptake. This link could be more firm and sustainable if we employ an integrated approach, i.e. the use of bio-augmented RP-enriched organic fertilizer. In this way, we would be employing a sustainable approach to meet the needs of crops for P in an environment friendly way. The use of this approach would not only be helpful in the restoration of degraded soils, but would also be helpful in minimizing organic wastes that could be composted to make organic fertilizer. The use of indigenous sources of RP would help minimize the energy use during its conversion into chemical phosphatic fertilizers. The product would help the farmers in reducing their expenditures to purchase chemical fertilizers, and would also reduce the import budget on national level. However, a few studies have reported its efficacy under pot and field conditions. More studies under controlled and field condition are needed to confirm these reports. Also there is a need to find the optimum ratio to mix organic fertilizer and RP, its time, and rate of application. Relative efficacy of different sources of organic fertilizers to make bio-organo-phos could also be searched out. Their post-harvest effects on soil physico-chemical properties and on microbial community structure could also be found out in future.

A variety of organisms are involved in P cycling in soils, and microorganisms are probably the most important ones. However, most of the soil microbes have not been cultured successfully [148]. In the future, new culture-independent methods like LMW RNA profiling and PCR based on nucleic acid composition are required to study the function and ecology of microbes involved in P cycling in soils [149]. The techniques mentioned have been found not only independent of culture media composition or growth phase of microorganisms but also are precise and reproducible. These techniques also made it possible to utilize different biotechnological tools like amplification of targeted genes or to quantify their expression. Overall, these techniques have opened new horizons in order to solve out the puzzle related to

microbial community ecology and to assess the survival and persistence of specific inoculants under different environmental conditions.

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