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Improvements Availability Phosphorus (P) On Acid Soil Through Utilization Of Phosphorus (P) Solubilizing Microbial And Organic Matter.

Muh. Jayadi^{1*}, Burhanudin Rasyid¹, Asmita Ahmad¹, Rina Mariana², and Ahdin Gassa³.

¹Soil Sciences Department, Agriculture Faculty, Hasanuddin University ²Alumnus of Soil Sciences Department, Agriculture Faculty, Hasanuddin University ³Department of Plant Pests and Diseases, Faculty of Agriculture, The University of Hasanuddin

ABSTRACT

Phosphorus (P) as an essential nutrients has lower availability on acid soils because it has been fixation by Al, Fe and Mn element. To increase the availability of P in acid soils, it can be used phosphate rock (BF), organic matter and Phosphate Solubilizing Microbial (MPF). The purpose of this research is testing the MPF ability to dissolve BF at different levels fineness on pikovskaya liquid media and to study the effect of MPF combined with various compositions of compost to increase the availability of P in acid soils. There are two combination to dissolve BF in 60, 120 and 230 mesh on pikovskaya liquid media. The first combination is AB (Aspergillus sp. and Bacillus sp.) and the second is ABP (Aspergillus sp., Bacillus sp. and Pseudomonas sp.). Dissolved P is defined by Murphy and Riley method. There are two test for P availability in acid soil with pot; 1) using zeolite 5% with four treatment; there are a) BF with MPF combined with rice straw compost 100% (KJ), b) rice straw compost 75% + Gliricidia compost 25% (KJG), c) BF with MPF combined with rice straw 75% + cow manure 25% with zeolite 5% (KJS), d) BF with MPF combined with rice straw compost 75% + 12.5% Gliricidia compost + cow dung 12.5% (KJGS), and 2) not using zeolite but with same treatment as the point one. Availability P determined by Bray II method. The result showed that P solubility in BF 230 mesh on pikovskaya liquid media with ABP increasing 105.29% while with AB only 80.30% comparing to controls. BF fineness of 230 mesh increasing the solubility of P is better than the other sizes. ABP combination with a fineness of 230 mesh BF improve the solubility of P 120.39% comparing to controls. MPF treatment increases the availability of P2O5 10.25% - 38%, while the compost treatment was 17.12% - 27.79%. Utilization ABP with BF (230 mesh) produce the highest P soluble in liquid pikovskaya media. The best availability of P in acid soils was on a combination of compost KJS + zeolite 5% with MPF1 (48, 64%).

Keywords: Rock phosphate, compost, phosporus solubilizing microbial, pikovskaya, acid soil.

*Corresponding author:



INTRODUCTION

Efforts to increase food production gets serious attention from the agriculture ministry for food supply to get target of food self-sufficiency. In Indonesia, dry lands are generally reacted sourly, and have wide spread around 102.8 million ha from 148 million ha of the total dry lands [16]. Seeing the extent of the land reacted sourly, it is necessary to get attention so that the utilization can be optimized. According [1,33], the main obstacle for acid soil was the low content of cation exchange capacity (CEC), base saturation and C-organic, and high contenct of aluminum (AI), iron (Fe), manganese (Mn), phosphorus (P) fixation. It also was sensitive to erosion and poor biotic elements.

Phosphorus (P) is an essential nutrient for plants are required in large numbers. P is the main component of nucleic acid, phospholipids, ATP and some coenzyme. The use of triple superphosphate (TSP) that is easily soluble have a very low effectiveness on acid soils, only about 10-25% [13]. The low effectiveness due to the fixation of P by Fe and Al. Thus in acid soil P deficiency is often as a factor limiting plant growth. Therefore, P fertilization should be done to increase agricultural production

Phosphate rock is one of the alternative P sources that can be applied on acid soils because its relatively cheap price than super phosphate, containing much P and Ca, and slow release, so it can improve the efficiency of fertilizer on dry land [27]. BF utilization can help to reduce soil acidity [15]. BF applications with 1000 kg ha-1 in Oxisol and Ultisol will increase maize yields between 30-90% over five seasons [27].

Rocks phosphate have low solubility, and difficult to meet the needs of plants. Rocks phosphate have to decompose in advance so that it can be used by plants. One way to increase the solubility of the phosphate rock is mixtured with organic materials from agricultural waste or manure, and it can combine with microbial phosphate solvent either alone or in combination. It can increase the availability of P in Ultisol [21].

Several types of bacteria that can dissolve P are *Bacillus* sp. and *Pseudomonas* sp. [32,23,41,40]. Fungus group that have a high ability to dissolve phosphate is *Penecillium* sp. and *Aspergillus* sp. [2,8,20,18,22]. Improved solubility BF by microbes caused by decreased the pH and the excretion of organic acids [6].

The use of organic materials can be a positive influence on BF solubility in soil, it is caused by the presence of organic acids. The organic acid can dissolve the Ca-P, Al-P and Fe-P through its ability in cations complexation so that P available to plants [39]. Other than that the organic material can also improve the cation exchange capacity (CEC), soil bearing capacity, nutrients availability and phosphorus absorption efficiency. Organic matter can produce N, P, K, S and other nutrients [11]. Organic matter compost can be enhanced through the addition of zeolite that has a high CEC.

Zeolites utilization in composting can improve chemical properties and increase the N content of compost. Increasing N content occurred through adsorption by zeolite, and can be released back for the plants absorbed [36]. Binding cations such as Ca, K and Na by zeolite occurs in two ways adsorption, namely; in the pores of the matrix and through the exchange of cations. [24] stated that zeolite given on fertilizers can help to hold nutrients thus allowing longer available for absorbed by plants, reducing nutrient losses and improve use efficiency of N and K fertilizer.

Through the utilization of organic material with zeolite and microbial phosphate solvent, so the availability of P from phosphate rock can be improved. Thus the problem of P in acid soils can be resolved.

MATERIALS AND METHODS

MPF Ability Test to Dissolve Rock Phosphate in Liquid Media

MPF ability to dissolve phosphate did with liquid pikovskaya media. Into 25 ml Pikovskaya liquid medium was add 5 g 1000 ml BF-1 with subtlety respectively 60, 120 and 230 mesh, then it autoclaved at 121°C temperature for 20 minutes. Into each sterile media are inoculated MPF with three treatments: (1) without the MPF, (2) a mixture of *Aspergillus niger* (JTM 2) + *Bacillus* sp. (JTM 3) or AB, (3) a mixture of *Aspergillus niger* (JTM 2) + *Bacillus* sp. (JTM 10) or ABP. Into Pikovskaya solution



was added 1 ml (1x10⁸ cfu/ml) for *Bacillus* sp. (JTM 3) and *Pseudomonas* sp. (JTM 10), while the *Aspergillus niger* fungus (JTM 2) was added 1 ml (1x106 cfu/ml). BF and MPF degree of fineness combination produce 9 treatments. The inoculation treatment is placed on a shaker for 10 days at 120 rpm. The suspension was filtered with Whatman 40 and then centrifuged for 15 minutes at a speed of 10,000 rpm. Determination of P soluble in media with [17] method, using Spectrophotometer at 693 nm wavelength which is calibrated with KH2PO4 standard curve

MPF Ability Test and Organic Materials to improve P availability of Rock Phosphate in Acid Soil

Testing capabilities of isolates MPF in dissolving BF on pikovskaya media was followed on acid soil in the pot with a dose equivalent to 100 kg P ha-1. Acid soil samples that used in ths treatment has properties such as; silty clay loam texture, pH H2O (4.37), C-organic (1.28%), Total-N (0.27%), P-available (6.85 mg.kg-1), cation exchanged (Ca, Mg, K and Na) respectively to 2.52, 1.55, 0.03 and 0.24 cmol (+) kg-1), cation exchange capacity (19.65 cmol (+) kg-1), base saturation (22.09%), Al-dd (5.88 cmol (+) kg-1). Compost used is rice straw composted, gliricidia leaves composted, and cow dung composted.

Compost composition testing has two units, namely: (1) compost with 5% zeolite, and (2) compost without zeolite. Treatments are arranged with a randomized block design and analyzed in a separate plot, which consists of 4 treatment composition of compost as main plot and 4 treatment MPF as a subplot. The composition of the compost on unit 1 and unit 2 were combined with the MPF (mixture of *Aspergillus* sp., *Pseudomonas* sp. and or *Bacillus* sp.).

Compost composition treatment as the main plot, namely: (1) 100% straw (KJ), (2) 755 straw + 25% Gliricidia leaves (KIG), (3) 75% straw + 25% cow manure (KJS), and (4) 75% straw + 12.5% gliricidia leaves + 12.5% cow manure (KJGS), each composition of the compost is given 5% zeolite (Z) and without zeoilt. Chemical properties of compost are listed in Table 1.

No.	Compost Composition	Total-N (%)	P ₂ O ₅ (%)	К (%)	CEC (cmol kg ⁻¹)	рН
1	KJ	0,66	0,35	2,66	37,17	7,21
2	KJ + Z 5%	0,61	0,36	2,59	29,26	7,02
3	KJG	1,19	0,63	2,31	31,68	6,44
4	KJG + Z 5%	0,97	0,42	2,26	33,25	6,46
5	KJS	1,06	0,71	2,35	34,85	7,25
6	KJS + Z 5 %	0,95	0,35	2,27	35,77	7,14
7	KJGS	1,02	0,58	2,37	37,82	6,76
8	KJGS +Z 5%	1,00	0,73	2,36	35,72	6,80

Table 1. Characteristic of compost chemical properties that use in this study

MPF Treatment as subplots, namely; (1) without MPF (MPF0), (2) a mixture of *Bacillus* sp. JTM3 + *Aspergillus* sp. JTM6 + *Pseudomonas* sp. JTM10 (MPF1), (3) a mixture of *Aspergillus* sp. JTM2 + *Bacillus* sp. JTM3 + *Pseudomonas* sp. JTM10 (MPF2), (4) a mixture of *Aspergillus* sp. JTM2 + *Pseudomonas* sp. JTM10 (MPF3). This research was conducted at Soil Microbiology Laboratory and Soil Chemistry Laboratory, Department of Soil Sciences, Faculty of Agriculture Hasanuddin University.

RESULTS AND DISCUSSION

Ability of MPF to dissolve phosphate rock in pikovskaya liquid media

Ability to dissolve phosphate rock (BF) quantitatively of the 3 isolates of phosphate-solubilizing microbe (MPF) tested in pikovskaya liquid media containing BF with different smoothness showed that there was a decrease in pH of liquid media inoculated by MPF (AB, ABP) compared to control pH (without MPF inoculation). Incubation of MPF for 12 days decreased the media pH between 4.68 – 6.23, while control remained in neutral range, namely pH 7.57 – 7.93 (Table 2). Solubility of BF that was higher in pikovskaya liquid



media was obtained in ABP treatment with the smoothness of BF 230 mesh, namely 55 mg P l^{-1} , that was the lowest in the treatment without MPF and BF 60 mesh (23.61 mg P l^{-1}).

No	Treatment	Content of dissolved P (mg P I ⁻¹)	pH of pykovskaya liquid media
1	ABP + BF 230 mesh	55.09	4.68
2	ABP + BF 120 mesh	50.88	5.24
3	ABP + BF 60 mesh	47.97	5.39
4	AB + BF 230 mesh	46.65	5.54
5	AB + BF 120 mesh	45.41	5.71
6	AB + BF 60 mesh	43.90	6.23
7	Ktrl + BF 230 mesh	25.80	7.57
8	Ktrl + BF 120 mesh	25.58	7.75
9	Ktrl + BF 60 mesh	23.61	7.93

Table 2. Solubility of P of phosphate rock and change of pH of pikovskaya media by M	1PF
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Remarks:

AB : *Aspergillus* sp. + *Bacillus* sp.

ABP : Aspergillus sp. + Bacillus sp. + Psudomonas sp.

Solubility of BF based on the effects of treatment of smoothness level showed that the smoother the BF the higher the solubility of BF, from the BF smoothness 60 mesh (38.49 mg P $|^{-1}$) to become 42.51 mg P $|^{-1}$ on BF 230 mesh or 10.44 % higher. Solubility of BF based on the effects of MPF treatment showed that ABP (51.31 mg P $|^{-1}$) > AB (45.32 mg P $|^{-1}$) > control 25 (mg P $|^{-1}$), with respective pH of 5.10, 5.83 and 7.75 in a row (Figure 1). Therefore, it was seen that in ABP treatment whose isolates were more diversed, P dissolved was higher. This was probably because of the more diversed amounts and types of organic acids that were produced. Each type of organic acids has different abilities in dissolving phosphate [4].

Higher solubility of BF in liquid media (Figure 1) was also related to lower pH. Composite of ABP isolates decreased the media pH from neutral to pH 5,10 and dissolved 51.31 mg P I^{-1} of BF. AB isolates decreased media pH media to pH 5.83 and dissolved 45.32 mg P I^{-1} BF, while the one without phosphate-solubilizing microbe, the solution pH remained neutral (7.73) and dissolved BF was only 25 mg P I^{-1} . Therefore, combination treatment of MPF composite *Aspergillus* sp + *Bacillus* sp + *Psudomonas* sp (ABP) and BF having smoothness of 230 mesh produced higher dissolved P compared to other treatments.



Figure 1. Effects of MPF (ABP and AB) treatment and BF smoothness on solubility of BF and pH of pikovskaya solution.

Solubilization of P by MPF in liquid media is caused by organic acid production, pH decrease and the presence of chelates toward kations by organic acid, so P becomes dissolved [18,25,7]. Decrease of media pH in MPF treatment was caused by organic acid production [26] and by MPF respiration [12]. Decrease of pH

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accompanied by organic acid production will increase P solubility [5,25,6]. Decrease of pH causes the change in organic acid produced by MPF was dominantly in citrate acid [9]. Citrate acid has bigger power to dissolve P than do malate and oxalate acids [4]. Meanwhile, solubilization of P at relatively higher pH is caused by organic acid chelation toward Ca ion in tricalcium phosphate [12].

Effects of Phosphate-Solubilizing Microbe and Compost with or without Zeolite on the Availability of P in Acid Soil

Availability of nutrient element of P was low in acid soil due to high fixation in soil and due to low diffusion distance (0.02 cm) compared to N (1 cm) [39] therefore, plants often underwent problems of nutrient element of P. Low availability of P in acid soil occurred because it was adsorbed on clay mineral surface due to the formation of complex with hydro oxide Fe and Al [31], and the occurrence of deposit of P caused by free ions Al^{3+} and Fe^{3+} in soil [10].

Results of variety print toward availability of P_2O_5 in soil showed that treatment of sub-sector of phosphate-solubilizing microbe (MPF) and main sector (compost composition) individually and in combination significantly affected on P available in soil. BNT test at trust level of 95% (Table 3) showed that straw compost (KJ) without MPF was significantly lower compared to other compost compositions, while if combined with MPF differences, between compost compositions were not seen. Nevertheless, the highest result was obtained in the treatments of combinations of KJG and MPF3 (12.68 mgkg⁻¹), KJS and MPF1 (12.62 mgkg⁻¹), and the lowest was obtained in the treatment of combination of KJ and MPF0 (7.03 mgkg⁻¹) on compost without zeolite.

Compost	P ₂ O ₅ available in soil (mgkg ⁻¹)					
treatment	MPF0	MPF1	MPF2	MPF3		
	Without addition of zeolite in compost					
КJ	7.03 с у	11.18 ax	8.32 by	10.78 ax		
KJG	10.79 b x	11.66 ab x	10.96 bx	12.68 a x		
KJS	10.39 c x	12.62 a x	10.67 bc x	11.67 ab x		
KJGS	10.75 a x	11.27 ax	11.12 ax	10.85 a x		
	Addition of zeolite 5% in compost					
KJZ	8.32 c y	13.85 a x	11.43 bx	13.82 a x		
KJGZ	11.13 b x	11.58 ab y	11.51 ab x	12.95 a x		
KJSZ	10.43 c x	13.96 a x	11.27 bc x	12.35 ab x		
KJGSZ	9.68 c xy	12.57 a xy	11.80 ab x	10.47 bc y		

Table 3. Effects of treatment of compost and phosphate-solubilizing microbe (MPF) combination on P₂O₅ available in soil

Remarks: Numbers followed by the same letter (a,b,c) in the same line, and the same letter (x, y) in the same column means that they are not significantly different at the level of test BNT 0.05. with differentiating value 1.862 for line and 2.101 for column (without zeolite) and 1.843 for line and 0.857 for column (zeolite 5%).

KJ = Rice straw; KJG = Rice straw+Gliricidia spp. leaf; KJS = Rice straw+cow faeces; KJGS = Rice straw+Gliricidia spp. leaf+cow faeces; Z = zeolite 5%.

Treatment of compost accompanied by zeolite 5% resulted in P_2O_5 to be the most available, namely in the combination of KJSZ and MPF1 (13.96 mgkg⁻¹), combination of KJZ and MPF1 (13.85 mg kg⁻¹) and combination of KJZ and MPF3 (13.82 mgkg⁻¹) and the least in the treatment of KJZ and MPF0 (8.32 mgkg⁻¹). Overall, when compared on average between the treatment that was not given zeolite (10.67 mgkg⁻¹) and the treatment that was given zeolite 5% in compost (11.69 mgkg⁻¹), it could be seen that there was an increase of P_2O_5 available in soil amounting 9.58 % due to the addition of zeolite. It was formerly reported by Omar *et al.* (2011) that composite of sago waste, zeolite and urea could increase changeable ammonium, K, Ca, Mg and P availability.

When looked closely, the single effect of compost and MPF (Figure 2) on P availability, it was obvious that in the treatment of compost without zeolite (KJS), the highest result was 11.34 mgkg⁻¹, while in the treatment of compost with zeolite, treatment of KJSZ gave the highest result (12.00 mgkg⁻¹) and the lowest was given in the treatment of KJ (8.83 mgkg⁻¹) or 35.39% lower. When compared between treatment of BF without compost and treatment with BF + compost, P availability increased 17.12 % - 27.79 % whereas in

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treatment without BF and without compost (KT), P available was only 6.83 mgkg⁻¹, increasing to 9.39 mgkg⁻¹ in treatment that was given BF or the increase was 36.28%.



Figure 2. P₂O₅ available in soil in compost treatment

The increase of P available through compost application originated from the presence of humus as the result of the process of decomposition. Humus consisted of complex composites such as fulvate (AF) and humate acid (AH) that was the source of negative content in soil. The negative content mainly originated from carboxyl group dissociation (R-COOH) and the fenolic hydroxyl (R-COH) [3,38], alcoholic hydroxyl, kuinoid, ketonic (C=O) and coordination group such as amine [34]. Through those functional groups, humus can react with metal ions such as AI, Fe, Ca and Mg forms chelate compounds, so it can minimize its reactivity toward P and therefore P becomes more available. Result of organic material decay in soil in the form of humate material is about 70 - 80 % [31]. Besides, in organic material mineralization process, there will be a release of nutrients needed by plants such as N, P, K, Ca, Mg and S.



Figure 3. P₂O₅ available in soil in phosphate-solubilizing microbe (MPF) treatment

In MPF treatment (Figure 3), it was shown that isolate of composites of *Aspergillus sp., Bacillus sp.,* and *Pseudomonas sp.* (MPF1) that gave the highest result was MPF1 treatment (12.99 mgkg⁻¹) or 40.59 % higher than the treatment without phosphate-solubilizing microbe (MPF0) (9.24 mgkg⁻¹). Treatment with MPF increased the availability of P_2O_5 in acid soil between 10.25% and 38.48 %, compraed to the treatment without MPF. The ability of microbe to dissolve P from both bacteria group such as *Pseudomonas* and *Bacillus* and fungi group such as *Penicillium* and *Aspergillus* was already stated by [35]. [35,28] state that the ability to dissolve P is caused by the presence of organic acid secretion such as formic acid, acetate acid, propionate, lactate, glicolate, fumarate and succinic. The low pH of organic acids that are produced result in the breaking of

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phosphate binding. Besides, hydroxide acid can form chelate with Ca, Al and Fe so solubilization and utilization of phosphate are more effective.

MPF microorganism acivities excreting organic acids such as monocarboxilate acid (acetate acid), dicarboxilate (oxalate acid, fumarate) and tricarboxilate/polycarboxilate (citrate acid, glutamate), can increase P available in soil. Mechanism of P preparation due to organism acid is decrease in pH, increase in chelation toward kation that binds P, competition with P in soil adsorption site and formation of complex with metal ions such as Al, Fe and Ca, so P ion is released from the binding, so it becomes available for plants [42,14,30. [19] states that some species of fungi such as genus *Aspergillus* have higher abilities in dissolving phosphate compared to bacteria. Therefore, there is a good opportunity to develop them in tropical acid soil, because fungis favor growth environment that is acid.

CONCLUSION

Phosphorus higher solubility on the media pikovskaya is on a mixture of isolates; *Aspergillus sp., Bacillus sp., and Pseudomonas sp.* compared with a mixture of isolates; *Aspergillus sp., and Bacillus sp.* Phosphorus (P) solubility is higher in BF fineness of 230 mesh size compared to other size. The combination of *Aspergillus sp., Bacillus sp. and Pseudomonas sp.* with BF fineness of 230 mesh increase the solubility of P 120.39% compared to controls. MPF treatment on acid soils increase the availability of P₂O₅ in range 10.25% - 38.48%, while the compost treatment was in range 17.12% - 27.79%. Phosphorus (P) availability in acid soils is highs on a combination of compost KJS + 5% zeolite with a mixture of *Bacillus sp* JTM3+ *Aspergillus sp* JTM6+ *Pseudomonas sp.* JTM10 (MPF1) (13.96 mg kg⁻¹) or 48.64% compared to the treatment without compost and MPF.

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