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Production of phospho-vermicompost by earthworms mediated bio-conversion of organic residues and rock phosphate

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Abstract

A glass house experiment was conducted to develop phospho-vermicompost using different combination of organic residues amended with rock phosphate and earthworms at the Soil Science Division, BINA, Mymensingh. The experiment was conducted in a Completely Randomized Design with eight treatments and three replications. The treatments were as T₁:50% Cowdung (CD) + 50% Mustard straw (MST), T₂:50% CD + 50% Water hyacinth (WH), T₃:50% CD + 50% Rice straw (RST), T₄:50% CD + 25% WH + 25% RST, T₅:50% CD + 50% MST + 4% Rock phosphate (RP), T₆:50% CD + 50% WH + 4% RP, T₇:50% CD + 50% RST + 4% RP, T₈:50% CD + 25% WH + 25% RST + 4% RP. About 150 earth worms (Eisenia foetida or Red wiggler earthworms) were released on partial decomposed residues into the pots. At the end of the incubation, population of earthworms, total bacteria and phosphate solubilizing bacteria were determined from prepared vermicompost. pH, organic carbon, total N, P, K, S, available P and alkaline phosphatase activity were also determined from prepared vermicomposts. pH was found almost similar in all the treatment combinations but with rock phosphate amended treatments showed greater pH than without rock phosphate amended treatments. The treatment T₈ (50% cowdung +25% water hyacinth +25% rice straw with 4% rock phosphate powder + red wiggler earthworms) showed significantly highest population of earthworms, total bacteria, phosphate solubilizing bacteria (PSB) and alkaline phosphatase activity (ALPA) and gave lower C:N and C:P ratio which might be resulted the higher amount of nutrients including available P in mature vermicompost. However, among the treatments, the treatments T₈ gave the highest total N (1.42%), P (1.45%), K (1.52%) and S (0.35%) and available P contents than that of other treatments which indicated the better quality of phospho-vermicompost. Therefore, 50% cowdung +25% water hyacinth +25% rice straw with 4% rock phosphate powder and red wiggler earthworms could be used for the production of phospho-vermicompost. The developed phospho-vermicompost could be used for supplement of phosphatic fertilizer and other chemical fertilizers in the cultivation of different crops and also could be saved of chemical fertilizers.

Key words: Phosho-vermicompost, rock phosphate, organic residues, PSB population, phosphatase activity, nutrients availability

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Introduction

Organic matter and plant nutrients of Bangladesh soils are declining day by day due to soil mining with

intensively crop cultivation for feed the ever rising population. Exploiting of plant nutrients did not

replenish with proper organic manuring. Different ways could be found to replenish the soil organic matter like green manuring, application of cowdung, farmyard manure, crop residues etc. Those materials have detrimental effects on the environment and they contained also low available plant nutrients. But vermicompost is prepared with different organic residues with the action of earthworms and native microorganisms. Microorganisms and earthworms are important biological organisms helping nature to maintain nutrient flows from one system to another and also minimize environmental degradation (Lazcano et al., 2018; Singh and Amberger, 1990). The earthworms ingested the decomposing residues and leave it as excreta in their life activities. Suhane (2007) reported that total bacterial counts exceeded 10⁻¹⁰/g of vermicompost and it included nitrobacter, azotobacter, rhizobium, phosphate solubilizers and actinomycetes. Several findings also showed considerable increase in total viable counts of actinomycetes and bacteria in the worm treated (Haritha Devi et al. 2009). The increase of microbial population may be due to the congenial condition for the growth of microbes within the digestive tract of earthworm and by the ingestion of nutrient rich organic wastes which provide energy and also act as a substrate for the growth of microorganisms (Tiwari et al. 1989). Worm casts contain higher activities of cellulase, amylase, invertase, protease, peroxidase, urease, phosphatase and dehydrogenase (Sharpley and Syers 1976; Edwards and Bohlen 1996) which accelerate the decomposition during the vermicomposting than normal composting. Therefore, vermicompost get more stable condition with lower carbon, nitrogen and phosphorus ratio and higher available plant nutrients than normal compost (Joshi et al. 2015).

In order to maintain the soil fertility and productivity, integrated nutrient management with sustainable organic fertilizers is very important (Souzaa *et al.*, 2013; Parastesh *et al.*, 2019). Rock phosphates are applied in agricultural field to improve the availability of P in soil. However, the major disadvantage of

applying rock phosphate is low dissolution rate resulting low availability which do not match to the P demand of crops (Edwards et al., 2010). Thus, finding ways to increase the solubility of rock phosphate is of great interest. Many infrastructures, energy, money and technical knowledge are required for manufacturing of phosphatic fertilizers by using rock phosphate in the industry. But phosphate rich organic fertilizer like phospho-vermicomost can be produced easy and cheap through bioconversion of organic materials and rock phosphate with effective earthworms (Shiraz et al., 2020; Hashemimajd and Golchin, 2009; Garg and Kaushik, 2005). Phospho-vermicompost could be made by mixing of rock phosphate with various organic materials through decomposition with the help of earthworms (Mohammady et al., 2010; Lime et al., 2012) which can meet the full demand of phosphorus fertilizer for many crops. It may play a vital role for restoring of soil fertility and soil health and saving of phosphatic fertilizer and other chemical fertilizers in crop cultivation. But information is scanty about the preparation of phospho-vermicompost in Bangladesh. Therefore, the present study has been undertaken to Phospho-vermicompost develop using different combinations of organic residues with rock phosphate and earthworms.

Materials and Methods

A glass house experiment was conducted to develop phospho-vermicompost using different combination of organic residues with rock phosphate and earthworms at the Soil Science Division, BINA, Mymensingh during 2019-2020. The experiment was conducted in a Completely Randomized Design with eight treatments and three replications. The treatments were as follows:

T₁: 50% Cowdung (CD) + 50% Mustard straw (MST)

 $T_2:50\%$ CD + 50% Water hyacinth (WH)

T₃:50% CD + 50% Rice straw (RST)

T₄: 50% CD + 25% WH + 25% RST

T₅:50% CD + 50% MST + 4% Rock phosphate (RP)

T₆:50% CD + 50% WH + 4% RP

T₇: 50% CD + 50% RST + 4% RP

 T_8 : 50% CD + 25% WH + 25% RST + 4% RP

Nutrient contents of different organic residues have been given in the Table 1. Rock phosphate (@ 4%) was mixed well with the mixtures of organic residues as treatment plan into the plastic pots (50 L) and all the pots were pre-incubated until three weeks. After softening and partial decomposed of residues then 150 earth worms (*Eisenia foetida* or Red wiggler earthworms) were released into all the plastic pots. All the pots were covered with gunny bag to make dark condition. Small amount of water was sprayed if necessary to avoid the dryness of the residues.

Table 1. Nutrient contents in different organic residues used in the experiment.

Organic residues	Org C (%)	%N	%P	%K	%S	C:N ratio	C:P ratio
Water hyacinth	45	0.65	0.35	1.8	0.23	70.7	128.6
Cowdung	38	0.65	0.3	0.46	0.23	58.5	126.6
Rice straw	48	0.67	0.17	1.4	0.13	85.7	282.3
Mustard straw	46	0.57	0.13	1.3	0.20	80.7	353.8

After 90 days of incubation, the residues were completely decomposed and no bad odor was observed. So phospho vermicomposts were ready for collection. Then the samples were collected from each pot for chemical analysis and estimation of earthworms, total bacteria, phosphate solubilizing bacteria and phosphatase activity. The prepared samples were analyzed for pH, organic carbon, total N, P, K and S and available P. Plant residues, cowdung and prepared phospho-vermicompost were analysed with the standard methods which were as follows:

pH of prepared phospho-vermicompost: pH was measured by a glass electrode pH meter using phospho-vermicompost: water suspension of 1:2.5 as described by Jackson (1967).

Organic carbon from organic residues and prepared phospho-vermicompost: Organic carbon was determined by wet oxidation method as described by Black (1965) from residues, and phosphovermicompost. The underlying principle was used to oxidize the organic matter with an excess of 1N K₂Cr₂O₇ in presence of conc. H₂SO₄ and conc. H₃PO₄ and to titrate the excess K₂Cr₂O₇ solution with 1N

FeSO₄. The results were expressed in percentage (Page *et al.*, 1989).

Total N from residues and prepared phosphovermicompost: Total N content was determined following micro-Kjeldahl method as described by Jackson (1967) from residues and vermicompost. Each sample was digested with H₂O₂, conc. H2SO4 and catalyst mixture (K₂SO₄:CuSO₄.5H₂O:Se in the ratio of 100:10:1). After completion of digestion, made the volume to 100ml. Distillation was performed with adding of 40% NaOH into the digest. The distillate was received in 2% boric acid (H₃BO₃) solution and 4 drops of mixed indicator of bromocresol green and methyl red solution. Finally, the distillate was titrated with standard H₂SO₄ (0.01N) until the color changed from green to pink. Then amount of N was calculated.

Total P, K and S from organic residues and prepared phospho-vermicompost: Total P, K and S were determined following micro-Kjeldahl method as described by Jackson (1967). About 0.5 g of samples was transferred into dry clean 100 ml kjeldahl flasks. 10 ml of di-acid mixture (HNO₃:HClO₄ = 2:1) were

added into the flask. After leaving for a while the flasks were heated at temperature slowly raised to 200°C. The contents of the flaks were boiled until they became sufficiently clear and colorless. After cooling the digests were transferred into 100ml volumetric flaks and the volumes were made up to the mark with distilled water. Phosphorus was determined by developing blue color by SnCl₂ reduction of phosphomolybdate complex and measuring intensity of color by calorimetrically at 660 µm wave length and the readings were calibrated to the standard P curve. Potassium was determined from the aliquot by flame photometer (Black, 1965) and calibrated with a standard curve. Sulphure was determined from aliquot by calorimetrically at 440 µm wave length (Williams and Steinbergs, 1959) and the readings were calibrated to the standard curve.

Available phosphorus from prepared phosphovermicompost: Available phosphorus was extracted from the phospho-vermicompost samples by shaking with 0.5 M NaHCO₃ solutions at pH 8.5 following the method of Olsen and Sommers (1982). The extracted phosphorus was determined by developing blue color by SnCl₂ reduction of phosphomolydate complex and measuring the intensity of color calorimetrically at 660 nm wave length and the readings were calibrated to the standard P curve.

Counting of earthworms: Earthworms were counted at the end of the incubation i.e. at the mature of vermicompost from each pot. Visible earthworms were carefully sorted from the decomposted materials manually and counted.

Estimation of total bacteria in prepared vermicompost: The dilution plate method was used for enumeration of total bacteria from prepared vermicompost (Subba Rao, 1993). The ingredients of the methods were soil extract –glucose agar medium in which 1.0 g glucose, 0.5g K₂HPO₄, 0.1 g KNO₃, sterilized soil extract 100ml, distilled water 1000ml, pH adjusted 6.5 and agar 20 g and autoclaved. Soil extract was prepared by autoclaving 500 g of fresh soil

with the addition of 1000 ml tap water at 121°C for 20 min at 15 PSI and filtered through Whatman No. 1 filter paper. Serial dilutions were prepared by weighing of 10 g vermicompost samples in 90 ml sterile water blank and shaked vigorously. This makes 10⁻¹ dilution. After shaking, immediately transfered 1 ml of suspension from 10⁻¹ dilution aseptically and discharge into 9 ml sterile water blank and shaked again. This makes a dilution of 10⁻². Dilutions were made up to 10⁻⁷ followed by this procedure. Then 1 ml was taken from dilutions 10⁻⁴, 10⁻⁵, 10⁻⁶ and 10⁻⁷ and pour in sterilized Petri dishes with three replicates in each dilution. Each Petri dish contained about 15 ml of soil extract agar medium, melted previously and kept in a water bath at 45°C. Quickly rotate the Petridish so as to mix the medium with the inoculum. Allow the agar to solidify. The plates were incubated inverted position for 5-7 days at 30°C. The number of bacteria was calculated in per gram of dry vermicompost and expressed in colony forming unit (CFU g-¹vermicompost) and finally data were transferred in Log value as descrived by Cappuccino and Sherman (1999).

Determination of phosphate solubilizing bacteria (PSB) in prepared phosphor-vermicompost: PSB populations were counted from the mature vermicompost at the end of the incubation. The samples were collected from each pot. The samples were processed immediately after collection, otherwise stored in plastic bags at 4 °C until processed. PSB were counted by serial dilution plate technique (Subba Rao, 1999) as like as above total bacterial count. One gram of vermicompost diluted in 9 mL sterile water blank which gave 10⁻¹ dilution. Then 10⁻² to 10⁻⁶ dilutions were spread on Pikovskaya's solid medium containing tricalcium phosphate (TCP) (Pikovskaya, 1948). The colonies that appeared as discrete clear zones (halo zones) around them indicated the dissolution of TCP was assumed to be phosphate solubilizers and the PSB colonies were counted. PSB populations were expressed in the colony forming unit (CFUg-1 vermicompost) as described by Cappuccino and

Sherman (1999) and finally data were transferred in value.

Assay of phosphatase activity in prepared phosphovermicompost: The phosphatases are mainly of two types such as acid and alkaline phosphatases. Acid phosphatases exist in acidic condition while alkaline phosphatases exist in alkaline condition. All the vermicompost were exist in alkaline condition. Therefore, alkaline phosphatase activity (ALPA) were assayed from the vermicpmpost. Phosphatase activities were assayed according to the methods as described by Tabatabai and Bremner (1969). The activity of phosphatase enzyme was assayed by colorimetric method using p-nitrophenyl-phosphate (pNPP) as substrate. One gram of vermicompost was taken in a 30 mL test tube and 0.2 mL of tolune, 4 mL of modified universal buffer (MUB), 1 mL of pnitrophenyl-phosphate solution were added and mixed the contents by shaking. All the test tubes were tightly closed by parafilm and incubated at 37°C for one hour then turned the suspension yellow in colour. After incubation all the test tubes were brought out and removed the parafilm. 1 mL of 0.5M CaCl₂ and 4 mL of 0.5 M NaOH were added into the test tube to stop the reaction. All the test tubes were swirled for few seconds and filtered the suspension through a Whatman No.1 filter paper. Absorbance of yellow coloured filtrate was taken at 420 nm in spectrophotometer. Finally, ALPA was calculated by a reference to calibrate with the standard p-nitrophenol (pNP) and the results were expressed in terms of product of pNP formed g⁻¹ vermicompost h⁻¹.

Statistical analysis: All data obtained were analyzed and processed using one-way ANOVA (Analysis of variance) with significance level of 0.05 through Statistix 10 software packages (Analytical Software, 2105 Miller Landing Rd, Tallahassee, FL 32312).

Results and Discussion

Effects of different treatments on pH, organic carbon and nutrient contents in phospho-vermicompost: The

pH, organic carbon and nutrient contents (N, P, K and S) of prepared phospho-vermicompost with or without rock phosphate amended are presented in the Table 2. The pH was almost similar in all the treatment combinations after vermicomposting (Table 2) but with rock phosphate amended vermicompost showed greater pH than without rock phosphate amended treatments. Greater pH may be attributed to the decomposition of nitrogenous substrates resulting in the production of ammonia which forms a large proportion of the nitrogenous matter was excreted by earthworms (Champar-Ngam *et al.*, 2013). The present results are supported with those earlier findings.

After vermicomposting all the treatments showed lower organic carbon (Table 2) than its parent materials (Table 1) might be due to the action of earthworms and microorganisms harbored into the earthworms guts. However, the lowest organic carbon was recorded in the treatment T₈ where amended with rock phosphate which indicated more decomposition and allowed to stand more stable phospho-vermicompost. Albanell *et. al.*(1988) reported that chemical analysis showed vermicompost had a lower organic carbon. The present findings are well supported by those findings (Garg *et al.*, 2005).

Among the treatments, the treatments T_8 gave the highest total N (1.42%), P (1.45%), K (1.52%) and S (0.35%) contents (Table 2) than that of other treatments which indicated the better quality of phosphovermicompost. Maximum available P (Figure 1) was also found with the treatment T_8 and the lowest available P was observed with the treatment T_1 . Microorganisms and earthworms are important biological organisms helping nature to maintain nutrient flows from one system to another (Lazcano *et al.*, 2018; Singh and Amberger, 1990). Hashemimajd *et al.* (2004) reported that higher amounts of nitrogen, potassium and total phosphorous and micronutrients were recorded in the vermicompost compared to the parent materials.

Table 2. pH, organic carbon	O.C) and total N, P, K and S contents in with or without rock phosphate amended
vermicompost.	

Treatments	рН	%OC	%N	%P	%K	%S
T ₁	7.73±0.1	16.2±0.1	1.1±0.1	0.45±0.05	1.4±0.2	0.28±0.06
T ₂	7.95±0.05	16.4±0.2	1.23±0.08	0.45±0.02	1.80±0.3	0.30±0.02
T ₃	7.82±0.12	17.3±0.17	1.17±0.05	0.52±0.02	1.70±0.05	0.34±0.01
T ₄	7.9±0.1	16.8±0.4	1.2±0.05	0.50±0.03	1.75±0.08	0.34±0.05
T ₅	8.05±0.15	15.5±0.5	1.23±0.1	1.2±0.1	1.35±0.07	0.32±0.04
T ₆	8.13±0.13	15.1±0.3	1.35±0.04	1.37±0.05	1.4±0.06	0.30±0.07
T ₇	8.10±0.2	15.9±0.1	1.37±0.03	1.38±0.04	1.45±0.12	0.33±0.11
T ₈	8.15±0.15	15.2±0.36	1.42±0.07	1.45±0.06	1.52±0.08	0.35±0.03

± indicates standard deviation; **Note:** T₁: 50% Cowdung (CD)+ 50% Mustard straw (MST), T₂:50% CD +50% Water hyacinth (WH), T₃:50% CD+ 50% Rice straw (RST), T₄: 50% CD +25% WH + 25% RST, T₅:50% CD+50% MST+ 4% Rock phosphate (RP), T₆:50% CD +50% WH+ 4% RP, T₇: 50% CD+ 50% RST+ 4% RP and T₈: 50% CD +25% WH + 25% RST + 4% RP.

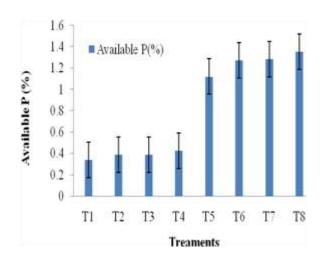


Figure 1. Available P contents with or without rock phosphate amended vermicompost (Verticalbars indicate standard errors).

Note: T₁: 50% Cowdung (CD)+ 50% Mustard straw (MST), T₂:50% CD +50% Water hyacinth (WH), T₃:50% CD+ 50% Rice straw (RST), T₄: 50% CD +25% WH + 25% RST, T₅:50% CD+50% MST+ 4% Rock phosphate (RP), T₆:50% CD +50% WH+ 4% RP, T₇: 50% CD+ 50% RST+ 4% RP and T₈: 50% CD +25% WH + 25% RST + 4% RP.

Therefore, the present study claimed that 50% cowdung +25% water hyacinth +25% rice straw with 4% rock phosphate and earthworms (T_8) could be used for the production of phosphate rich vermicompost i. e. Phospho-vermicompost. Yan *et al.* (2012) reported that the extractable P was 17% higher in vermicompost with the addition of rock phosphate. In addition, extractable macronutrients N and K were also found to be significantly higher in vermi-composting with the addition of rock phosphate. The present findings are well agreement with those findings.

Effects of different treatments on earthworms, total bacteria and PSB population and alkaline phosphatase activity in prepared phosphovermicompost: The treatment T₈ showed significantly highest population of earthworms, total bacteria, phosphate solubilizing bacteria (PSB) and alkaline phosphatase activity (ALPA) followed by the treatments T₆ and the lowest was observed in the treatment T₁ in prepared phospho-vermicompost. The results indicated that organic residues amended with rock phosphate increased all the recorded biological indices in mature vermicompost than without rock phosphate amended treatments (Table 3). The interaction between earthworms and microorganisms were non-thermophilic biodegradation of organic wastes (Arancon *et al.*, 2004). Several findings showed considerable increase in total viable counts of bacteria in the worm treated compost (Haritha Devi *et al.* 2009).

Suhane (2007) reported that total bacterial counts exceeded 10¹⁰/g of vermicompost and it included nitrobacter, azotobacter, rhizobium, phosphate solubilizers and actinomycetes. The increase of microbial population may be due to the congenial condition for the growth of microbes within the digestive tract of earthworms which provide energy

and also act as a substrate for the growth of microorganisms (Tiwari *et al.* 1989). The present results are well supported with those findings. Maximum PSB population and phosphatase activity were recorded in the treatment T₈ which might be enhanced to increase the available P in the rockphosphate amended treatments than nonamended treatments. The present results are well supported by earlier findings that vermicomposting increased P bioavailability, microorganisms and earthworms; including organic acid and phosphatases production which solubilizes inorganic P (Scervino *et al.*, 2010; Saha *et al.*, 2008; Champar-Ngam *et al.*, 2013).

Table 3. Effects of different treatments on number of earthworms, total bacteria and phoshate solubilizing bacteria (PSB) and alkaline phosphatase activity (ALPA) in prepared vermicompost (VC).

Treatments	Earthworms pot ⁻¹ (no.)	Total bacteria (Log CFU g ⁻¹ VC.)	PSB (Log CFU g ⁻¹ VC)	ALPA (μg þNPg ⁻¹ VC
T_1	919c	10.2d	3.3d	20.0d
T_2	1134d	11.6b	3.6d	27.3cd
T_3	938e	10.5cd	3.3d	30.3c
T_4	1123d	11.0c	3.7d	35.0c
T ₅	1383bc	10.9с	4.6c	73.0d
T_6	1519ab	12.7a	6.7a	91.0a
T_7	1333c	12.6a	6.1b	79.0b
T_8	1619a	12.8a	6.9a	97.2a
% CV	7.05	6.5	5.23	8.69

In a column, the values having same letter do not differ significantly at 5% level of probability as per LSD; **Note:** T_1 : 50% Cowdung (CD)+ 50% Mustard straw (MST), T_2 :50% CD +50% Water hyacinth (WH), T_3 :50% CD+ 50% Rice straw (RST), T_4 : 50% CD +25% WH + 25% RST, T_5 :50% CD+50% MST+ 4% Rock phosphate (RP), T_6 :50% CD +50% WH+ 4% RP, T_7 : 50% CD+ 50% RST+ 4% RP and T_8 : 50% CD +25% WH + 25% RST + 4% RP.

Effects of different treatments on C:N and C:P ratio in prepared phosphor-vermicompost: After vermicomposting all the combination of organic residues showed lower C:N and C:P ratio (Fig.2) than its original materials. But both of C:N and C:P ratio

were more decreased when rock phosphate were added to the residues (Figure 2) resulted higher contents (Table 2) of total nutrients (N, P, K and S) including available P (Fig. 1) in the mature phophovermicompost. However, lowest C:N and C:P ratio

were observed with the treatment T₈ which indicated more maturity occurred in the phopho-vermicompost. C:N and C:P ratio are good indicator for the degree of decomposition and mineralization of nutrients. During the process of biooxidation, CO₂ and N is lost and loss of N takes place at a comparatively lower rate and vermicompost showed significantly less C:N ratio as they underwent intense decomposition (Lazcano *et al.* 2008). Atiyeh *et al.* (2000) also reported that earthworm activity reduced C:N ratio in manure. The results of the present study are well supported with those findings.

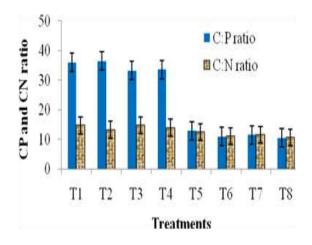


Figure 2. Effect of different treatments on C:N and C:P ratio in mature vermicompost

Note: T_1 : 50% Cowdung (CD)+ 50% Mustard straw (MST), T_2 :50% CD +50% Water hyacinth (WH), T_3 :50% CD+ 50% Rice straw (RST), T_4 : 50% CD +25% WH + 25% RST, T_5 :50% CD+50% MST+ 4% Rock phosphate (RP), T_6 :50% CD +50% WH+ 4% RP, T_7 : 50% CD+ 50% RST+ 4% RP and T_8 : 50% CD +25% WH + 25% RST + 4% RP.

The results indicated that the phospho-vermicompost could be produced through the bioconversion of earthworms by using of 25% cowdung, 25% water hyacinth, 25% rice straw enriched with 4% rock phosphate. Prepared phospho-vermicompost contained higher amount of total N, P, K and S and available P which can be applied as organic fertilizer for the supplement of nutritional demand of the crops and

could be saved inorganic fertilizer in the cultivation of crops. The developed phospho-vermicompost also could be used in the cultivation of different crops for full supplement of phosphatic fertilizers. Kumari and Ushakumari (2002) reported that high and diverse populations of native microorganisms favor biochemical reactions and vermicompost enriched with rock phosphate showed high P bioavailability which might be led to enhance yield and uptake of nutrients in the crops. The present findings are well agreements with those findings.

Conclusion

From the results, it can be concluded that 50% cowdung +25% water hyacinth +25% rice straw + 4% rock phosphate powder with red wiggler earthworms (*Eisenia foetida*) could be used for the production of phospho- vermicompost which may use as organic fertilizer for the supplement of phosphorus and other plant nutrients (N, K, S etc.) for the production of different crops and could be reduced the usage of chemical fertilizers.

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