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Assessment of Phosphorous Release from Bat Guano with Respect

to Their use as Organic Fertilizers in Crop Production

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ABSTRACT

Bat guano from Kisarawe cave A (BGK-A), Kisarawe cave B (BGK-B) and Sukumawera cave (BGS) in Tanzania were studied in a laboratory incubation experiment for 112 days to assess the phosphorus release patterns and establish the pick periods of P mineralization. Total P contents from BGK-A, BGK-B and BGS were 8.55, 7.03 and 3.45 % respectively. Bat guano from each deposit mixed with soil at varying rates of 0, 10, 20, 40 and 80 mg P 200 g⁻¹ soil. The experiment was arranged as 3×5 factorial in randomized complete design (RCD) with three replications. Results showed a gradual increase of P availability from 28 days to 84 days of incubation; followed by a gradual decrease up to the 112th day of incubation. P release (availability) from the studied guano was in the order of BGK-B < BGS < BGK-A. BGK-A indicated higher potential as an alternative P source for agricultural application. P-source and application rate had a significant interaction effect (P < 0.05) on P release at all incubation intervals when P was applied at a rate of 80mg kg⁻¹ soil. The soil pH showed very strong negative correlation (r > 0.8) with P released in the soil throughout incubation period. It was concluded that P release from the three bat guano deposits is gradual and reaches the pick in almost three months after application. Thus, guano should be applied at least two months before planting for more effectiveness of P release.

Keywords: Bat guano, Phosphorus release, Incubation, soil pH.

1.0 INTRODUCTION

Phosphorus (P) is an essential plant nutrient required for optimum crop growth [1]. It is the second most limiting plant nutrient after nitrogen (N) in crop production [2]. Phosphorus plays vital roles in almost all plant processes including energy transfer, phosphate held as a part of the chemical structures of adenosine diphosphate (ADP) and adenosine triphosphate (ATP). It is the source of energy that drives large number of chemical reactions within the plant including phosphorylation process [3]. In photosynthesis, P utilizes light energy in the presence of chlorophyll to combine carbon dioxide and water into simple sugars, with the energy being captured in form of ATP which is available as an energy source for the many other reactions that occur within the plant. Through genetic transfer, P is a vital component of the substances that are building blocks of genes and chromosomes which involve in carrying genetic code from one generation to the next [4]. Also P involves in transportation of nutrients through the plant cells, movement of nutrients within the plant through cell membranes depends largely on energy whereby P in form of ATP compounds provide that energy [4]. In cereal crops such as rice, P increases panicle numbers, seed-setting rate and grain weight [5, 6]. Moreover, it stimulates tillering, and often hastens maturity [7, 8]. It improves plant health and reduces the incidence and severity of many fungal diseases such as powdery mildew [3, 9, 10].

Most of the tropical soils including Tanzanian soils are low in plant available phosphorus (P) [11]. The low level of plant available P in the tropical soils is due to either low levels of P in soil parent materials, highly weathered soils with high P fixing capacities or inadequate amounts of applied P fertilizer to replenish P losses through crop removal and other pathways [12]. Furthermore, industrial fertilizers are too limited in access and expensive for most

smallholder farmers. This justifies the need for exploiting new and affordable fertilizers such as bat guano which could improve soil fertility and crop production while minimizing environmental pollution risks associated with misuse of chemical fertilizers. Guano deposits are found in a number of places in Tanzania and are continuously being deposited by bats and thus, they are renewable. Most research works have been conducted and reported about phosphorous and other nutrients release patterns of different manures other than bat guano [13-17]. In Tanzania, little information is known on the nutrients release patterns of guano when used as soil amendment and source of plant nutrients. Furthermore, the recommended rates of guano found in Tanzania for agriculture are not yet known. The objective of this study was to assess the phosphorous release from bat guano with respect to their use as organic fertilizers in crop production.

2.0 MATERIALS AND METHODS

2.1. Geological settings of guano deposits

2.1.1. Kisarawe guano deposits

Kisarawe guano deposits are found in Kisarawe district, 25 km west of Dar-es-salaam City (Fig.1). The site is located at $38^{\circ} 78 \text{ E} / 7^{\circ} 20 \text{ S}$. Formally the caves were used for mining kaolinite mineral in nineteen fifties to early nineteen seventies when they were abandoned and nowadays the caves are hosting bat colonies. The parent material of the caves is kaolinitic sandstone. The sandstone hosting bat guano is surrounded to the north and western side by clay bound gravels and to the eastern side by superficial white buff sands (Fig.1).



Figure 1: Geological setting of Kisarawe guano deposits

2.1.2 Sukumawera guano deposit

Sukumawera guano deposit is found at Maji moto sub village, near Songwe river, in Mbozi district. The site is located at $33^{\circ} 22$ E / $8^{\circ}89$ S. The cave is made of travertine (Carbonate rock) within the parent rock which is mainly dominated by Precambrian garnet biotite gneiss (Fig. 2).



Fig.2. Geological setting of Sukumawera guano deposit

2.2 Sampling and analysis of bat guano

Three composite samples of guano were randomly collected from Kisarawe caves (A and B) and Sukumawera guano deposits. The samples were air-dried in a glasshouse, ground and sieved to pass through a 0.5 mm sieve, labeled and packed in bags and transported for determination of all total elemental composition at the Geological Survey of Tanzania (GST) laboratory by using X-ray Fluorescence (XRF01) pressed powder without binder. The analysis was done by using *Maniple Analytical Software*. While sub samples sieved through a 2 mm sieve and transported to the Soil and Geological Sciences laboratory at Sokoine University of Agriculture (SUA), in Morogoro Tanzania for determination of physical properties including moisture content (MC) and loss on ignition (LOI). Chemical properties determined were pH, Electrical conductivity (EC), organic carbon (OC) and total N as described by [18, 19].

2.3 Soil sampling and analysis

A bulk soil sample was collected at 0-20 cm depth from the Soil and Geological Sciences experimental field, using the random sampling method in two diagonals. The soil was air dried, crushed and sieved through a 2 mm sieve for physical and chemical analysis. Particle size distribution was determined by the hydrometer method after dispersing soil sample in sodium hexametaphosphate solution [20]. Thereafter, the corresponding soil textural class was determined by using the USDA textural class triangle [21]. The pH of the soil was determined using a glass electrode pH meter in 1:2.5 (soil: water suspension) [22]. Electrical conductivity was measured in 1:2.5 (soil: water) by using conductivity meter [22]. Organic carbon was determined by Walkley and Black method using wet oxidation by potassium dichromate [23]. Total N was determined by the micro-Kjeldahl digestion procedure followed by distillation [18]. Available P was extracted following the procedure described by [24] and determined by using neutral ammonium-acetate saturation method (NH₄OAc, pH 7) followed by Kjeldahl distillation method. Exchangeable K, Ca, Mg and Na were determined from the ammonium-acetate filtrates by Atomic Absorption Spectrophotometer [20].

2.4 Experimental design

The experiment was designed as a 3×5 factorial experiment laid down in a Completely Randomized Design (CRD) replicated three times. It consisted of two factors which were three P-sources from Kisarawe A, Kisarawe B and Sukumawera deposits at five different levels of application (Table 1). Bat guano was mixed thoroughly with soil, and

the mixture was incubated in plastic containers covered with aluminum foil at the top to avoid contamination and covered with cotton wool at the bottom to avoid leaching. It was placed on the laboratory bench at 25^{\pm} 1°C for 112 days. Incubation moisture was maintained around 50% of field capacity by adding a predetermined amount of distilled water.

P rate	Level of P applied from each source (mg P 200 g soil ⁻¹)			
(mgP kg ⁻¹)	BGK-A	BGK-B	BGS	
P_0	0	0	0	
P_{10}	0.023	0.028	0.06	
P ₂₀	0.046	0.056	0.12	
P_{40}	0.092	0.112	0.24	
P_{80}	0.184	0.224	0.48	

Table 1: P rates used in an incubation experiment

2.5 Data collection

After every 28 days, 5 g sub-sample of the incubated mixture was collected by using sterilized spatula cleaned using a cotton wool between sampling events. The samples were analyzed for pH in a 1:2.5 (soil: water) suspension [22] and P was extracted by the Olsen extraction method [24], followed by quantification of extractable P using ascorbic acid colorimetric method [25].

2.6 Statistical analysis

Analysis of variance was performed on extractable P determined at different intervals of incubation using GenStat Discovery 15thedition Software. Treatment means were separated using Duncan New Multiple Range Test at the 5% of probability level.

3.0 RESULTS AND DISCUSSION

3.1 Physico-chemical properties of the soil used in the incubation experiment

The physico-chemical properties of the soil used in the incubation study are as presented in Table 2. According to [26], the soil was slightly alkaline with pH of 7.2. Most plant nutrients are optimally available for plants between 6.5 to 7.5 pH [27]. [26] revealed that when the pH is low, P can be fixed by Al, Fe and Mn and at higher pH values above 7.5 P is precipitated by calcium and become insoluble. The pH of experimental soil was therefore within a range that guaranteed availability of P released from guano.

The electrical conductivity (EC) of the soil was 561 mS/cm. According to [28] the level of EC is categorized as normal. [29] reported that under anaerobic condition EC has a tendency of showing positive correlation with phosphorus concentrations. The texture of the soil was sand clay. Sandy clay soils are likely to influence the availability of P which is mineralized after amend the soil with bat guano. [30] and [31] reported that soil texture affects the availability of P by influencing soil organic matter (SOM) accumulation through mineralization and immobilization of SOM by microbial activity. The organic carbon (OC) of the soil was rated as very high while total N was medium [26]. Olsen extractable P was relatively higher than the critical value for deficiency (4.25 mg P kg⁻¹) as reported by [32]. Moreover, [28] reported that Olsen extractable P higher than 6 mg P kg⁻¹ is regarded as medium.

Table 2. Chemical and physical properties of son conected for incubation experiment					
Parameter	SI-unit	Value	Rating [28][26]		
pH (H ₂ O)		7.2	Normal		
EC	mS/cm	561	Normal		
OC	%	11.05	Very High		
Total N	%	0.48	Medium		
Olsen Extractabl P	mg/Kg	6.59	Medium		
Exch.Calcium	Cmol (+)Kg ⁻¹	7.97	Medium		
Exch.Sodium	cmol (+) Kg ⁻¹	3.98	Very High		
Exch. Magnesium	cmol (+) Kg ⁻¹	5.12	High		
Exch. Potassium	cmol (+) Kg ⁻¹	6.07	Very High		
CEC	cmol (+) Kg ⁻¹	20.2	Medium		

 Table 2: Chemical and physical properties of soil collected for incubation experiment

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Particle size			
Sand	%	49.3	
Clay	%	41.1	
Silt	%	9.6	
Textural class			Sand Clay

3.1 Total elemental compositions of Kisarawe and Sukumawera bat guano

Table 3 shows the result of the total elemental composition of Kisarawe and Sukumawera bat guano used in this study. The results showed relatively higher total P (8.55%) from BGK-A than BGK-B (7.03%) and BGS (3.45%). [33] reported total P content of Kisarawe bat guano in the range of 7 - 15% P. Higher P concentration in Kisarawe caves A and B might be due to high accumulation of daily fresh bat faeces [35]. Total P content determined in Sukumawera guano (3.45%) was lower and different from total P content of 20 - 41% previously reported by [33] using guano from the same deposit. Such differences might have been caused by decomposition of bat guano followed by gradual leaching of nutrients including P [35, 36, 37].

Element	Guano type	and total composition (%)	
	BGK-A	BGK-B	BGS
Al	5.82	11.11	Trace
Si	4.21	10.11	2.34
Р	8.55	7.03	3.45
S	7.49	4.25	2.87
K	11.04	6.34	3.98
Ca	6.70	4.92	45.67
Ti	0.36	1.98	0.35
Mn	7.36	1.40	0.31
Fe	2.71	4.41	4.70
Zn	1.25	0.86	0.38
Sr	0.09	0.10	1.23
Pd	0.11	0.14	0.05
Cu	0.04	0.05	0.03
Pb	< 0.01	0.01	< 0.01
Zr	0.01	0.03	0.01
Sr	0.01	0.01	0.21
Rh	0.062	0.07	0.03

Table 3: Total elemental compositions of Kisarawe and Sukumawera bat guano

Potassium (K) is among the major nutrient elements required by plants. Total K in guano from Kisarawe cave A was found to be higher (11.4%) compared to total K obtained from Kisarawe cave B (6.3%) and Sukumawera cave (4.0%). The differences could be attributed to variations in the nature and composition of parent material around the caves and that of food consumed by bats. Generally, total P and K contents of guano from Kisarawe caves A and B were above 5% recommended by FAO as a minimum for any material to qualify as a fertilizer the rate at which the nutrients are released from the guano would still influence its potential for use as P source in crop production. Other nutrient elements contained in guano include calcium which was higher in guano from Sukumawera deposit (45%) than the other two deposits mainly because of high Ca contents in parent material of Sukumawera cave. The parent material of Sukumawera cave is mainly composed of gneiss and travertine rocks which contain amphibole, pyroxene and calcite minerals [38].

Total Manganese in the three bat guano was low, similar to the findings reported by [33]. Furthermore, total S in the three bat guano was also low. The data reported by [33] was contrary to these findings. [33] reported higher total S in Kisarawe caves than values observed in this study probably due to variation of total S contents between sampling points and time intervals as influenced by various factors. Apart from Si and Al which were relatively higher in

Kisarawe cave A and B deposits, heavy metals (Pb, Pd, Rh,Ti, Sr, and Zr) were found in relatively smaller amounts in the three deposits suggesting that application of guano as organic fertilizer is safe and can't be associated with high risks of Potential toxic elements (PTEs) accumulation in the soil environment.

3.2 Physico-chemical properties of guano used in the experiment

Table 4 shows some physico-chemical properties of bat guano used in this study. The physical chemical properties assessed include pH, electrical conductivity (EC), moisture content (MC), loss on ignition (LOI), organic carbon (OC) and Total nitrogen. The pH of Kisarawe bat guano from caves A and B (BGK-A and BGK-B) was quite different. Guano from cave A was slightly acidic with a pH of 6.1, while pH of guano from cave B (4.3) was extremely acid [26, 28]. Extremely low pH of BGK-B coupled with high levels of Al and Si are likely to restrict P release for plant uptake from this guano because solubility (activity) of both Al and Si which have high affinity for P increases with decreasing pH hence increasing P fixing sites in the soil. The pH of guano from caves A and B were different although these guanos are from the same geographical location but found in different caves. Their differences are possibly caused by high amount of organic carbon 27.95% in cave B compared to 21.45% in cave A. [39] reported that soil which has high organic matter which has a relation and estimated by the determination of soil organic carbon (SOC) has greater capacity of holding hydrogen ions; therefore it has high ability of reserving acidity. Moreover, [27] explained that cation and anion exchange capacity are largely determined by the charges of the SOM which are influenced by pH. High amounts of organic matter typically have higher cation exchange capacity (CEC), thus able to bind more cations such as calcium or potassium, aluminum and hydrogen ions which have a tendency of lowering pH. In addition pH differences may be contributed by the nature of rocks and rock forming minerals in and around the caves.

Parameter		Guano type	
	BGK-A	BGK-B	BGS
pH (water)	6.1	4.3	6.3
ECe (mS cm ^{-1})	17.77	3.28	28.53
OC (%)	21.45	27.95	35.10
MC (%)	14.55	19.71	21.69
LOI (%)	26.23	7.7	15.88
Total N (%)	4.08	5.51	5.57

Table 1: The physico-chemical properties of Kisarawe and Sukumawera bat guano

3.2 Effect of incubation time on P release

The amounts of P released and determined as Olsen extractable P during incubation for 112 days are depicted in Fig. 3. P releases were generally in three regions; the first region was fast release of P and was immediately after day 0 to day 56 of incubation. The second region was slow release which was from 56 to day 84 days of incubation. The third region in which P release was decreasing started from day 84 to day 112. These results were in line with findings reported by [40] and [41]. Both studies reported increase in available P followed by decrease in the release of P. Studies of [42] and [43] showed that as the time of incubation increases the P availability in soil decreases for both organic and inorganic P sources due to rapid microbial immobilization of the added sources. The trend can also be due to fixation of released P by both soil and guano components such as Al, Fe and Si with high affinity for P. Although BGK-B had higher total P than BGS, BGK-B released less amount of P at all sampling intervals further suggesting that elevated levels of Al and Si in BGK-B restricted P release or fixed P after release from the guano making it less available for plant uptake.



Figure 3.P release from different guano at varying incubation intervals.

3.3 Effect of P- rates and interaction between P-sources and rates on P release

The P release significantly increased with increasing application rates of all P- sources (Table 5). Application of guano at a rate of 80 mg kg⁻¹ resulted into the highest level of P release from all P sources. These findings are similar to those of [41]. They reported that phosphate release increase significantly with increase in initial phosphorus levels and decreased with increasing incubation period. The P released was significantly (P < 0.05) higher for BGK-A followed by BGS and BGK-B (Fig. 3).

P-rates (mg kg ⁻¹)	Days of incubation				
	0	28	56	84	112
P0	5.02 a	4.98 a	5.21 a	5.6 a	5.45 a
P10	6.99 b	10.26 b	13.43 b	14.85 b	11.17 b
P20	7.85 c	10.92 c	14.59 bc	16.58 c	12.94 c
P40	8.77 d	12.01 d	15.40 c	18.55 d	14.31 d
P80	9.40 e	14.16 e	17.55 d	20.22 e	15.60 e
Mean	7.61	10.46	13.24	15.16	11.89
CV %	4.8	4.5	11.4	4.2	5.7
F-pro	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

Table 2: Effect of P-rates on P release at varying incubation intervals

Means in the same column followed by the same letter(s) are not significantly different at 5% level of significance according to Duncan New Multiple Range Test. Means in each column analyzed separately.

Furthermore, there was highly significant (P < 0.05) interaction effect between P-sources and rates on P released in soil following incubation (Table 6). The overall mean results showed significantly higher P release (P < 0.05) from BGK-A and BGS at the rate of 40 and 80 mg kg⁻¹ as compared to P released from equivalent rates of BGK-B. These differences were due to variations in their pH, Al and probably Si contents. BGK-A and BGS were slightly acidic while that of BGK-B was strongly acidic with higher total Al and Si contents (Table 4). According to [44] fertilizers with lower pH prevent P availability in soil due to P fixation.

P-Source Rates		Days of incubation				
(шу ку	;)	0	28	56	84	112
BGK-A	0	5.28 ab	5.18 a	5.79 a	5.50 ab	5.19 a
BGK-A	10	8.82 f	13.68 f	17.93 e	20.47 g	16.31 fg
BGK-A	20	9.30 fg	14.14 f	18.81 e	22.02 h	17.44 g
BGK-A	40	9.61 gh	14.48 f	19.13 e	23.69 i	18.66 h
BGK-A	80	10.09 h	15.36 g	20.54 e	25.41 j	19.28 h
BGK-B	0	4.65 a	4.76 a	4.74 a	6.23 b	6.15 a
BGK-B	10	5.59 b	8.21 b	10.69 b	11.35 c	7.70 b
BGK-B	20	6.38 c	9.01 bc	11.18 bc	13.07 d	9.59 c
BGK-B	40	7.25 e	10.40 d	12.18 bcd	12.73 d	10.79 d
BGK-B	80	8.12 e	11.78 e	13.10 bcd	14.66 e	11.88 d
BGS	0	5.12 ab	5.01 a	5.11 a	5.07 a	5.02 a
BGS	10	6.54 c	8.88 bc	11.66 bc	12.73 d	9.50 c
BGS	20	6.54 c	9.60 c	13.79 cd	14.66 e	11.80 d
BGS	40	7.88 e	11.14de	14.90 d	17.33 f	13.47 e
BGS	80	10.01 h	15.32 g	19.01 e	20.30 g	15.65 f
Mean	ı	7.61	10.46	13.24	15.16	11.89
CV %	,)	4.8	4.5	11.4	4.2	5.7
F-pro)	< 0.001	< 0.001	0.003	< 0.001	< 0.001

Table 3: Interaction effect of P-sources and rates on P released at varying incubation intervals

Means in the same column followed by the same letter(s) are not significantly different at 5% level of significance according to Duncan New Multiple Range Test. Means in each column analyzed separately.

3.4 Effect of pH on P release

The pH of the soil was correlated with P release from 0 to 112 days of incubation. The results showed a very strong negative correlation between soil pH and P released (r > 0.8) as shown in figures 4- 6. This implies that as the time goes on, the P release was decreasing in the meantime the soil pH was increased. As P is higher the pH become lower and vice versa. The results are in agreement with the findings of [45] who reported negative correlation between available P and pH values of soil. In BGK-A the correlation was highest in day 112 (r = -0.982) and lowest in day 56 (r = -0.928) (Fig. 4 e, c). In BGK-B the correlation was highest in day 0 (r = -0.985) and lowest during day 56 (r = -0.884) (Fig. 5 a, c). Similarly in BGS, result showed that correlation was highest in day 0 (r = -0.998) and lowest in day 56 (r = -0.943) (Fig. 6 a, c). These results showed that as the soil pH decrease in a range of 5.5-6.5 the P availability increases. Below the soil pH of 5.5, the release of P in soil become difficult because P reacts with iron and aluminum to form insoluble compounds that makes it unavailable to plants. These results are in line with observations made by [46]. The results also agreed with findings attained by [47]. [47] reported that phosphorus availability is higher in moderately acidic soil. Moreover, in this study, the pH decreased as the rate of guano increases probably because of acidic nature of the guano. This finding is supported by [48] who reported the addition of organic resources to soils may result in pH changes due to microbial activity during residue breakdown. Also, [45] reported that application of phosphorus fertilizers can cause the decrease of the pH values in soils.



Figure 4: Correlation of P released from BGK-A at 0, 10, 20, 40 and 80 mg P kg⁻¹ with pH at (a) day 0, (b) day 28, (c) day 56, (d) day 84and (e) day112 of incubation



Figure 5: Correlation of P released from BGK-B at 0, 10, 20, 40 and 80 mg P kg⁻¹ with pH at (a) day 0, (b) day 28, (c) day 56, (d) day 84 and (e) day112 of incubation



Figure6: Correlation of P released from BGS at 0, 10, 20, 40 and 80 mg P kg⁻¹ with pH at (a) day 0, (b) day 28, (c) day 56, (d) day 84 and (e) day112 of incubation

CONCLUSIONS AND RECOMMENDATIONS

Guano from Kisarawe cave A (BGK-A) decomposed more rapidly in the soil and released more available P than guano from Kisarawe cave B (BGK-B) and that from Sukumawera (BGS). In all three P sources, application of guano at a rate of 80 mg P kg⁻¹ of soil resulted into the highest level of P release at 84 days of incubation but increasing the duration of incubation beyond 84 days resulted into a gradual decrease in P release to 112 days. Furthermore the study found a negative correlation between P release from guano and pH of soil. These results implied that P released from guano could be much more available for plant uptake when applied three months earlier before planting especially for annual crops. However, field trials are recommended to validate the results of incubation study.

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