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Mineralization of phosphorus and sulfur in concentrated organic fertilizer under anaerobic laboratory conditions

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Abstract

Understanding nutrient release from manure or compost through mineralization is essential for meeting crop nutritional requirements, ensuring timely fertilizer application, and improving nutrient use efficiency. This study investigated the mineralization of phosphorous (P) and sulfur (S) in concentrated organic manures under anaerobic laboratory conditions. Treatments included T₁= Mustard oil Cake, T₂= Fish meal, T₃= Bone meal, T₄= Biochar enriches organic fertilizer, T₅= Vermicompost and T₆= control. Physicochemical properties such as available phosphorus (P), available S, pH, and electrical conductivity (EC) were assessed using standard protocols. The mineralization process was monitored for 180 days at 25°C. Results showed significant variations in P, S, pH, and EC under anaerobic conditions. Mustard Oil Cake exhibited the highest release of S, P, pH, and EC, followed by Fish Meal in most cases. Variation in pH release was observed among treatments throughout the study period. Appropriate selection and application of organic manures in correct quantities are crucial for providing essential nutrients, enhancing crop nutrient use efficiency, and formulating accurate fertilizer recommendations to maximize the benefits of organic manures.

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Introduction

Agriculture significantly impacts Bangladesh's economy, contributing 15% to GDP and employing 43% of the workforce (Bangladesh Economic Review, 2023; Statistical Yearbook Bangladesh, 2023). Technological advancements, mechanization, increased chemical use, higher cropping intensity, and adoption of high-yielding and hybrid varieties have boosted crop production. However, these changes have led to adverse effects such as topsoil depletion, nutrient extraction, water contamination, poor living and working conditions for laborers, rising production costs, and declining rural economic and social conditions (Agriculture Sector Review, 2023).

Soil, being fragile and dynamic, needs regular maintenance and nourishment for sustained productivity and stability. Relying solely on chemical fertilizers is insufficient to prevent the depletion of organic matter and nutrient mining. Incorporating organic sources such as cow dung, chicken manure, bioslurry, compost, and green manure is essential (Prado *et al.*, 2022; Ding *et al.*, 2021; Vanotti *et al.*, 2020; Hammerschmiedt *et al.*, 2021). Organic manures improve soil organic matter, enhance physical and chemical properties, boost microbial activity, and reduce metal toxicity (Escobar and Hue, 2008). Soil organic matter mineralization releases significant amounts of nitrogen, phosphorus, and sulfur, along with some micronutrients (Rahman *et al.*, 2013).

To enhance soil fertility and optimize crop yield, a systematic approach using both organic and inorganic fertilizers is essential. Effective management of organic waste materials is crucial for sustainable farming. Concentrated organic manures, rich in nutrients like sulfur (S) and phosphorus (P), show promise, but research on S and P mineralization in anaerobic conditions is limited. Understanding the mechanisms controlling nutrient release and availability is vital for improving nutrient management strategies and mitigating environmental impacts (Niyungeko *et al.*, 2020; Lisowska *et al.*, 2022).

Anaerobic conditions, characterized by limited oxygen, are common in agricultural settings like waterlogged soils and anaerobic digesters. In these environments, anaerobic microbial communities break down organic matter, releasing soluble forms of sulfur and phosphorus through microbial activity and organic matter decomposition (Kacprzak *et al.*, 2023; Rigby *et al.*, 2023).

The mineralization of soil organic matter gradually releases significant macronutrients, reducing nutrient loss and enhancing plant uptake. Manures consistently release nutrients over time, ensuring prolonged plant availability without substantial depletion. The key biogeochemical process improving soil fertility and crop yield is the mineralization of carbon (C) and nitrogen (N) from manures (Rahman *et al.*, 2013; Cai *et al.*, 2016).

Phosphorus is vital for plant growth and productivity, involved in processes like photosynthesis, respiration, and protein synthesis. Limited phosphorus availability can lead to persistent soil deficiencies, hindering agricultural productivity (De la Fuente *et al.*, 2013; Jensen *et al.*, 2023).

The rate of phosphorus mineralization is influenced by factors like season, climate, soil organic matter, soil depth, and the C/P ratio of organic matter (Jalali *et al.*, 2014). Soil properties such as moisture, organic matter, and clay content significantly affect phosphorus distribution and movement (Xiao *et al.*, 2012). Soils with excessive phosphorus are prone to leaching and runoff (Carpenter and Bennett, 2011).

Sulfur is essential for chlorophyll creation and protein synthesis in plants. Insufficient sulfur leads to adverse metabolic and visual effects. Organic sulfur mineralization, a key source of sulfur for plants, constitutes over 95% of the sulfur in manures, crop wastes, and fertilizers (Reddy *et al.*, 2001; Ghani *et al.*, 2001).

The process of S mineralization is influenced by biological activity, organic compounds, and soil

properties (Islam and Dick, 1998). The release kinetics of phosphorus and sulfur from different manures in various soils, particularly in Bangladesh, are not well-studied. There is insufficient evidence on the conversion of organic nutrient sources in soils under both aerobic and anaerobic conditions (Risberg *et al.*, 2017; Sakadevan *et al.*, 1993).

Researching sulfur and phosphorus mineralization in Bangladesh is crucial due to their essential roles in agriculture, impacting crop yield and quality (Schott *et al.*, 2023; Pagliari *et al.*, 2020). Understanding these processes can optimize fertilizer use, reduce costs, and minimize environmental pollution. It is vital for managing soil nutrient depletion, enhancing soil fertility, and promoting sustainable farming. Improved nutrient management supports food security through stable and increased production. Additionally, this research informs policy decisions on agricultural practices and environmental conservation, ensuring the long-term health of the agricultural sector and ecosystems.

Materials and methods

Soil sample collection and preparation

The soils samples used for this study were collected at a depth of 0–15 cm from the Field Laboratory of Soil Science Department, Hajee Mohammad Danesh Science and Technology University, Dinajpur-5200. The soil sampling site lies between located in between 25°10' and 26°04' north latitudes and in between 88°23' and 89°18' east longitude, and the experimental soil was characterized as Non-Calcareous Grey of Piedmont plain and Tista Floodplain soil under the agro-ecological region of the AEZ. Composite soil samples were transported to the laboratory, air-dried on brown paper, crushed, and sieved through a 2 mm mesh to remove unwanted materials. The sieved soils were stored in polyethylene bags and incubated anaerobically at 25°C for 21 days. Prior to incubation, all soil samples were kept in a dry and cool place.

Analysis of initial soil samples

The initial soil samples were analyzed for physicochemical properties: textural class, bulk

density, cation exchange capacity (CEC), pH, organic carbon (OC), total nitrogen (N), exchangeable potassium (K), available phosphorus (P), and available sulfur (S). Particle size and textural class were determined using the hydrometer method and Marshall's Triangular Coordinates. Bulk density was measured with the core sampler method. Soil pH was assessed in a 1:2.5 soil-water ratio using a glass electrode pH meter. CEC and OC were estimated by the sodium saturation and Walkley-Black methods, respectively. Total N was determined by the semi-micro Kjeldahl method, available P by the Olsen method, exchangeable K by the NH_4OAc extraction method with a flame photometer, and available S by a CaCl_2 extractant solution with a spectrophotometer.

Collection of different manures and determination of their chemical composition

The investigation used seven categories of organic fertilizers: Tea Waste (TW), Banana Peel (BP), Egg Shell (ES), Mustard Oil Cake (MOC), Fish Meal (FM), and Biochar Enrich Organic Fertilizer (BEOF). Tea Waste and Banana Peels were collected locally and prepared in the lab, while Egg Shells were gathered from households, and MOC and FM from a fertilizer shop. BEOF was sourced from an organic fertilizer shop. After collection, all materials were dried and blended. Total phosphorus (P) and sulfur (S) in the initial manure samples were measured using colorimetric and turbidimetric methods with a spectrophotometer, following digestion with a di-acid mixture ($\text{HNO}_3\text{-HClO}_4$ 3:1).

Experimental setup

The mineralization of phosphorus (P) and sulfur (S) in different manures was assessed anaerobically using destructive sampling. Plastic pots were filled with 50g of air-dried soil, and manures (TW, BP, ES, MOC, FM, BEOF) were mixed with the soil at a rate of 20 ton/ha (0.5g manure per 50g soil). Control treatments had no added manure. Moisture was maintained at field capacity (25% moisture), and the pots were incubated for 90 days at 25°C. Each treatment was replicated three times.

Organic fertilizer incubation in soil

For measuring phosphorus (P) and sulfur (S) mineralization in anaerobic incubation, plastic cups (7 cm diameter, 12 cm height) were used. The containers were placed in a dark place at $25 \pm 2^\circ\text{C}$ for 90 days, wrapped with parafilm to reduce water loss, and kept in an air-conditioned room with proper aeration. Soil moisture content was recorded every 15 days and adjusted with deionized water to maintain specific moisture levels. Destructive sampling was conducted, with the first sampling done the day after incubation to establish preliminary soil content. The initial weights of the cups with soil were also recorded.

Analysis of soil samples after manure application

Soil sampling was conducted from each replicate using destructive methods at 1, 15, 30, 45, 60, 75, and 90 days after applying organic manure. Phosphorus availability in the soil was determined using the NaHCO_3 extraction method (pH 8.5), and available sulfur was quantified using a CaCl_2 extractant solution followed by turbidity measurement with a spectrophotometer. Data were adjusted for moisture content and presented on an oven-dry basis.

Statistical analysis

Data were analyzed using ANOVA with Statistix 10, applying a repeated measures design. Mineralization kinetics were modeled using Sigma-Plot 14.0 with

non-linear regression (Levenberg–Marquardt algorithm). Post hoc Tukey–Kramer tests identified differences among modeled values of P_0 , S_0 , and rate constant k . All tests were significant at $P < 0.05$, unless stated otherwise.

Results and discussion

Effect of different treatments on sulfur (%) release content of the soil sample

The release of sulfur varied significantly across treatments and over time. Under anaerobic conditions, mustard oil cake (T₁) had the highest sulfur release at 26.76% on Day 1, followed by fish meal (T₂) at 24.59% (Table 1). Bone meal (T₃) had 20.32%, while the control (T₆), biochar-enriched organic fertilizer (T₄), and vermicompost (T₅) had the lowest releases (12.30%, 17.15%, and 18.11%, respectively). By Day 15, T₁ and T₂ still showed the highest sulfur levels (32.77% and 29.11%), with T₃ at 20.11% and T₆ at 10.62%. On Day 30, T₁ and T₂ maintained the highest releases (42.18% and 41.90%), while T₆ had the lowest (16.72%). By Day 45, T₁ led with 24.89%, and T₆ had 12.59%. On Day 60, T₁ and T₂ both had the highest sulfur levels (91.99% and 91.02%), compared to T₆, T₄, and T₅ with lower levels. On Day 75, T₁ showed the highest sulfur release (102.98%), followed by T₂ (101.32%) and T₃. T₆ had 80.08%. By Day 90, T₁ and T₂ had the highest releases (84.76% and 79.85%), while T₆ had the lowest (60.95%).

Table 1. Effect of different treatments on sulfur (%) content of the soil sample

Treatment	1 Day	15 Day	30 Day	45 Day	60 Day	75 Day	90 Day
T ₁	26.76 a	32.77 a	42.18 a	24.89 a	91.99 a	102.98 a	84.76 a
T ₂	24.59 ab	29.11 a	41.90 a	18.89 b	91.02 ab	101.32 a	79.85 ab
T ₃	20.32 bc	20.11 b	32.45 b	17.31 b	84.05 bc	92.79 ab	72.91 bc
T ₄	18.11 cd	15.78 c	26.01 bc	17.07 b	80.08 cd	87.32 b	69.18 c
T ₅	17.15 cd	15.69 c	24.79 c	15.86 bc	78.73 cd	86.75 b	68.97 c
T ₆	12.30 d	10.62 d	16.72 d	12.59 c	72.71 d	80.08 b	60.95 d
LSD	6.14	3.85	6.86	4.26	7.67	13.03	7.99
Level of significance	**	**	**	**	**	*	**

Mean with similar letter do not differ significantly. T₁= Mustard oil Cake, T₂= Fish meal, T₃= Bone meal, T₄= Biochar enriches organic fertilizer, T₅= Vermicompost and T₆= control

Sulfur release after mineralization in modified soils is influenced by the sulfur concentration of decomposing materials (Reddy *et al.*, 2001; Islam *et al.*, 1998; Islam *et al.*, 2021). Mustard oil cake,

derived from mustard seeds, has high sulfur content due to glucosinolates, sulfur-containing compounds. Fish meal also contains significant sulfur from sulfur-containing amino acids like cysteine and methionine.

Among the materials studied, mustard oil cake exhibits the highest sulfur release due to its naturally high sulfur content and rapid decomposition rate, which facilitates quicker nutrient release into the soil.

Effect of different treatments on phosphorus (%) release content of the soil sample

The release of phosphorus content varied significantly among different treatments under anaerobic conditions. On Day 1, the T1 treatment (mustard oil cake) had the highest phosphorus release (4.99%), while the T6 (control) had the lowest (3.44%). By Day

15, T1 maintained the highest release (5.50%), with T6 remaining the lowest (4.82%). On Day 30, T1 again had the highest release (5.63%), and T6 the lowest (4.93%). On Day 45, all treatments showed similar results, but T1 was highest (6.65%) and T6 lowest (5.71%). By Day 60, T1 had the greatest phosphorus release (6.51%), and T6 the least (5.77%). On Day 75, T1 and T2 had the highest releases (14.12% and 14.04%, respectively), with T6 the lowest (12.82%). Finally, on Day 90, T1 exhibited the highest phosphorus release (20.24%), while T6 remained the lowest (18.06%) (Table 2).

Table 2. Effect of different treatments on phosphorus (%) content of the soil sample

Treatment	1 Day	15 Day	30 Day	45 Day	60 Day	75 Day	90 Day
T ₁	4.99 a	5.50 a	5.63 a	6.65 a	6.51 a	14.12 a	20.24 a
T ₂	4.96 a	5.39 ab	5.38 ab	6.61 a	6.32 a	14.04 a	20.02 ab
T ₃	4.46 ab	5.28 abc	5.24 ab	6.56 a	6.30 ab	13.54 ab	19.55 ab
T ₄	4.31 ab	5.21 abc	5.05 b	6.48 a	6.26 ab	13.45 ab	19.53 ab
T ₅	4.18 ab	5.00 bc	5.01 b	6.40 a	6.22 ab	12.94 ab	19.07 bc
T ₆	3.44 b	4.82 c	4.93 b	5.71 a	5.77 b	12.82 b	18.06 c
LSD	1.28	0.49	0.58	0.96	0.53	1.20	1.04
Level of significance	NS	NS	NS	NS	NS	NS	**

Mean with similar letter do not differ significantly. T₁= Mustard oil Cake, T₂= Fish meal, T₃= Bone meal, T₄= Biochar enriches organic fertilizer, T₅= Vermicompost and T₆= control

Research by Ghosh *et al.* (2012) showed that applying mustard oil cake (MOC) increases soil organic carbon, enhancing microbial biomass and facilitating phosphorus solubilization. Tiwari *et al.* (2018) further noted that MOC boosts populations of phosphorus-solubilizing bacteria (PSB), leading to greater mineralization of organic phosphorus and improved

phosphorus availability for plants. Additionally, Das *et al.* (2020) found that MOC offers a steady release of phosphorus over time compared to traditional fertilizers, while also enhancing soil structure and microbial diversity. This slow-release characteristic contributes to sustained phosphorus availability throughout the growing season.

Table 3. Effect of different treatments on pH release content of the soil sample

Treatment	1 Day	15 Day	30 Day	45 Day	60 Day	75 Day	90 Day
T ₁	6.63 ab	6.60 a	6.83 a	6.57 ab	6.67 ab	6.60 a	6.57 a
T ₂	6.70 ab	6.67 a	6.27 c	6.77 a	6.53 b	6.60 a	6.60 a
T ₃	6.57 b	6.53 ab	6.57 b	6.57 ab	6.57 b	6.57 a	6.50 a
T ₄	6.67 ab	6.33 b	6.63 b	6.43 ab	6.57 b	6.60 a	6.30 b
T ₅	6.73 a	6.37 b	6.53 b	6.47 ab	6.60 b	6.60 a	6.50 a
T ₆	6.73 a	6.50 ab	6.60 b	6.37 b	6.87 a	6.70 a	6.20 b
LSD	0.15	0.20	0.12	0.34	0.22	0.21	0.14
Level of significance	NS	*	**	NS	*	NS	NS

Mean with similar letter do not differ significantly. T₁= Mustard oil Cake, T₂= Fish meal, T₃= Bone meal, T₄= Biochar enriches organic fertilizer, T₅= Vermicompost and T₆= control

Effect of different treatments on pH release content of the soil sample

The release of pH content varied significantly among treatments on different days. On Day 1, treatments T1

and T5 had the highest pH release at 6.73%, while treatment T3 (bone meal) recorded the lowest at 6.57%. By Day 15, T2 and T1 again showed the highest pH levels (6.67% and 6.60%, respectively), whereas

T4 had the lowest at 6.33%. On Day 30, T1 reached the peak pH release of 6.83%, with T2 showing the least at 6.27%. On Day 45, T2 (using fish meal) recorded the highest pH level (6.77%), while the control T6 had the lowest at 6.37%. On Day 60, T6 showed the greatest pH release at 6.87%, statistically similar to T1 (6.67%), whereas T2 had the lowest at 6.53%. On Days 75 and 90, treatments released similar pH levels, with the maximum from T2 (6.60%) and the minimum from the control (6.20%). Overall, the treatments demonstrated varying pH releases throughout the study period, with T2 and T1 consistently showing higher levels (Table 3).

The initial pH of the materials and the environmental conditions, such as moisture and temperature, also play crucial roles in determining the final pH during decomposition. Some soil characteristics, such as type, 26 27 depth, temperature, moisture content, pH, C/N ratio and complex carbohydrate content influence the mineralization of organic manures in soil (Sleutel *et al.*, 2009). A study by Sinha *et al.* (2020) showed that mustard oil cake application can lead to an increase in soil pH, particularly in acidic

soils. This is due to the calcium carbonate content in the cake, which acts as a liming agent.

Effect of different treatments on EC release content of the soil sample

The release of EC content varied significantly across different treatments and days. On Day 1, all treatments showed similar EC release, with T1 having the highest at 0.18%, followed by T2 (0.17%) and T3 (0.16%). On Day 15, T1 and T2 had the highest EC releases (0.66% and 0.49%, respectively), while the control (T6) had the lowest (0.11%). By Day 30, T1 continued to show the highest EC release (0.70%), with T2 at 0.39% and T6 at 0.10%. On Day 45, T1 and T2 had the highest releases (0.67% and similar), with T6 remaining lowest (0.11%). By Day 60, T1 had the highest release (0.50%), T2 was second (0.31%), and T6 still had the lowest (0.11%). On Day 75, T1 maintained the highest EC release (0.37%), with T2 second (0.22%) and T6 lowest (0.11%). On Day 90, T1 had the highest release (0.35%), similar to T2 (0.32%) and T3 (0.28%), while T6, T4, and T5 had the lowest releases (0.13%, 0.17%, and 0.17%, respectively) (Table 4).

Table 4. Effect of different treatments on EC release content of the soil sample

Treatment	1 Day	15 Day	30 Day	45 Day	60 Day	75 Day	90 Day
T ₁	0.18 a	0.66 a	0.70 a	0.67 a	0.50 a	0.37 a	0.35 a
T ₂	0.17 a	0.49 ab	0.39 b	0.51 a	0.31 b	0.22 b	0.32 a
T ₃	0.16 a	0.15 b	0.13 c	0.14 b	0.17 c	0.16 bc	0.28 a
T ₄	0.15 a	0.12 b	0.12 c	0.14 b	0.14 c	0.13 c	0.17 b
T ₅	0.15 a	0.12 b	0.12 c	0.11 b	0.11 c	0.12 c	0.16 b
T ₆	0.13 a	0.11 b	0.10 c	0.11 b	0.11 c	0.11 c	0.13 b
LSD	0.06	0.39	0.04	0.26	0.07	0.08	0.08
Level of significance	NS	*	**	**	**	**	**

Mean with similar letter do not differ significantly. T₁= Mustard oil Cake, T₂= Fish meal, T₃= Bone meal, T₄= Biochar enriches organic fertilizer, T₅= Vermicompost and T₆= control

Mustard oil cake is likely to release the highest EC (Electrical Conductivity) under anaerobic conditions compared to bone meal, fish meal, biochar, and vermicompost due to its readily decomposable organic matter content. Mustard oil cake is rich in soluble nutrients and proteins, which break down quickly in the absence of oxygen, releasing electrolytes that contribute to a higher EC. In contrast, bone meal and fish meal have a 28 29 slower decomposition rate due to complex organic

structures. Biochar, while high in carbon, holds onto nutrients tightly, and vermicompost, though nutrient-rich, releases them more gradually (Bera *et al.*, 2014).

Conclusion

The result of the experiment shows that the maximum available phosphorus and sulfur was recorded from mustard oil cake and the minimum available sulfur was recorded from the control treatment. Mustard oil cake and fish meal increase

pH most and control treatment decreases pH most. Mustard oil cake is also responsible for greater electrical conductivity. Composition of manures, local management techniques in terms of treatment, storage and field application and ambient climatic conditions have a significant impact on nutrient mineralization. The nutrient release pattern should be developed first considering the soil condition, and then there must be adequate coordination of nutrient input and crop demand based on the data obtained from farm manure mineralization research. The use of manure at the right time and in the right amount will prevent nutrient shortage or over usage in crop production, provide balanced fertilization and, ultimately, save our ecosystem.

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