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Determination of Bio-fertilizer Quality (N, P, K, Mg and Ca) of Digestate of Laying Birds Droppings Co-digested with Broiler Droppings in a 50:50 Ratio

Hafsat OO^{1*}, Ambeatrice K¹, Alfa IM², Okpanachi U¹

ABSTRACT

This study evaluated the bio-fertilizer quality of digestate obtained from the anaerobic co-digestion of laying birds and broiler droppings in a 50:50 ratio. The study's objectives were to ascertain the bio-fertilizer quality (N, P, K, Mg, and Ca) of laying bird and broiler (50:50) droppings before anaerobic co-digestion of the substrate and evaluate the impact of the anaerobic co-digestion process on the bio-fertilizer quality of the digestate and substrate. The initial measurements of the substrate showed the following values: pH (6.9), temperature (29.03°C), nitrogen (0.28%), phosphorus (2.01%), potassium (2.58%), magnesium (0.68%), and calcium (3.01%). After the anaerobic co-digestion, significant changes were noticed. The result showed that the pH increased by 4.3% to 7.2, while the temperature rose by 10.9% to 32.20°C. Nitrogen concentration was reduced significantly by 70.0% to 0.09%, whereas phosphorus declined by 32.8% to 1.35%. The potassium content was reduced by 23.6% to 1.97%, magnesium by 11.8% to 0.60%, and calcium by 7.0% to 2.8%. The result indicated substantial differences ($p \leq 0.05$) between the substrate and digestate for temperature, nitrogen, phosphorus, and potassium, although pH, magnesium, and calcium variations were not statistically significant. Regardless of these reductions, the digestate retains substantial nutrient concentrations, affirming its value as a bio-fertilizer and using digested poultry droppings as bio-fertilizers can augment soil fertility and improve sustainable agricultural practices.

Keywords: Bio-fertilizer, Substrate, Digestate, Anaerobic Co-digestion, Poultry Droppings.

1. INTRODUCTION

Currently, organic droppings like poultry droppings are receiving more attention as fertilizers due to the rising cost of inorganic fertilizers coupled with the limited ability of inorganic fertilizers to improve soil quality (Arancon et al., 2008). The

poultry droppings is an exceptional source of organic fertilizer which contains a high percentage of nitrogen, phosphorus, potassium, and other vital nutrients readily available for plant uptake as compared to other organic sources (Garg & Bahl, 2008; Mohamed et al., 2010).

Poultry droppings can pose a potential threat to the health of human beings and have a harmful impact on the surrounding natural environment, particularly the soil and quality of water (Myszograj & Puchalska, 2012). In this regard, digestate the residue of biogas material, a fully fermented nutrient-rich material, could be used as an alternative to mineral fertilizer (Eickenscheidt et al., 2014; Koszel & Lorencowicz, 2015; Vázquez-Rowe et al., 2015).

Digestate comprises microbial biomass, semi-degraded organic matter, and inorganic compound, and, therefore, can be used as soil conditioners on farmlands (Albuquerque et al., 2012). The digestate contains more readily available nutrients than the substrate products, which makes it better for crop fertilization (Lansing et al., 2010; Garfi et al., 2011; Goberna et al., 2011).

Several publications demonstrated the effectiveness of digestate as a suitable nutrient source in agriculture (Chantigny et al., 2008; Arthurson, 2009; Panuccio et al., 2016; Muscolo et al., 2017). There was a high degree of freedom of the total amount of mobile organic fertilizers when digestate was used in soil with an increase in soil N and N use efficiency, in comparison to the direct soil incorporation of green droppings or crop postharvest residues (Möller and Müller. 2012).

Bio-Fertilizers are one of the most effective agricultural technologies, and using them in the field is highly beneficial as they contain beneficial living microorganisms that improve the soil fertility by colonizing the rhizosphere of the plant or interior and boosting plant growth by increasing nutrient supply (N, P, K, S, etc.) to the host plant through natural processes including nitrogen fixing, phosphorus solubilization; when used to seeds, plants, or soil (Ghosh, 2004; Bhavyaa et al., 2017; Mahanty et al., 2017).

The use of digestate bio-fertilizers is one of the essential components of integrated nutrient management, as they are cost-effective and are renewable sources of plant nutrients for sustainable agriculture (Grigatti et al., 2011). The demand for digestate bio-fertilizers is dependent on compliance with quality standards (Albuquerque et al., 2012). Although there is a lot of research on the production of biogas from different substrates, there is very little data on the fertilizer and sanitary quality of digestate from anaerobic digestion in scientific publications. However, the fertilizer potential of digestate from farm and agro-industrial residues was investigated (Albuquerque et al., 2012). Also, Alfa et al. (2014) assessed the bio-fertilizer quality of digestate from the digestion of cow dung and poultry droppings.

Co-digestion offers several advantages over the digestion of individual substrates. It can lead to increased biogas production due to the synergistic effects of the different feedstocks (Holm-Nielsen et al., 2009). The combinations of complementary substrates can provide a balanced nutrient composition, enhance the buffering capacity, and improve the overall process stability (Mata-Alvarez et al., 2011).

2. MATERIALS AND METHODS

Study Area

The experiment was carried out at the Teaching and Research Farm in the Department of Animal Production, University of Jos, Naraguta Campus, Jos-North Local Government Area (LGA) of Plateau State (Fig.1). Jos-North LGA receives an annual rainfall ranging from 1,400 mm to 1,500 mm (55.1 to 59.1 inches), with the peak precipitation occurring between July and August (Ishaya et al., 2008). The mean annual temperature ranges from 21°C to 25°C (69.8°F to 77°F), with the coolest months being December and January (Ishaku et al., 2012).

Materials

The following are the materials (equipment) used in this study;

- i. A Multifunction Water Quality Tester (model number EZ-9909SP) was used to determine the pH and temperature of the Substrate and the digestate.
- ii. New Sacks: New sacks were used as a medium to aid the drying process of the droppings and for storage.
- iii. Personal protective equipment such as farm coat, hand gloves, nose masks, and boots.
- iv. Measuring cylinder: for measuring equal Volume of poultry droppings to water for slurry preparation.
- v. Weight balance: was used for measuring the Weight of the wet and dry droppings.

- vi. Sieve: was used to separate and sieve out litter materials.
- vii. Droppings from laying birds and broiler birds was used as the biomass.
- viii. Fresh cow dung was used as inoculum.
- ix. Water.
- x. Wooden spatula: was used for stirring during the slurry preparation.
- xi. An eighty (80) liter galvanized steel bio-digester with accessories.
- xii. Sample collection bottles.

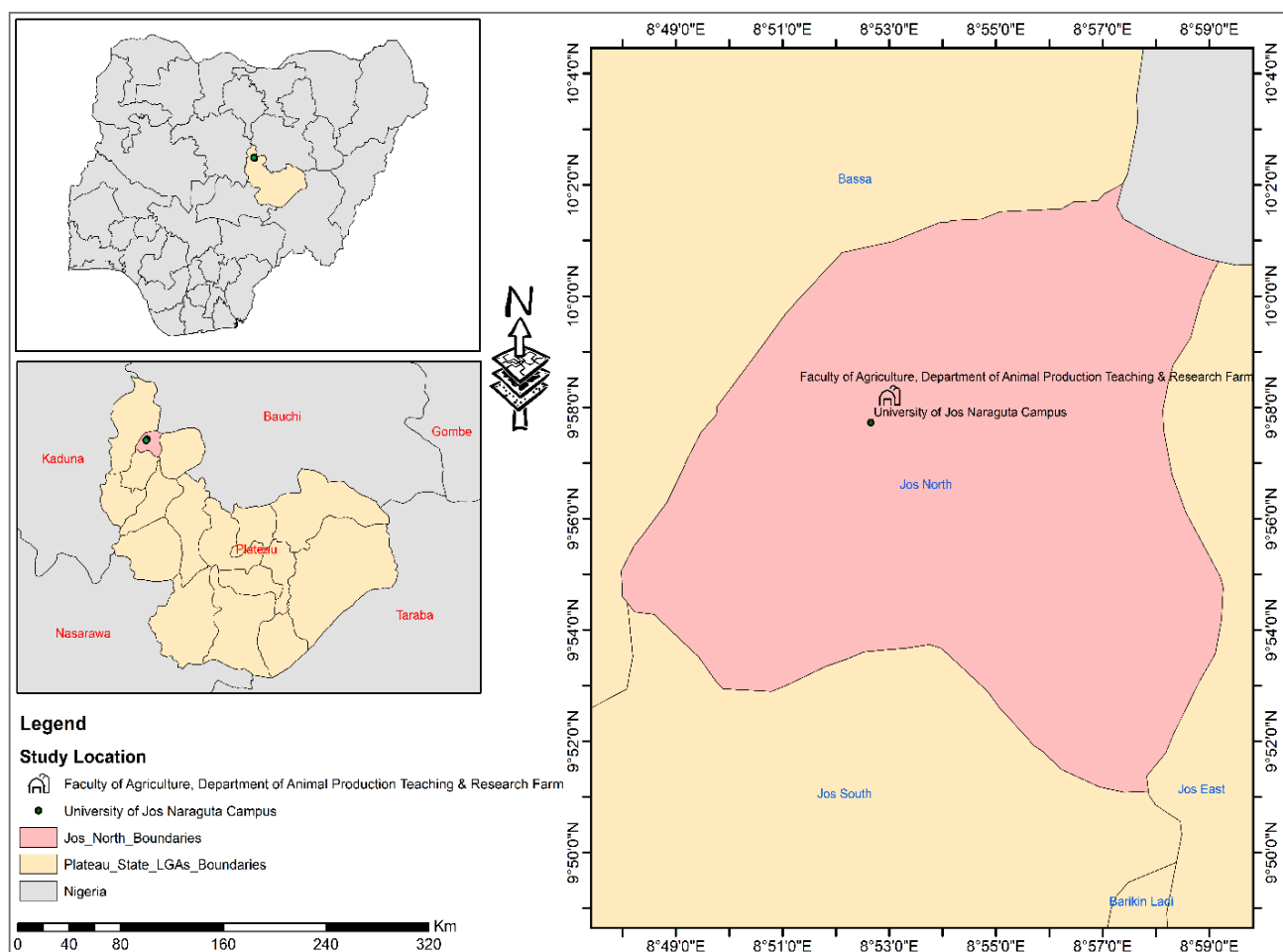


Figure 1: Map of Nigeria Showing Plateau State and the Study Area (OSGOF, 2024)

Biomass Collection

The droppings of the laying bird and broiler were obtained from the pen house at the Teaching and Research Farm in the Department of Animal Production, University of Jos, Naraguta Campus. The droppings of the birds that were wet was weighed and poured on new sacks (as a biosecurity measure to prevent contamination) and left to sundry (Plate 1). The dried bird droppings were manually sieved to extract the feed ingredients, weighed, and then placed in new sacks for storage.

Design and Development of Bio-Digester

The eighty (80) litre bio-digester used was designed at the Department of Civil Engineering in the Faculty of Engineering, University of Jos, and the fabrication took place at Dilimi, Fabrication Market, Jos-North, Plateau state (Plate 2). The galvanized metal sheet was used for fabrication due to its high corrosion resistance.



Plate 1: Sun-drying of Droppings

Slurry Preparation

The biomass (droppings) and water were mixed in equal proportion (1:1) by Volume to form a slurry. The biomass was made from a combination of 50% and 50% dried layer and broiler droppings. Five (5) litres of broiler bird droppings, five (5) litres of laying bird droppings, and ten (10) litres of water were combined to create the slurry, which was then put into a 20-litre measuring cylinder. The slurry was stirred with a wooden spatula to ensure there were no lumps.



Plate 2: Bio-digester Layout

Sample Collection and Digester Loading

Samples of 1.5 litres of the slurry were collected in sterilized bottles (sterilized using hot water) before being loaded into the bio-digester (substrates) and after the retention time (the digestate) for laboratory analysis. A Multifunction Water Quality Tester (model number EZ-9909SP) was used to detect the temperature and pH of the substrate and digestate of the droppings,

The slurry was introduced into the eighty (80) litres bio-digester through the inlet pipe. In order to seed the bio-digester and initiate the anaerobic digestion process, 1.6 litres of fresh cow dung was utilized as the inoculum. The bio-digester was sealed completely and placed outside the shed, above the ground level, since warm temperatures from sunlight exposure can raise the metabolic rate of the bacteria in the biodigester and accelerate a higher rate of organic matter breakdown.

Laboratory Analysis

After sample collection, the Substrate and digestate samples were brought to Optima Kings Research Center and Laboratories, which is situated at No. 4 Miango Road, Kuffang, Jos, Nigeria. The Nitrogen (N), Phosphorus (P), Potassium (K), Calcium (Ca), and Magnesium (Mg) contents of the samples were examined to ascertain their bio-fertilizer quality.

Determination of Nitrogen Content

The protein content in the collected samples was ascertained using the micro Kjeldahl method, as stated by AOAC-979.09 (AOAC, 2005). The micro Kjeldahl flask was filled with ten (10) grams of each sample taken from the substrate and digestate. Five (5) millimetres of concentrated tetra-oxo-sulphate (vi) acid (concentrated. H₂SO₄) and a tablet of selenium were added. The same treatment was applied to another five (5) millimetres sample, which was then measured. The flask was digested at a red-hot temperature in a fume cupboard for two hours. A volumetric flask was filled with the digests.

In the meantime, a clear solution was collected after two hours to demonstrate the digestion. Each of the transferred digests was diluted to 50 ml with distilled water. The distillate was recovered in 10 millilitres of 4% NaOH after ten millilitres of each dilution were pipetted onto powered heat. 50 ml of the distillate from each duplicate was titrated with 0.02 NH₂SO₄ to produce a pink colour. Each mixture was distilled using steam-powered heat, and the distillate was collected into 10 ml of 4% boric acid solution containing three drops of mixed indicator. The percentage of protein was calculated by multiplying the percentage of nitrogen in each by a factor of 6.25. The total Nitrogen content was calculated by using the relationship that 1 ml of H₂SO₄ = 14 mg of H₂SO₄ thus,

$$N_2 = \frac{100 \times N \times 14 \times Vt}{W} \times T - B$$

Where;

T = Titre value of the sample

B = Blank titer value

Ca = Volume of digest distilled

Vt = Total Volume of digest

N = Normality of acid used

W = Weight of sample used

Determination of Phosphorus Content

Phosphorus content was ascertained using the molybdate method, which involves hydroquinone as a reducing agent (Onwuka. 2005). 0.5 ml of the mineral digest was mixed with 1.0 ml of ammonium molybdate, 1.0 ml of sodium sulphate, and 1.0 ml of hydroquinone and mixed before being left to stand for half an hour. A colorimeter set at 660 nm was used to measure the blue color that formed compared to a standard.

Determination of Calcium Content

The titration method, as described by Pearson (1991), was used to determine the calcium content. 250 ml conical flask was pipetted with around 10 ml of each sample, and 25 ml of KOH was added along with a pinch of calcine indicator. The combination was titrated against an ethylene diamine tetra-acetate (EDTA) solution to obtain the endpoint. The Volume of calcium in the sample is equal to the Volume of EDTA.

Determination of Potassium Content

A.O.A.C. Method 958.02 (Potassium in Fertilizers) was used to determine the amount of potassium in the samples (AOAC, 2005). The samples were digested with ammonium oxalate, (NH₄)₂C₂O₄, and hydrogen peroxide, H₂O₂. After digestion, the samples were titrated with BAC solution (benzalkonium chloride) using a sodium tetraphenyl boron (STPB) indicator to measure the potassium.

Determination of Magnesium Content

AOAC 964.01D method was used to determine the magnesium content (AOAC, 2005). A 500 ml volumetric flask was filled with one gram of the sample, and 350 ml of water was added and boiled for an hour (1h). After boiling, it was allowed to cool, diluted to

Volume, mixed, and filtered. The solution prepared was filled into a beaker in aliquot portions, and the pH was adjusted with KOH to ascertain the magnesium content.

Study Duration

Thirty (30) days were spent monitoring the experimental setup.

Statistical and Data Analytics

The IBM Statistical Package for Social Science (SPSS) for Windows version 20.0 was used to do statistical analysis, which included an independent t-test at $p \leq 0.05$ level of significance.

3. RESULTS AND DISCUSSION

The results of laboratory analyses of the bio-fertilizer quality of the poultry droppings before co-digestion and the effectiveness of the anaerobic co-digestion process on the bio-fertilizer quality in the substrate and digestate from laying birds and broiler droppings, respectively, are shown in Table 1-2. Additionally, the percentage differences in these parameters before and after the digestion process are shown in Table 2. The physical properties and the nutrient content of the substrate and digestate are visually compared in Figures 2 and 3, respectively. The statistical analysis of the co-digestion process shown in Table 3 compares the properties of the substrate and the digestate that results.

Table 1: Bio-Fertilizer Quality of Laying Birds and Broiler (50:50) Droppings Before (Substrate) Anaerobic Co-Digestion

Parameters	Substrate
pH	6.9
Temperature (°C)	29.03
Nitrogen (%)	0.28
Phosphorus (%)	2.01
Potassium (%)	2.58
Magnesium (%)	0.68
Calcium (%)	3.01

Keys: °C = Degree Celsius, % = Percentage

Table 1 presents various parameters and their respective bio-fertilizer quality values for the substrate of laying bird droppings co-digested with broiler bird droppings in a 50:50 ratio. According to Table 1, the pH value of 6.9 indicates that the poultry droppings are almost neutral. Most crops benefit from a pH level within the optimal range (6.0 to 7.0) for nutrient availability and microbial activity in the soil. The near-neutral pH also helps prevent the volatilization of ammonia, thereby retaining nitrogen in a form that plants can use more efficiently. The result is consistent with a study by Ravindran et al. (2017), who found that the pH of substrate poultry droppings was within the range of 6.94.

The temperature of 29.03°C is typical for fresh poultry droppings, which supports the activity of mesophilic microorganisms and is essential for the initial stages of organic matter breakdown and nutrient cycling. Hence, maintaining the entire biological activity in the substrate before anaerobic digestion begins is essential. In the study of Bouzid and Djadi (2015), the range of temperature for substrate poultry droppings was reported to be 28°C. The result of this study corresponds with the findings of these authors.

The low nitrogen content (0.28%) may be caused by the initial Substrate state, where some nitrogen may be present in forms that are not immediately available to plants. Typical values for poultry droppings are often within the range of 2.40% (Ravindran *et al.*, 2017). The initial substrate condition, where some nitrogen may be present in forms that are not instantly available to plants, could cause the low nitrogen content. The phosphorus concentration is relatively high (2.01%). According to Omer and Hasan (2018), in their studies, phosphorus level was recorded to be between (2.3%) in substrate poultry droppings. Phosphorus is an essential bio-fertiliser content due to its high percentage in this study, as it supports strong root systems and early plant growth stages. The potassium level (2.58%) in this study closely aligns with the study of Omer and Hasan (2018), who recorded potassium as 2.18%. Plants require potassium for

different biological processes, such as photosynthesis and water regulation, and at adequate levels to improve disease and drought resistance.

This study found a 0.68% level of magnesium, which is consistent with the findings from Omer and Hasan (2018), who recorded a magnesium level of 0.62%. This content level can improve plant growth because magnesium is rarely found in soils. The calcium content in the substrate droppings is relatively high (3.01%), which is in line with the calcium level recorded in the study of Omer and Hasan (2018), which shows that the calcium level of substrate poultry droppings is 4.96%. The calcium level in the droppings can help improve soil structure, promote root development, and enhance plants' overall nutrient absorption capacity.

The findings show that substrate poultry droppings have a balanced nutrient profile with particular phosphorus, potassium, and calcium strengths. The essential macronutrients and near-neutral pH make poultry droppings a valuable bio-fertilizer.

Table 2: Effect of the Anaerobic Co-Digestion Process on the Bio-Fertilizer Quality in the Substrate and Digestate from Laying Birds and Broiler (50:50) Droppings

Parameters	Substrate	Digestate	% Difference
pH	6.9	7.2	4.3
Temperature (°C)	29.03	32.20	10.9
Nitrogen (%)	0.28	0.09	-70.0
Phosphorus (%)	2.01	1.35	-32.8
Potassium (%)	2.58	1.97	-23.6
Magnesium (%)	0.68	0.6	-11.8
Calcium (%)	3.01	2.8	-7.0

Keys: °C = Degree Celsius, % = Percentage

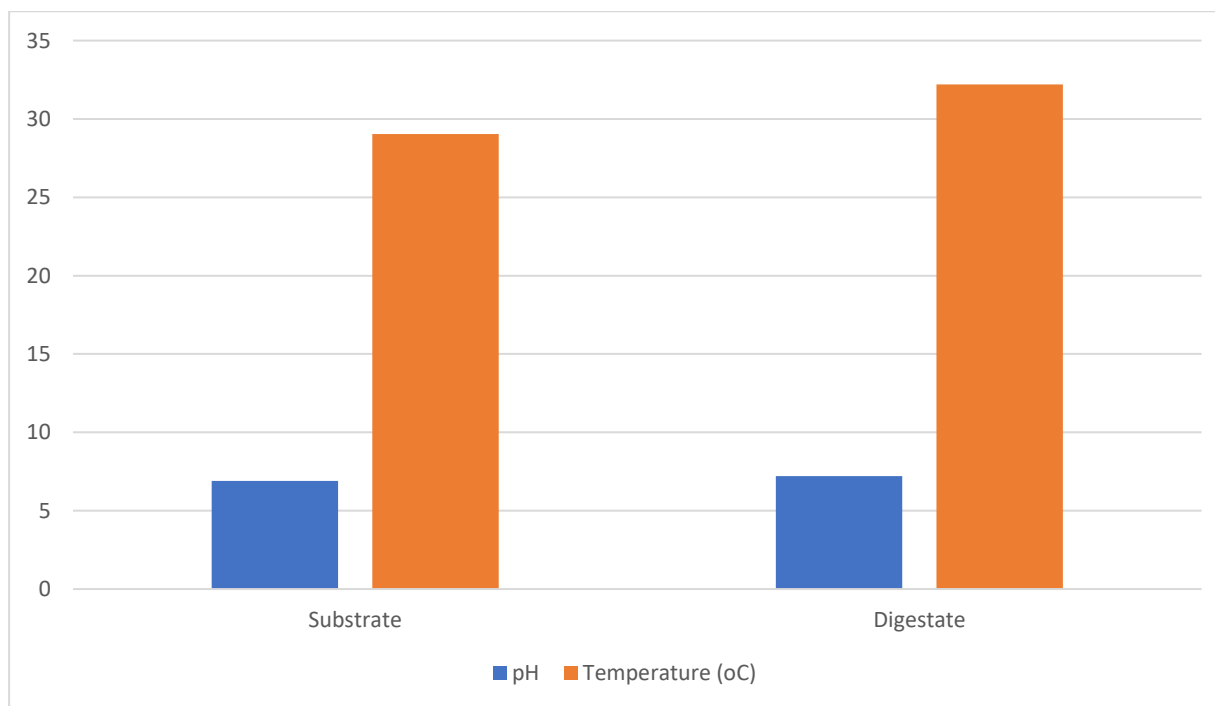


Figure 2: Visual Comparison of the Physical Properties of the Substrate and Digestate

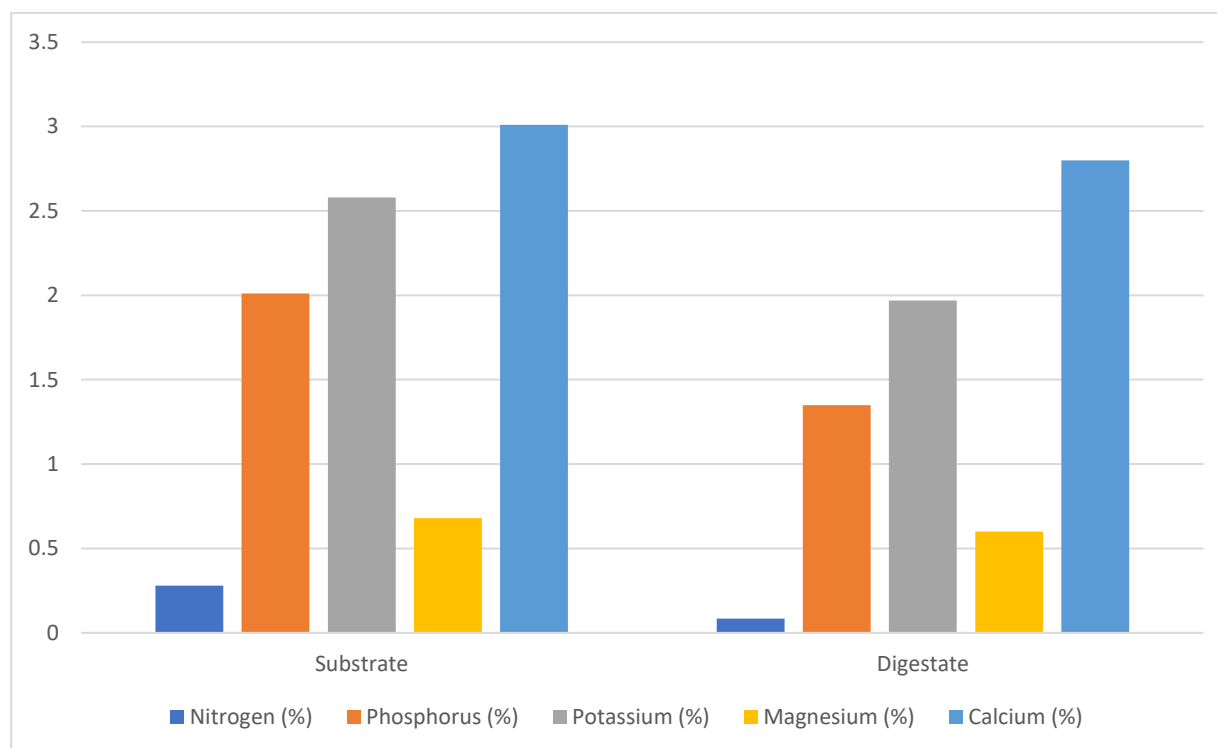


Figure 3: Visual Comparison of the Nutrient Content of the Substrate and Digestate

Table 2 and Figures 2-3 show that the pH of the substrate sample was 6.9, indicating a slightly acidic environment. In contrast, the digestate sample had a pH of 7.2 (a 4.3% increase), indicating a slightly alkaline environment. The result is consistent with the study by Lukehurst et al. (2010), noted a slight change in pH level from 6.9 before digestion to 7.6 after the digestion process.

The temperature of the digestate was 32.2°C, which is warmer than the temperature of substrate sample which is 29.03°C—a difference of 10.9%. This temperature rise can be beneficial, as it often boosts the efficiency of microbial activity. As a result, it helps to break down organic matter more effectively and promotes the release of nutrients. This result aligns with what Insam et al. (2015) found, indicating that mesophilic anaerobic digestion usually works best at temperatures between 30°C and 40°C for digestate samples.

The nitrogen content decreased significantly from 0.28% in the substrate sample to 0.09% in the digestate sample (a 70% reduction). The reduction is more pronounced than what some other studies have found. Tambone et al. (2010) reported reductions of nitrogen by 15-30% in various organic substrates after undergoing anaerobic digestion. The reduction in nitrogen content may be attributed to the volatilization of ammonia during the digestion process. Although a low nitrogen content in the digestate sample may seem like a disadvantage, it is important to recognize that the remaining nitrogen is likely to be in a more stable and plant-available form, which enhances its effectiveness as a fertilizer (Möller & Müller, 2012). The phosphorus content also decreased from 2.01% to 1.35% (a 32.8% reduction) after co-digestion. In the study conducted by Avedun et al. (2023), there was a slight decrease in phosphorus levels in the digestate samples, dropping to 0.28%. Additionally, the potassium content was reduced from 2.58% to 1.97%, a decrease of 23.6%. This finding is noteworthy because potassium is usually conserved during anaerobic digestion. The recorded potassium level after digestion was slightly lower than that of Avedun et al. (2023), who noted a post-digestion level of 2.09%. The reduction in nutrient levels can result from leaching or microbial uptake during digestion. Potassium is crucial for plant water regulation and enzyme activation, so even with the reduction, the digestate material remains a valuable source of this nutrient.

There was a reduction in the magnesium content, dropping from 0.68% to 0.6%, (a 11.8% reduction). This finding aligns with the results observed by Anthony et al. (2006), which reported a significant reduction from the substrate to the digestate (i.e 1.98% to 0.80%). This minimal reduction indicates that the digestate retains most of its magnesium, ensuring its effectiveness in supporting plant growth.

The calcium content decreased from 3.01% to 2.8%, representing a 7% reduction after anaerobic co-digestion. Anthony et al. (2006) recorded a decrease from 3.31% to 2.24%. The slight decrease in calcium content is not significant enough to hinder the effectiveness of the digestate as a soil amendment, ensuring it still provides sufficient calcium for plant health.

Table 3: Statistical Analysis of the Effect of the Anaerobic Co-Digestion Process on the Bio-Fertilizer Quality in the Substrate and Digestate from Laying Birds and Broiler (50:50) Droppings

Parameters	Substrate Mean \pm SEM	Digestate Mean \pm SEM	P \leq 0.05
pH	6.90 \pm 0.10	7.20 \pm 0.10	0.17
Temperature ($^{\circ}$ C)	29.03 \pm 0.02 ^b	32.20 \pm 0.20 ^a	0.04
Nitrogen (%)	0.28 \pm 0.02 ^a	0.09 \pm 0.00 ^b	0.05
Phosphorus (%)	2.01 \pm 0.02 ^a	1.35 \pm 0.02 ^b	0.00
Potassium (%)	2.58 \pm 0.02 ^a	1.97 \pm 0.02 ^b	0.00
Magnesium (%)	0.68 \pm 0.02	0.60 \pm 0.02	0.11
Calcium (%)	3.01 \pm 0.20	2.80 \pm 0.20	0.54

Keys: SEM = Standard Error Mean, % = Percentage, $^{\circ}$ C = Degree Celcius

Table 3 indicates that pH levels increased from 6.90 \pm 0.10 in the substrate to 7.20 \pm 0.10 in the digestate; however, this difference is not statistically significant. The findings align with Gobind et al. (2018), who noted a pH range of 6.10 to 7.79 for digestate produced from the anaerobic digestion of chicken droppings. The slight rise in pH suggests that anaerobic digestion has a buffering effect, maintaining pH levels within a narrow range due to the production of bicarbonates and ammonia, which is crucial for microbial activity and the subsequent application of the digestate as bio-fertilizer.

The study shows a significant temperature increase from 29.03 \pm 0.02 $^{\circ}$ C to 32.20 \pm 0.20 $^{\circ}$ C during the anaerobic digestion process. The findings are consistent with Anthony et al. (2006), who noted a temperature rise from 32 $^{\circ}$ C to 33.5 $^{\circ}$ C during the anaerobic co-digestion of poultry droppings. There was a significant reduction in nitrogen content, decreasing from 0.28 \pm 0.02% to 0.09 \pm 0.00%. This finding aligns with the study by Omer and Hasan (2018), which reported a decline in nitrogen levels from 0.51% to 0.25% during the anaerobic digestion of poultry droppings. This decrease is primarily attributed to ammonia volatilization and microbial activity, which had effect on the fertilizer value of the digestate and underscore the necessity for additional nitrogen supplementation.

Phosphorus content significantly decreased from 2.01 \pm 0.02% to 1.35 \pm 0.02%. Also, Avedun et al. (2023) reported a phosphorus level of 0.28% after anaerobic co-digestion of animal droppings and sawdust. This consistent reduction in phosphorus content suggests leaching or transformation into less bioavailable forms during digestion, emphasizing the need for phosphorus supplementation to maintain fertilizer quality.

Potassium decreased significantly from 2.58 \pm 0.02% to 1.97 \pm 0.02%. This finding aligns with Avedun et al. (2023), who reported a reduction of 2.1%. The potassium losses during anaerobic digestion result from leaching and microbial uptake, highlighting the necessity for potassium supplementation to improve fertilizer effectiveness. This study found no significant changes in magnesium (0.68 \pm 0.02% to 0.60 \pm 0.02%) and calcium (3.01 \pm 0.20% to 2.80 \pm 0.20%) contents, consistent with Avedun et al. (2023), who reported stability in magnesium and calcium levels during anaerobic digestion of dairy droppings. The stability suggests that these minerals remain largely unaffected by digestion, maintaining their availability in the digestate for plant use.

4. CONCLUSION

This study demonstrates that co-digestion of the droppings of laying birds and broilers through an anaerobic process resulted in high digestate temperature, a reduction in nitrogen, phosphorus, and potassium content while pH, magnesium, and calcium were stable. These findings align with recent research that also indicates nutrient losses, underscoring the advantages of anaerobic co-digestion for improving the quality and stability of bio-fertilizers. Regardless of these reductions, the digestate retains significant nutrient content, affirming its value as a bio-fertilizer. It is recommended that higher application rates be necessary when utilizing digestate compared to substrate due to its lower nutrient content.

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Author contributions

Hafsat OO: Collected and sorted data, conducted experiments and field studies, and also prepared the first draft of the manuscript, covering the primary analysis and results interpretation.

Ambeatrice K: Collected and sorted data and conducted experiments and field studies.

Alfa IM: Created & constructed the Bio-digester Layout. Monitored the research process and progress, provided guidance, ensured the project aligned with the research ethics, and reviewed and edited the manuscript.

Okpanachi U: Monitored the research process and progress, provided guidance, ensured the project aligned with the research ethics, and reviewed and edited the manuscript.

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Conflict of interest

The authors declare that there are no conflicts of interests.

Ethical approval

Not applicable.

Informed consent

Not applicable.

Data availability

All data associated with this study will be made available based on the reasonable request to corresponding author.

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