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Development of an organic fertilizer bioreactor for the bioconversion of dried chicken manure into organic liquid solution

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Abstract Animal manure is an excellent source of nutrients that could support plant nutrients. The result showed that the optimum hydraulic retention period for bioprocessing of dried chicken manure using an organic fertilizer bioreactor is between 7 and 14 days. Within the optimal hydraulic retention periods, the organic fertilizer bioreactor was able to maintain the optimal operating parameters for bioconversion of dried chicken manure to organic liquid fertilizer such as dissolved oxygen (6.67 – 6.73 mg/L), pH (8.83 – 8.88), electrical conductivity (2,061 – 2,096 μS/cm) and total dissolved solids (937 – 947 ppm). In terms of nutrient content, the organic liquid fertilizer produced at optimal hydraulic retention period contained total nitrogen of 233 – 267 ppm, nitrate of 99 – 129 ppm, total phosphorus of 100 – 200 ppm, and total potassium of 400 - 467 ppm. The presence of nitrate suggested that the organic fertilizer bioreactor was able to convert inorganic forms of nitrogen present in the dried chicken manure. The seed germination bioassay suggested that the organic liquid fertilizer produced at optimal hydraulic retention period using an organic fertilizer bioreactor did not inhibit the germination of the cucumber seeds compared to dried chicken manure. The result provided a new methodology for processing dried chicken manure to produce organic liquid fertilizer that could be used as a plant nutrient source alternative to commercial fertilizers.

Keywords: Aerated chicken manure, Biofilter, Circular economy, Manure tea, Nutrient reuse

Introduction

One of the main agricultural sectors in the Philippines is the chicken industry. The overall chicken inventory in the country as of October 2021 was 190 million birds (Philippine Statistics Authority, 2021). This number of chickens could produce a significant amount of dung, unconsumed feed,

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feathers, litter, and other types of agricultural waste. In the Philippines, there are approximately 2.46 million metric tons of manure released yearly from the poultry business (Calub *et al*., 2016). Chicken manure contains a significant amount of nutrients that could support plant nutrient requirements. However, using raw animal excrement as an organic amendment without prior processing has a detrimental environmental impact (Manogaran *et al*., 2022).

It is urgently necessary to develop, adapt, and deploy cutting-edge manure management technologies and methods before land application at a time when agriculture is faced with rising challenges in maintaining crop and animal productivity while conserving soil, water, and air resources (Millner, 2009). One of the five major problems in waste management within a circular economy was the development of technology for nutrient reuse, and this might be achieved by utilizing biological, chemical, and physical techniques as part of treatment systems (Vanotti *et al*., 2020).

The common chicken manure treatment methods for nutrient reuse were anaerobic digestion and composting process. Digestate produced from the anaerobic digestion of animal manure using a biogas digester contains essential elements needed by the plant (Möller & Müller, 2012). However, the fresh fruit output is adversely impacted by using digestate as a source of nutrients for hydroponic tomato cultivation (Mupambwa *et al.*, 2019). On the other hand, vermicomposting of animal manure resulted in a positive result on plant growth and yield, but the process would take up to 6 months (Yadav and Garg, 2011) before the vermicompost could be used as fertilizer. In addition, most of the nitrogen present in manure during the composting period was lost due to ammonia volatilization.

The extraction of nutrients from animal manure through the aerobic fermentation process is gaining attention as an alternative animal manure treatment for nutrient reuse. Aerated animal manure extracts were used as a nutrient source for hydroponic lettuce and kale production and successfully grown using turkey manure extract, however, yield using aerated cow and chicken manure extract was significantly lower when compared to the nutrient solution (Tikasz *et al*., 2019). It has been proposed that adding a pre-processing step could raise nitrate concentrations while lowering ammonium levels in manure extracts, thus reducing toxicity.

The study aimed to develop a batch-type organic fertilizer bioreactor (OFB) for the bioprocessing of organic fertilizer, specifically, dried chicken manure. This research also aimed to investigate the performance of OFB in terms of the quality of produced organic liquid solution (OLS) at different hydraulic retention periods (HRP). A phytotoxicity test of the produced OLS was done to assess the effect of different HRP and unprocessed chicken manure on the seed germination index of cucumber seeds.

Materials and methods

Design and fabrication of the OFB

The design concept was to develop a batch-type OFB that extract essential nutrients from organic nutrient sources through bioconversion with the aid of microorganisms. The OFB was designed for the continuous recirculation of OLS within the system, filtration of solid particulates in the organic fertilizer, and supply of continuous air in the system. The designed OFB maintained important operating parameters such as dissolved oxygen level, temperature, and pH level.

The OFB was comprised of an OLS tank, filtration tank, bioreactor tank, aeration system, and supply and discharge channel as shown in Figure 1. The materials were sourced from local shops within the vicinity of the study area. The OLS tank was made of a plastic container (50 L capacity) with a diameter of 40 cm and a height of 55 cm. It consisted of a feedstock vessel surrounded by a filter net (200 microns) that hold the dried chicken manure to allow the slow release of nutrients in the water.

The filtration tank was made of a plastic container (35 L capacity) with a diameter of 36 cm and a height of 40 cm. The filtration tank consisted of perforated plastic that enclosed the tube in the bottom connected to the organic liquid solution tank and a layer of fishnet (1m x 2.5m with 1 mm grid size), fiber wool (160 gram) enclosed with net and aquarium foam (30 PPI) with a thickness of 2.5 cm that filtered solid particulates present in organic liquid solution.

The bioreactor unit was made of the same plastic container as the filtration tank. It consisted of 500 g of K1 biofilm media that is suspended by diffused air from the aeration system during the processing of organic liquid fertilizer. It was a cylindrical shape material made of polyethylene plastic with a 10 mm diameter, a thickness of between 0.5 to 0.8 mm, a density of 0.123 g/L, and a specific surface area of 500 m^2/m^3. It is served as a carrier for nitrifying bacteria that processed the organic liquid fertilizer to produce nitrate.

The aeration system consisted of an air tube, air diffuser, and electromagnetic air pump (35 Watts with rated airflow of 65 L/min at 0.027 MPa) that delivered air to maintain dissolved oxygen with at least 2 mg/L or above. It served as a mixing mechanism in the organic liquid solution tank and bioreactor unit. The supply and discharge channel consisted of PVC pipes and fittings and an airlift pump which is responsible for the circulation of water in the system without the use of an additional water pump.

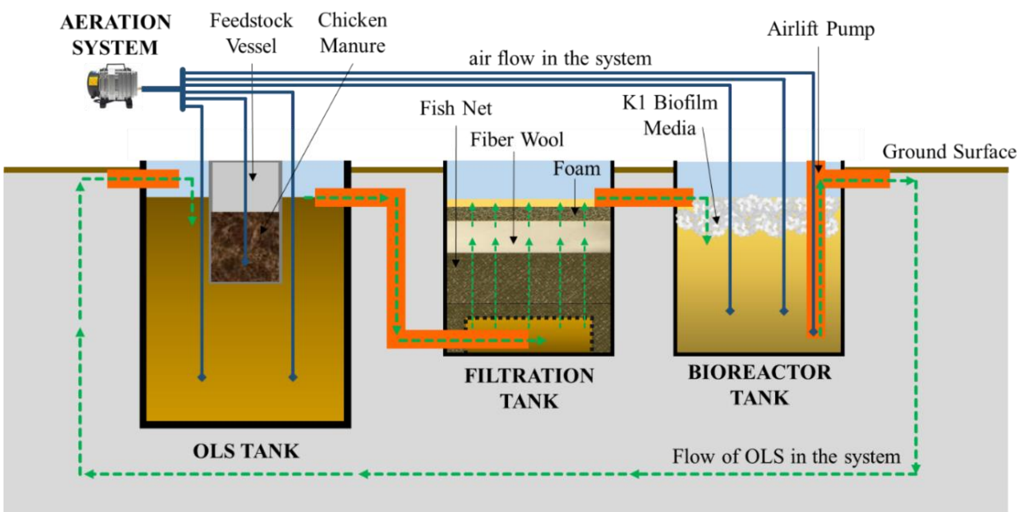

Figure 1. Schematic layout of the developed organic fertilizer bioreactor

Preliminary testing of the bioreactor

Preliminary testing and observation were conducted before the start of the operation of OFB. Each OFB was filled with 100 L of water (optimum operating level) without feedstock and operated for one day. In this procedure, checking for leaks in supply and discharge channels, uniformity of water discharge from the airlift pump, observation of changes in water level, and clogging of moving biofilm media were undertaken to ensure the smooth operation of the system during the bioprocessing of chicken manure using the OFB.

Feedstock preparation

The chicken manure was selected as a feedstock in the production of OLS using OFB. It was collected from the poultry farm of Central Luzon State University. It was laid in plastic and dried under greenhouse conditions (20 - 35^{o}C) for 2 weeks. After drying, the dried chicken manure (DCM) was loaded on the feedstock vessel of OFB. Samples of DCM were collected and placed in a closed container for the determination of physiochemical properties.

Vermicompost tea preparation

The vermicompost was selected as the source of beneficial microorganisms for the developed system. The preparation of vermicompost started with proportioning of test materials [carabao manure (60 kg), Madre de cacao leaves (10 kg), banana stem (10 kg), rice straw (20 kg), and mango leaves (20 kg)]. The materials were mixed and layered to create a small hill of materials. The layered materials were watered until fully saturated. It was inoculated with 50 g of Trichoderma to speed up the process of decomposition of materials. After the inoculation of Trichoderma, the layered materials were covered with mulching plastic films to create a high-temperature environment for decomposition. After two weeks, test materials were mixed thoroughly, and watered until they became saturated and covered with mulching plastic films. After another two weeks, the materials were mixed, collected, and put in the worm bin. After one week, the test materials inside the worm bin were inoculated with 1kg of worm (African night crawler sn. *Eudrilus Euginiae*). The vermicomposting process started after the inoculation of the worm. After two months, the by-products of vermicomposting were collected. The vermicompost tea was prepared with the bubbler bucket method using the recommended formulations of vermicompost and water (1:10 v/v) brewed for one day at room temperature (Torres *et al*., 2021).

Operation of OFB

The operation of the OFB was divided into two stages. The first stage involved the inoculation of beneficial microorganisms present in vermicompost tea into the system. The OFB was filled with 95 L of water. Five liters of produced vermicompost tea was added to OFB. The OFB was operated for fifteen days with no additional organic fertilizer and water to establish microorganisms in the biofilm media. The second stage involved the bioprocessing of the selected organic fertilizer. The OFB was loaded with 1kg of dried chicken manure based on 10g of chicken manure per liter of water (Tikasz *et al.*, 2019). The organic fertilizer bioreactors were operated for 28 days for the bioprocessing of dried chicken manure to produce OLS. Daily inspection and monitoring on both stages were done to ensure the smooth operation of the system.

Experimental set-up and sample collection

The experimental setup consisted of three fabricated OFBs served as replication in the study. The OFBs were placed beneath the soil and retained the top portion for the accessibility of placing materials needed for the bioprocessing of dried chicken manure and measurement of the operating parameters. The OFBs were operated under greenhouse conditions (temperature ranging from 23 – 35 ^{o}C) in the soil and water laboratory of the College of Engineering, Central Luzon State University, Philippines. The operation of the OFBs started after the conduct of preliminary testing and 15 days after the application of vermicompost tea in the system. Each OFB was loaded with 1 kg of DCM and placed inside the feedstock vessel. After loading, the bioprocessing stage of DCM into OLS started which lasted for 28 days. Four different HRPs (7 days, 14 days, 21 days, and 28 days) which serve as a treatment in the study were evaluated to assess the performance of OFB and to determine the effects of varying HRP on the physiochemical characteristics and phytotoxicity of produced OLS. No additional DCM and water were added during the bioprocessing stage. Two samples (250 mL) of OLS were collected at OFB during each HRP to assess the nutrient content and phytotoxicity of the produced OLS on each treatment.

Monitoring parameters and measurement

Dissolved oxygen (DO) level, electrical conductivity (EC) level, pH level, temperature, and total dissolved solids (TDS) were monitored daily between 7:00 – 8:00 AM and 1:00 – 2:00 PM throughout the bioprocessing period of feedstock using the developed OFB. A sample of 300mL was collected from the discharge channel of the bioreactor tank for the measurement of each parameter. DO level was measured using a portable DO meter (polarographic type DO probe, Lutron). EC level and TDS were measured using pen-type EC and TDS meter (AP-2 conductivity tester and AP-1 TDS tester, HM Digital). PH level was measured using a pH meter (hand-held pH meter, Hanna Instrument). The temperature was based on temperature readings from the DO meter and measured approximately 5 cm below the surface of OLS in the bioreactor tank. After measurement, the sample was returned to the OFB. Daily calibration was done on the DO meter before measurement while weekly calibration was done on the pH meter. EC and TDS meters were calibrated once and used throughout the monitoring period. The calibration methods of each instrument were based on the manuals provided by the manufacturer.

Physiochemical analysis of feedstock and produced OLS

The nutrient content of dried chicken manure and produced OLS at different HRPs using OFB were measured by an accredited laboratory

(Regional Soils Laboratory of Department of Agriculture – Regional Field Office III and Eminent Water Laboratory Center, San Fernando City, Pampanga, Philippines). The gravimetric method was used to determine the moisture content of the feedstock. The potentiometric method was used to calculate the pH. The Kjeldahl method was employed to estimate the total nitrogen in the sample. Total phosphorus and total potassium were determined using acid digestion, vanadomolybdate method, and flame atomic emission spectroscopy respectively. Nitrate content was measured using the cadmium reduction method. The Walkley-Black method (titrimetric) was employed to calculate organic carbon and organic matter.

Seed germination bioassay

A seed germination bioassay was conducted to determine the phytotoxicity of dried chicken manure and the produced OLS at different HRPs (7 days, 14 days, 21 days, and 28 days) which served as a treatment with three replicates of each treatment. The assays for organic fertilizer input were prepared by dissolving dried chicken manure with distilled water (1:10, w/v in g/mL) and shaking for 15 mins (Araujo & Monteiro, 2005; Ravindran *et al.*, 2014). The seed germination bioassay was based on the methods conducted by Tam and Tiquia, (1994). The filter paper was placed inside a 15 x 90 mm sterilized petri dish and wetted with 5mL of dissolved dried chicken manure in distilled water and samples of organic liquid solution from different HRPs. The same procedure was done for the control with distilled water. Ten seeds of cucumber (*Cucumis sativus*) were placed on the filter paper and incubated in the room without light at room temperature (20 - 25 ^{o}C) for five (5) days. Relative seed growth (RSG), relative root elongation (RRE), and germination index (GI) were computed with the following formula:

$$\mathrm{RSG}\ (\%) = \frac{\text{seeds germinated in samples}}{\text{seed germinated in control}} \times 100$$

$$\mathrm{RRE}\ (\%) = \frac{\text{average root length in sample}}{\text{average root length in control}} \times 100$$

$$\mathrm{GI} = \frac{\mathrm{RSG}\ \times \mathrm{RRE}}{100}$$

Statistical analysis

Repeated measures analysis of variance was utilized as statistical analysis in the study to determine whether the difference in HRPs of organic fertilizer in the system was significant in terms of the physiochemical characteristics and phytotoxicity of produced OLS. Holm method post hoc test was used to test significance among treatment means at a level of 0.05.

Results

Description and specification of the developed OFB

The function of OFB has been shown to extract the available nutrients available in feedstock introduced in the system and generated nitrate which was a preferable form of nitrogen for plants to assimilate. The OFB consisted of five major components, an OLS tank, filtration tank, bioreactor tank, aeration system, and supply and discharge channels as shown in Figure 3. The OLS tank held the dried chicken manure inside the vessel with a filter net within the bioprocessing period. The filtration tank separated the solid particulates in the OLS. The bioreactor tank processed the OLS with the aid of microorganisms present in the media to transform organic nitrogen present in the OLS into nitrate. The aeration system supported a continuous supply of dissolved oxygen in the OLS tank and bioreactor tank. The supply and discharge channel facilitated the recirculation of OLS in the system using water level flow and airlift pumps. The specification of the designed OFB is shown in Table 1.

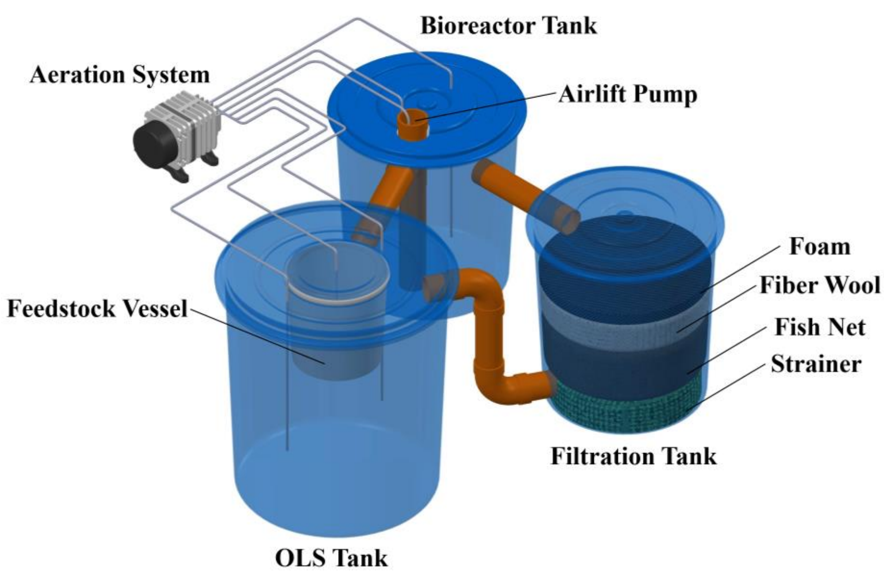

Figure 3. Design of organic fertilizer bioreactor for production of organic liquid solution

Table 1. Specification of the developed organic fertilizer bioreactor

Components	Specification
Overall structure of OFB	Dimension - 92 cm x 96 cm x 70 cm
	Weight - 22 Kgs
	Capacity - 100 Liters
OLS tank	Dimension - 40 cm dia., 55 cm height
	Feedstock capacity - 4 kgs organic fertilizer
Filtration Tank	Dimension - 36 cm dia., 40 cm height
	Consist of fiber wool, aquarium foam and layers of fish nets
Bioreactor Tank	Dimension - 36 cm dia., 40 cm height
	Consist of K1 media with specific surface area of 500 m^2/m^3
Aeration System	Airflow rate – 65 L/min equally distributed to OLS tank and bioreactor tank
	Power rating – 35 Watts
Airlift Pump System	Liquid flow rate - 3.58 $\pm$0.07 L/min

Physiochemical Characteristics of Feedstock

Table 2. Physiochemical properties of dried chicken manure used as feedstock in organic fertilizer bioreactor for production of organic liquid solution

Properties	Values
pH level	10.40
C/N ratio	12:1
Moisture content (MC)	31.77 %
Organic matter (OM)	27.09 %
Organic carbon (OC)	15.71 %
Total nitrogen (N)	1.30 (1.91^{db}) %
Total phosphorus (P_2O_5)	3.56 (5.22^{db}) %
Total potassium (K_2O)	3.38 (4.95^{db}) %

Note: db – weight percentage in dry basis

Result showed (Figure 4) the physiochemical characteristics of feedstock used in OLS production using OFB at different HRP (Table 2). The chicken manure processed in the OFB falls within the alkalinity range in terms of pH. The lower C/N ratio of DCM indicated the maturity of fertilizers ready to be used on soil fertilization. The presence of organic matter indicated the presence of microorganisms and organic residues as potential nutrient sources. In terms of nutrient content, the total NPK content of the DCM is 8.24% (12.08% on dry basis).

Performance of OFB in terms of maintaining the monitoring parameters at varying HRP

It showed the trend of different monitoring parameters within the processing period of feedstock using OFB (Figure 4). It is observed that there is an increase in the DO level of OLS from day 1 (6.05 mg/L) after the loading of chicken manure compared to day 28 (6.75 mg/L). The aeration system of the

OFB was able to maintain the optimum DO level (4mg/L and above) for the nitrification process. In terms of pH level and temperature, there was no observed large value of fluctuation over the bioprocessing period (Figures 4b and 4c). The OFB also maintained the optimum pH level and temperature for nitrification. It increased in EC level and TDS of OLS within the period of processing as shown in Figures 4d and 4e. The increasing observation indicated the continuous degradation of dried chicken manure in the feedstock vessel.

Result showed the effect of varying HRP on the DO level, pH level, temperature, TDS, and EC level of OLS produced using the developed OFB (Table 3). There was no significant difference in DO level ($p = 0.10$), pH level ($p = 0.06$) and temperature ($p = 0.25$) of OLS produced at different HRPs. However, there was a significant effect on the EC level ($p = 0.015$) and TDS ($p = 0.006$) of OLS produced at different HRPs. In terms of EC level OLS produced at 21 and 28 days HRP was significantly higher values compared to OLS produced at 7 days. OLS produced at 14, 21, and 28 days HRP were significantly higher in terms of TDS compared to OLS produced at 7 days HRP.

Performance of OFB in terms of Nutrient Content of OLS produced at varying HRP

It showed the nutrient content of OLS produced at different HRPs. The varying HRP significantly affects nitrate ($p = 0.002$) and total potassium ($p = 0.016$) content of OLS using OFB (Figure 5). However, there was no significant effect on the total nitrogen ($p = 0.07$) and total phosphorus ($p = 0.12$) of OLS produced at varying HRPs using OFB. An increasing trend of total potassium and decreasing trend of nitrate content of OLS were observed between HRP of OLS. The nitrate content of OLS produced at 7 and 14 days HRP was significantly higher values compared to OLS produced at 21 and 28 days HRP. In terms of total potassium content, OLS produced at 7 days HRP was shown to be lower value compared to OLS produced at 21 and 28 days HRP.

Table 3. Effect of different hydraulic retention periods on the dissolved oxygen level, pH level, temperature, total dissolved solids, and electrical conductivity of organic liquid solution using organic fertilizer bioreactor

Treatment (Hydraulic Retention Periods)	DO Level (mg/L)	pH Level	Temperature (°C)	EC Level (µS/cm)	TDS (ppm)
7 days	6.67 ± 0.02[a]	8.88 ± 0.08[a]	30.60 ± 0.08[a]	2,061.33 ± 19.16[a]	936.67 ± 1.36[a]
14 days	6.73 ± 0.02[a]	8.83 ± 0.03[a]	30.33 ± 0.06[a]	2,096.33 ± 4.76[ab]	946.50 ± 1.32[b]
21 days	6.75 ± 0.03[a]	8.82 ± 0.02[a]	30.23 ± 0.15[a]	2,119.67 ± 0.88[b]	949.00 ± 1.80[b]
28 days	6.78 ± 0.03[a]	8.75 ± 0.03[a]	30.28 ± 0.15[a]	2,124.17 ± 4.21[b]	948.33 ± 1.76[b]

Note: Mean ± represents standard error. Different letters show statistically significant differences at p-value = 0.05

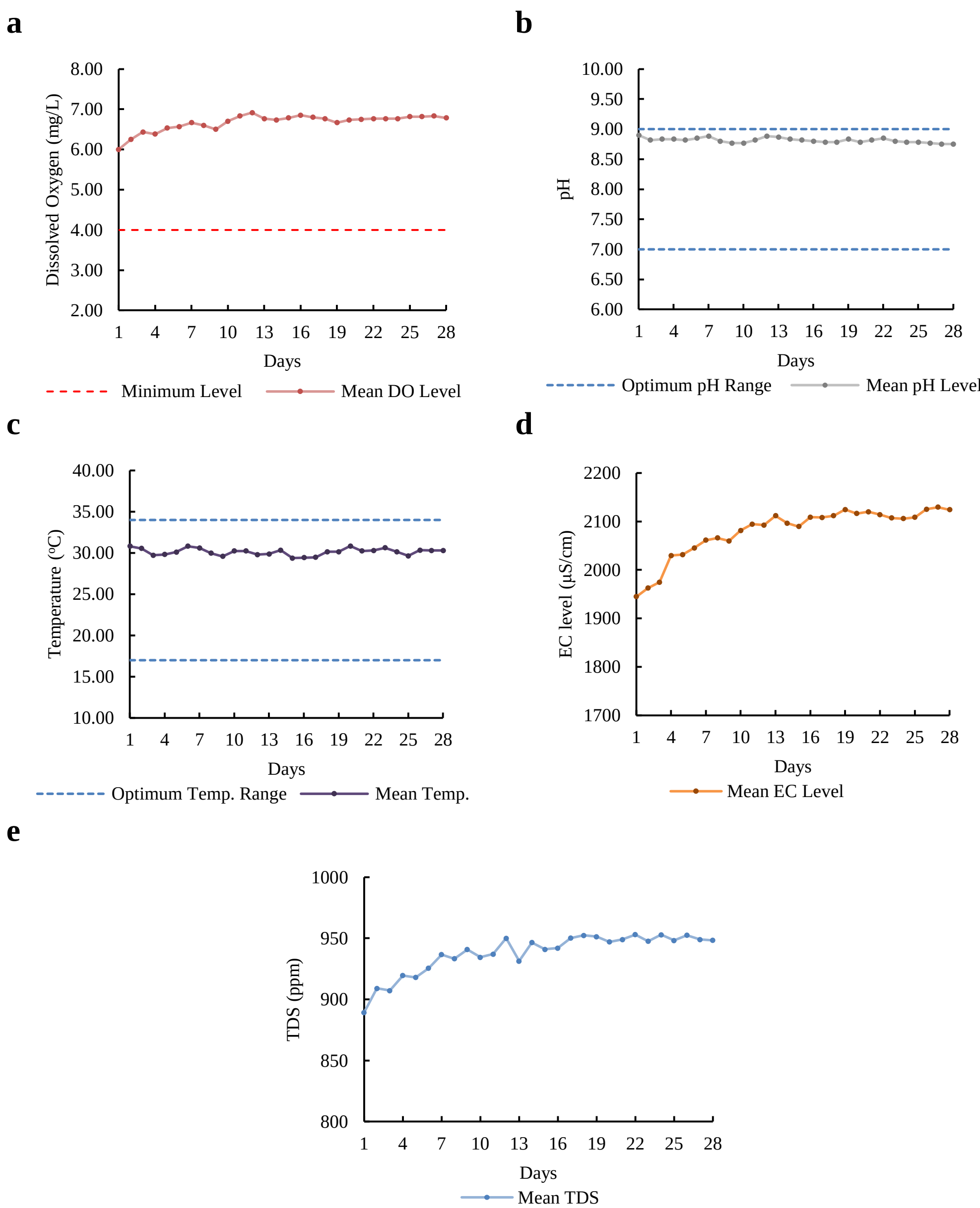

Figure 4. Trend of daily mean values of dissolved oxygen level (a), pH level (b), temperature (c), total dissolved solids (d), and electrical conductivity (e) of organic liquid solution using organic fertilizer bioreactor

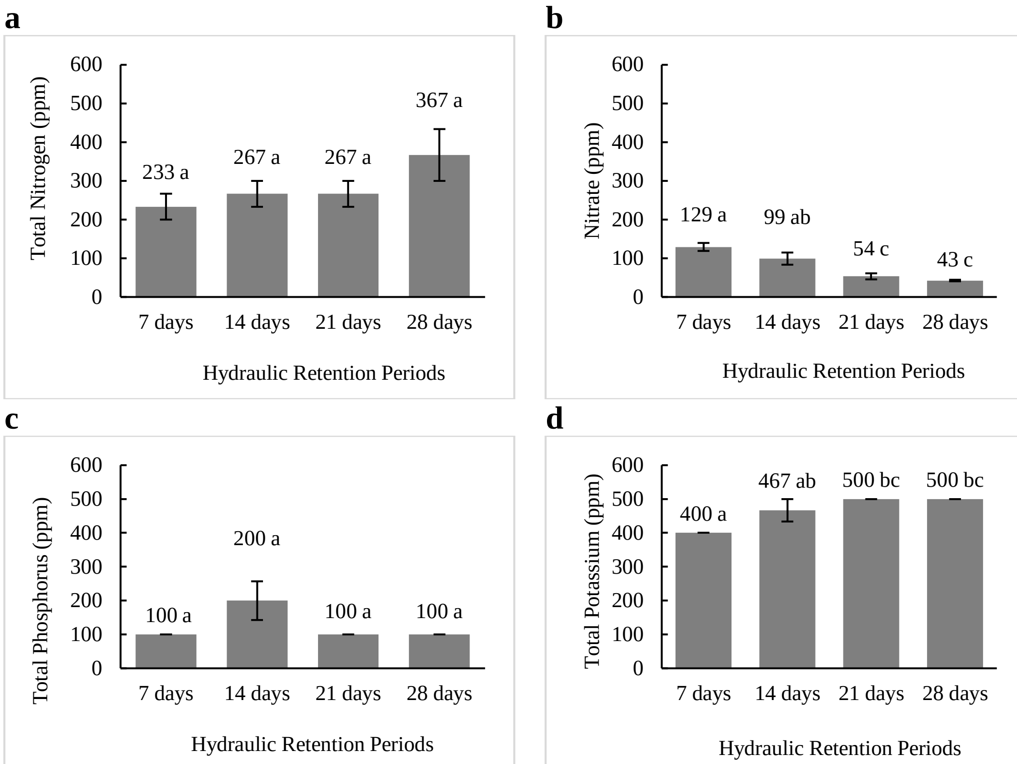

Figure 5. Quality of organic nutrient solution produced at different hydraulic retention periods using organic fertilizer bioreactor. Total Kjeldahl Nitrogen (a), Nitrate (b), Total Phosphorus (c), and Total Potassium (d). Different letters indicate statistically significant differences ($p < 0.05$). The error bar represents the standard error of the mean

Performance of OFB in terms of phytotoxicity of OLS produced at varying HRP

Result showed the parameters for assessing the phytotoxicity of OLS produced at varying HRP using OFB (Table 4). The unprocessed dried chicken manure (control) was significantly lower value of germination index, relative seed germination and relative root elongation of cucumber seeds as compared to OLS produced at different HRP ($p < 0.001$). On the contrary, there was no statistically significant variation in phytotoxicity indicators between OLS produced at four different HRPs.

Table 4. Relative seed germination, relative root elongation, and germination index of cucumber seeds using organic liquid solution produced at different hydraulic retention periods using organic fertilizer bioreactor

Treatment (Hydraulic Retention Periods)	Relative Seed Germination (%)	Relative Root Elongation (%)	Germination Index
Control	85.56 ± 1.11[a]	30.45 ± 0.93[a]	26.05 ± 0.81[a]
7 days	100.00 ± 0.00[b]	119.97 ± 2.76[b]	119.97 ± 2.76[b]
14 days	100.00 ± 0.00[b]	117.12 ± 4.16[b]	117.12 ± 4.16[b]
21 days	98.89 ± 1.11[b]	113.49 ± 1.44[b]	112.20 ± 0.89[b]
28 days	100.00 ± 0.00[b]	114.27 ± 2.59[b]	114.27 ± 2.59[b]

Note: Mean ± represents standard error. Different letters indicates statistically strong significant differences at p value < 0.001.

Discussion

Principle of Operation of the OFB

The purpose of OFB is to extract the nutrients present in the feedstock and to generate nitrate, which is the preferred form of nitrogen for crops to assimilate. The principle of operation of the OFB follows the nitrification process to valorize the nutrient-rich organic fertilizer such as chicken manure into an organic liquid solution rich in nitrate. Nitrification is the method of transforming nitrogen from its most reduced form, ammonia, to its most oxidized state, nitrate. This process is done in two stages by two different types of microbes, the ammonia-oxidizing microbes or archaea, and the nitrite-oxidizing microbes (Cáceres *et al*., 2018). The bioreactor tank through K1 bio media will serve as a medium for the colonization of nitrifying bacteria to convert a reduced form of nitrogen present in the OLS into nitrate. The aeration system will deliver the needed oxygen by the bacteria to oxidize ammonia present in the organic fertilizer into nitrate. The developed OFB has successfully extracted nutrients from DCM. The system was also able to produce nitrate from DCM.

Physiochemical Characteristics of Feedstock

In terms of the C/N ratio, the value of feedstock is slightly higher compared to the recommended range for nitrogen mineralization of organic fertilizer which was below 11 (Shinohara *et al*., 2011). The total nitrogen of chicken manure used was within the range of reported values of nitrogen content ranging from 1.5 – 8.7% (Wedwitschka *et al*., 2020). The total phosphorus and total potassium of the feedstock were slightly above the reported value of 2.95 % (Dady *et al*., 2021) and 2.5 ± 1.2% (Aksorn *et al*., 2022). In general, the physiochemical characteristic of the feedstock agrees with other reported values in the research utilizing chicken manure as a plant nutrient source.

Maintaining monitoring parameters at varying HRP

Dissolved Oxygen Level - Maintaining DO level above the critical level is necessary to ensure the efficient oxidation of ammonia into nitrate by nitrifying bacteria present in the surface media of OFB. DO levels ranging from 0.3 mg/L to as high as 4.0 mg/L have an impact on nitrification reactions in wastewater treatment facilities and in some cases that DO level greater than 4.0 mg/L is necessary to achieve optimum nitrification rates (Stenstrom and Poduska, 1980). In other studies, low DO levels (0.3 and 1.2 mg/L) inhibited the nitrification process in the aerated treatment pond system (Jechalke *et al*., 2011). On the other hand, introducing a high DO concentration (38 mg/L) in a laboratory fermenter treating sewage sludge does not inhibit the nitrification process (Charley *et al*., 1980). The OFB maintained the above minimum DO level for optimum nitrification of 4.0 mg/L and above within 28 days of the bioprocessing period. The result suggested that the designed aeration system of OFB was able to maintain a desirable DO level for the bioprocessing of chicken manure to produce OLS.

pH Level - One of the most critical environmental conditions that influence nitrifying bacteria activity is pH. In addition, nitrification in various biological systems is sensitive to pH fluctuations. The optimum pH level for nitrification can range from 7 to 9 with the optimum pH range from 7.2 – 8.8 for ammonia-oxidizing bacteria (AOB) and 7.2 to 9.0 for nitrite-oxidizing bacteria (NOB) (Chen *et al*., 2006). For plant application, the optimal pH level range in hydroponics system are between 5.5 to 6.5 and 6.5 to 7.0 for aquaponics (Tyson *et al*., 2004). The OFB was able to maintain the pH level (average of 8.78) within the optimum range pH level favorable for nitrifying bacteria as shown in Figure 4b, however, the pH value of OLS suggests it is not optimal for plant applications. On the other hand, lettuce cultivated on a hydroponics system utilizing processed chicken manure using a biofilter successfully flourished with a pH level of above 8.0 (Aksorn *et al*., 2022; Wongkiew *et al*., 2021).

Temperature - In most of the studies on bioreactors, the process of nitrification is substantially influenced by temperature. The optimum temperature range for nitrification varies depending on the type of environment. In the biofilter of the aquaponics system, the optimal temperature range for nitrifying bacteria is between 17 ℃ and 34 ℃ (Somerville *et al*., 2014). Other reports suggested that a temperature below 15 or above 32 would inhibit the activity of nitrifying bacteria (Qiao *et al*., 2008). However, as a result of the study on the effect of varying temperatures (19 ℃ to 32 ℃) in processing industrial wastewater using a moving bed biofilm reactor, the suggested optimum operating parameter is 27 ℃ (Majid and Mahna, 2019). The OFB operates within the optimum range of temperature needed to function the nitrifying bacteria as shown in Figure 4c. The placement of OFB beneath the soil could help to prevent the sudden fluctuation of temperature of OLS.

EC level and TDS - EC level and TDS serve as indicators of dissolved nutrients in the water from the organic nutrient source. EC levels of liquid fertilizer derived from agricultural waste varies in most of the literature depending on the dilution rate, type of organic nutrient sources, methods of production, and the ratio of water to organic fertilizer. The reported EC level of the produced fermented organic liquid fertilizers derived from varying ratios of agricultural residues and waste from factories ranges approximately between 1,500 to 3,500 μS/cm within a 28 day period (Phibunwatthanawong & Riddech, 2019). The EC values derived from aerated animal manure extracts used as plant nutrient source in hydroponics ranges between 550 to 1620 μS/cm (Tikasz *et al*., 2019). On the other hand, the reported TDS level of plant-based organic liquid manure fermented for 30 days ranges between 970 and 1844 ppm (Govere *et al*., 2011).

In terms of the effect of HRP on the EC level and TDS of OLS, the observed increasing pattern in the value agrees with other studies on the effect of time on the quality of organic liquid fertilizer. The effect of storage time significantly affects the EC level and TDS of municipal solid waste compost tea and a positive correlation between storage time and EC level and TDS was also observed (Vehniwal *et al*., 2020). The increasing trend of both parameters could be attributed to microbial activity in the media, slow release of nutrients from feedstock through steeping, continuous liquid movement, and liquid agitation due to the aeration system of OFB. The result suggests that at 14-day HRP and above, the OFB was able to extract a significant amount of nutrient present in the feedstock.

Nutrient Content of OLS produced at varying HRP using OFB

In scientific literature, chicken manure contains a significant amount of macronutrients that could support plant nutrient needs. The method of processing chicken manure to convert as organic fertilizer varies from physical, biological, and chemical methods. Aerated chicken manure with varying formulations (10g/L, 25g/L, and 50g/L) contains 0.5 – 2.5 ppm nitrate (Tikasz *et al*., 2019). Organic nutrient solution derived from chicken manure (20g/L) used as a plant nutrient source in hydroponically grown maize contains 184 ppm nitrogen, 59 ppm phosphorus, and 142 ppm potassium (Adeyemi *et al*., 2021). In comparison with widely used inorganic nutrient solution, Hoagland no. 2 nutrient solution contains 220 ppm nitrate, 24 ppm phosphorus, and 230 ppm potassium (Hoagland and Arnon, 1950).

The NPK content of the produced OLS at different HRPs using OFB was higher compared to other methods of nutrient extraction from chicken manure reported. The OFB was also able to generate nitrate which is the preferable nitrogen form of the plant compared to ammonia. In comparison with the inorganic nutrient solution, the NPK content of OLS was higher, however, the nitrate produced in OFB is not at par with the nitrate level of the inorganic nutrient solution. The reducing trend of nitrate could be attributed to the aerobic denitrification process in OFB. Numerous species of bacteria can engage in the process of denitrification, which uses nitrate as an oxidizing agent in an aerobic environment (Rajta *et al*., 2020). To utilize the nitrate produced using OFB, the OLS should be collected between 7 days and 14 days HRP. Future studies could be done on OFB using different volume feedstock to be loaded at optimal HRP.

Phytotoxicity of OLS produced at varying HRP using OFB

The germination index is a highly responsive metric that is used to assess the phytotoxicity of both solid and liquid biofertilizer. A germination index of more than 80% implies that the manure is safe of phytotoxicity, whereas index value of less than 80% suggests that the manure could be phytotoxic to plants (Barral and Paradelo, 2011). Germination indices of less than 50% have reportedly been found to be unsatisfactory for use in crop production (Ravindran *et al*., 2014). Unprocessed animal manure has a higher concentration of intermediary substances like ammonia and organic acids that causes toxicity to plants and have an impact on germinating seeds and root (Phibunwatthanawong and Riddech, 2019). The reported germination index of diluted 100% chicken manure extract (1g per 10mL, w/v) ranging from 1.9 to 2.6%, and in terms of reduced dilution percentage (50%) resulted in germination index, relative root elongation and seed germination lower than 80% using collard green seeds (Kebrom *et al*., 2019).

Based on the result of the phytotoxicity test, the produced OLS at different HRPs using OFB is above the phytotoxic range. The results suggested that the OFB was able to reduce the phytotoxicity of dried chicken manure with a minimum HRP of 7 days. Values above 100% of relative root elongation using OLS at different HRPs suggested that the nutrient content present in the OLS was able to stimulate roots in comparison with cucumber seed germinated using distilled water.

Generalization

The organic fertilizer bioreactor (OFB) was developed, fabricated, and evaluated to establish an optimum hydraulic retention period (HRP) on the bioconversion of dried chicken manure to produce an organic liquid solution (OLS) as a potential plant nutrient source. The result of the evaluation suggests that the optimum HRP for bioprocessing of dried chicken manure using OFB is between 7 days and 14 days. Within the optimal HRP, the OFB was able to maintain the optimal operating parameters for bioconversion of dried chicken manure to OLS such as dissolved oxygen (6.67 – 6.73 mg/L), pH (8.83 – 8.88), electrical conductivity (2,061 – 2,096 μS/cm) and total dissolved solids (937 – 947 ppm). In terms of nutrient content, the OLS produced at optimal HRP contains total nitrogen of 233 – 267 ppm, nitrate of 99 – 129 ppm, total phosphorus of 100 – 200 ppm, and total potassium of 400 -467 ppm. Successful production of nitrate suggests that the OFB was able to convert other forms of nitrogen present in the chicken manure such as organic nitrogen and ammonia. Seed germination bioassay suggests that the OLS produced at optimal HRP using OFB does not inhibit the germination of the cucumber seeds compared to dried chicken manure only. The result of the study provided a new methodology for processing animal manure to produce organic liquid fertilizer that could be used as a plant nutrient source alternative to commercial fertilizers. In addition, the result of the study can aid in the sustainability of nutrient reuse from animal waste, specifically chicken manure for OLS production as an alternative to chemical-based fertilizer. The OFB could also be applied as an alternative waste processing technology for poultry farms aiming for a bio-circular economy.

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