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Development of Small-scale Integrated Hydroponics—Animal Waste Bioreactor (AWB) for Romaine Lettuce (*Lactuca sativa L. var. longofolia*) Production

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ABSTRACT

As improper processing and disposal of animal waste cause negative impacts on the environment, the animal industry sector must shift to more sustainable practices to lessen these effects. Recently, the application of the circular economy concept in agriculture, using animal waste as part of nutrient cycling, has emerged as a sustainable approach. The study aims to develop and test the small-scale integrated hydroponics-animal waste bioreactor (AWB) for romaine lettuce production using chicken manure tea (CMT) derived from dried chicken manure as a primary nutrient source. Three integrated hydroponics-AWB systems, with varying concentrations of CMT at 1,000 ppm, 1,200 ppm, and 1,400 ppm total dissolved solids (maintained within an upper and lower bound of 50 ppm), were constructed, tested, and compared to conventional hydroponics that used a nutrient solution maintained at 1,000 ppm TDS. The test result suggests that the ideal concentration of CMT in the system is 1,000 ppm. Within the optimum manure tea concentration, the small-scale integrated hydroponics-AWB produced romaine lettuce with growth parameters comparable to conventional hydroponics. In addition, increasing the CMT concentration to 1,400 ppm negatively impacts the plant growth parameters of romaine lettuce. The developed small-scale integrated hydroponics-AWB system provides a viable approach for growing lettuce using animal waste as the major source of nutrients. The developed production system could help

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mitigate the negative environmental effects of improper handling and disposal of animal waste and dependence on chemical-based nutrient solutions in hydroponic crop production.

Keywords: Bioponics, chicken manure tea, mineralization, nutrient cycling, soilless culture, sustainable crop production

INTRODUCTION

The continuous increase of agricultural waste globally can be attributed to several factors. Due to the continuous increase in population worldwide, the demand for food from agriculture is also increasing (Valin et al., 2014). To meet this demand, farmers are growing more crops and raising more animals, which leads to increased agricultural waste (Borlaug, 2019; Bradford, 1999; Spiertz & Ewert, 2009). Moreover, the increase in agricultural waste could be attributed to industrialization, which uses artificially synthesized fertilizers, insecticides, and other chemical substances that augment crop productivity (Horrigan et al., 2002). Furthermore, problems with animal waste from the poultry and livestock sector resulted in nutrient pollution problems in land and water (Hu et al., 2019; Khoshnevisan et al., 2021).

Problems in agricultural waste could be solved by implementing sustainable crop production using farming technologies to improve crop yield and quality, as well as by reducing synthetic input. Hydroponics is deemed to be a sustainable method of crop production due to various environmental and economic benefits such as low water use, reduced pesticide applications, precise nutrient supply, space efficiency, and higher yield per area compared to conventional farming methods (De Clercq et al., 2018). However, the excessive use of chemical fertilizers as plant nutrient sources still contributes to soil, water, and air pollution, causing environmental and health problems (Savci, 2012). Moreover, manufacturing and transporting chemical fertilizers use fossil fuel-derived energy, contributing to net greenhouse gas emissions (Koga, 2008).

Due to its environmental benefits, there is a growing interest in using agricultural waste-derived fertilizer as an alternative plant nutrient source in a hydroponics system. Animal manure is one example of agricultural waste containing significant nutrients that could support crop growth. Although animal manure is a valuable soil amendment for crop production, improper management, processing, and utilization could negatively impact the environment and plants (Khoshnevisan et al., 2021; Ren et al., 2022). Due to the limited management of agricultural waste, there is an urgent need to implement a solution for the utilization and valorization of agricultural waste for sustainability, food security, and health security (Koul et al., 2022).

Several studies were conducted to utilize animal waste as a nutrient source in hydroponics as an alternative to the chemical-based nutrient solution. Bioprocessed animal waste using a biogas digester (digestate) was utilized as a substitute nutrient source in hydroponics. However, crop growth using different digestate and nutrient solution ratios was negatively impacted compared with conventional hydroponics (Mupambwa et al., 2019). In addition, with the use of aerated chicken and cow manure extract as a plant nutrient source in the ebb and flow hydroponics, the recorded aboveground wet and dry weights were lower compared to conventional hydroponics (Tikasz et al., 2019). Both studies suggested that low plant growth performance could be attributed to ammonia toxicity, which is the least preferable form of nitrogen compared to nitrate.

Chicken manure is a widely used organic fertilizer in agriculture due to its high nutrient content, such as nitrogen (N), phosphorus (P), and potassium (K) essential for plant growth (Waldrip et al., 2020). The typical nutrient analysis for chicken manure can vary depending on several factors, such as the bird's diet, age, and moisture content. A typical chicken manure on a dry basis (i.e., broiler and layer chicken manure as excreted) can contain around 5.0 and 5.4 % total Kjeldahl nitrogen, 1.29 and 1.32 % total ammonia nitrogen, 3.08 and 4.20 % total phosphorus, and 2.31 and 2.40 % total potassium (Ashworth et al., 2020; Collins et al., 1999). It also contains secondary and micronutrients like calcium, magnesium, sulfur, zinc, and copper. These nutrients fulfill specific roles in plant metabolic processes, whether soil-based or hydroponic. Chicken manure requires proper treatment and management to maximize its utility as a plant nutrient source. Fresh chicken manure contains high levels of ammonia, which can be phytotoxic.

In our recent study, we successfully designed an animal waste bioreactor (AWB) capable of producing significant amounts of nitrate within a 7 to 14-day bioprocessing period (Torres et al., 2023). Integrating our designed animal waste bioreactor into hydroponics could be a viable solution for using animal waste as a nutrient source. Thus, the present study aims to achieve the following objectives: to develop a small-scale integrated system of hydroponic system and animal waste bioreactor (AWB) for romaine lettuce production and to investigate the effect of varying concentrations of manure tea on the growth parameters of romaine lettuce compared to convention hydroponics system utilizing the commercial nutrient solution.

Integrating hydroponics systems and AWB can provide a more sustainable alternative plant nutrient source while reducing the negative impact of improper animal waste management on the environment. The results of this study not only offer valuable data for farmers and scientists but highlight the potential of circular economy principles to create more sustainable and efficient systems in the agricultural sector through nutrient cycling. Furthermore, the results of this study may have implications for other crops and different scale systems, providing valuable information in utilizing animal waste as a plant nutrient source.

METHODS

Location of the Study

The development and performance evaluation of an integrated hydroponics-AWB were conducted at the Land and Water Laboratory of the College of Engineering at Central Luzon State University in the province of Nueva Ecija, Philippines, from November 2022 to January 2023. The bioreactor system was assembled in a dome-type greenhouse, covered with a layer of polyethylene film and a 50% shade net to create an optimal growing environment. The greenhouse was 12 × 6 × 6 m (length × width × height) and lacked roof vents but was equipped with side screens to allow for proper airflow. The greenhouse also had a 40-cm masonry bottom, which served as the foundation for the system. Temperature and humidity inside the greenhouse were monitored using a humidity-temperature data logger (Benetech, GM1365) during the production phase of the study.

Design Concept of the Small-scale Integrated Hydroponics-AWB

The design concept for developing a technology for romaine lettuce production utilizing dried chicken manure (DCM) as a main source of nutrients consists of two major components: the hydroponics system and the AWB. Each component is composed of different parts to work as an integrated system, as shown in Figure 1.

The hydroponics system is composed of nutrient film technique (NFT) grow bed channels, a water delivery channel, a water discharge channel, and a support frame. The NFT grow bed channels are the primary component of the hydroponic system and are responsible for providing a space for the plants to grow. These channels are made of PVC

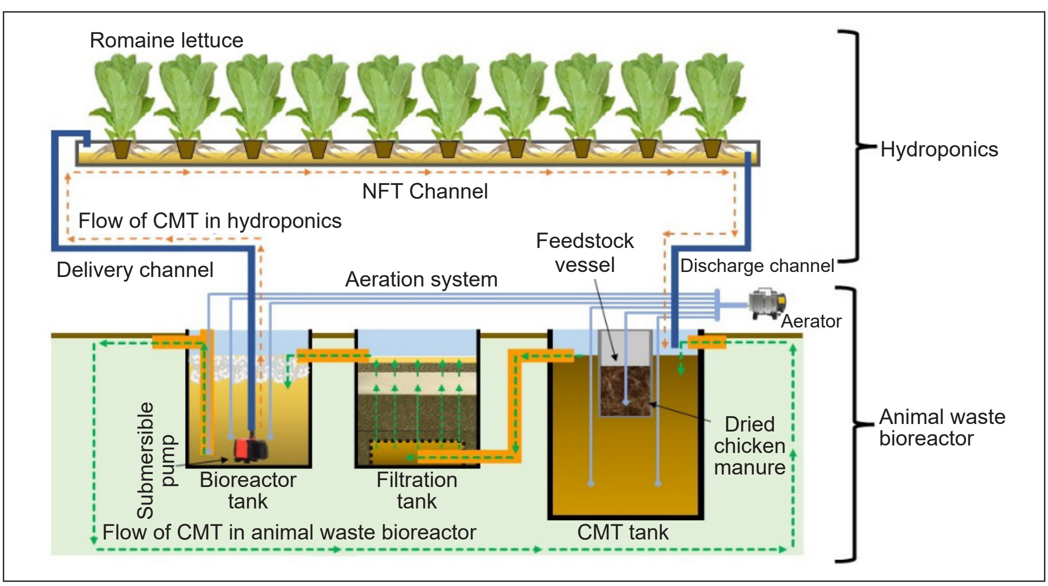

Figure 1. Schematic layout of the integrated hydroponics—animal waste bioreactor

pipe with a diameter of 5.08 cm and a length of 152 cm, providing an area for the plants to grow. The channels are designed with 10 evenly spaced holes, each measuring 5.08 cm in diameter and 15 cm apart, to ensure that the plants receive optimal water, nutrients, and sunlight.

The water delivery and discharge channel consists of a pipe, fittings, pump, and valve responsible for the flow of CMT from the AWB on the NFT grow bed channel. The water delivery channel consists of a PVC pipe with a diameter of 1.90 cm that connects the submersible pump (20 L/min), a gate valve to control the flow of water, and a 2 mm diameter flexible hose to distribute the flow of liquid fertilizer in each NFT grow bed channel. The water discharge channel is responsible for removing any excess water and debris. The support frame is made of slotted angle bars to hold the channels in place and provide structural stability to the system.

The AWB is composed of a bioreactor tank, CMT tank, filtration tank, water circulation channels, and aeration system. The CMT tank, made of a plastic container (50 L capacity, 40 cm diameter, 55 cm height), holds the DCM in a filter-lined vessel (200 microns) to slowly release nutrients into the water during the bioprocessing period. The filtration tank, made of a plastic container (35 L capacity, 36 cm diameter, 40 cm height), separates solid particulates in the CMT using perforated plastic, fishnet (1 m × 2.5 m with 1 mm grid size), fiber wool (160 g), and aquarium foam (30 PPI, 2.5 cm thickness) to filter the CMT.

The bioreactor tank processed CMT with nitrifying bacteria using K1 biofilm media (500g) suspended by diffused air from the aeration system. K1 biofilm media is a cylindrical polyethylene plastic (10 mm diameter, 0.5–0.8 mm thickness, 0.123 g/L density, 500 m^2/m^3 specific surface area) which serves as a carrier for nitrifying bacteria to convert ammonia present in CMT into nitrate. The aeration system supports a constant supply of dissolved oxygen in the CMT tank and bioreactor tank by using an air pump (35 Watts, rated airflow of 65 L/min at 0.027 MPa) to maintain a dissolved oxygen level of at least 2 mg/L or above. It also serves as a CMT tank and bioreactor unit mixing mechanism. The circulation of CMT in the system is facilitated using PVC pipes and fittings and an airlift pump, which recirculates the CMT without the need for an additional water pump.

Experimental Setup of the Study

The experiment was designed to test and compare the effect of varying concentrations (in terms of total dissolved solids in ppm) of CMT on the growth and yield of romaine using the developed integrated hydroponics-AWB system and a hydroponics system utilizing a commercial nutrient solution. The experimental setup consisted of four closed-loop hydroponics systems, which included three integrated hydroponics-AWB with CMT total dissolved solids (TDS) levels controlled to fall within the ranges of 1000 ± 50 ppm (CMT1), 1200 ± 50 ppm (CMT2), and 1400 ± 50 ppm (CMT3). In addition, one hydroponics system was maintained using a commercial nutrient solution as a control (CNS), with TDS levels maintained within the range of 1000 ± 50 ppm based on the recommended range between 800 and 1,200 ppm (Sace & Jr Natividad, 2015). Each setup consists of five NFT channels with ten plants, each with 50 plants per treatment.

Operation of the Small-Scale Integrated Hydroponics-AWB

The operations of the integrated hydroponics-AWB were divided into three phases. Phases 1 and 2 were based on recently conducted methods to produce CMT using AWB (Torres et al., 2023). Phase 3 involves the production stage using the developed system. In Phase 1, the AWB was inoculated with beneficial bacteria from vermicompost tea. The vermicompost tea was prepared using a 1:10 ratio of vermicompost to water and brewed for 24 hours at room temperature using the bucket bubbler method. Five (5) liters of the produced vermicompost tea were added to the AWB containing 105 liters of water. The system was operated for 15 days without additional DCM or water to establish microorganisms in the biofilm.

Phase 2 of the operation involved bioprocessing DCM to produce a nutrient-rich CMT as a source of nutrients for romaine lettuce. It was accomplished using AWB, loaded with varying amounts of DCM to achieve specific total dissolved solids (TDS) ranges for each treatment. Initially, CMT1, CMT2, and CMT3 were loaded with 1.0 kg, 1.5 kg, and 2.0 kg of DCM, respectively. The bioreactors were operated for 7 days to complete the bioprocessing process.

Phase 3 of the operation involved the utilization of a nutrient-rich CMT for romaine lettuce production. Initially, each system was supplemented with 10 g per 100 L chelated iron ethylenediamine-N, N’-di[(ortho-hydroxyphenyl) acetic] (EDDHA 6%). The prepared seedlings were transplanted to the NFT grow bed channel. The plant growing period set for this study is 30 days after transplanting. The plants were harvested for plant growth and development measurement at the end of the growing period.

Quality Monitoring and Management of CMT

CMT samples were collected from each system between 8:00 and 10:00 a.m. and were analyzed for various water quality parameters, including dissolved oxygen (DO), pH, total dissolved solids (TDS), and nitrate. These measurements were used to monitor and maintain the CMT quality required in the study. DO was determined using a pen-type auto-calibration DO meter (Lutron, PDO 519). pH level was measured using a pocket-sized pH meter (Hanna, HI98107). TDS was measured using a digital water tester (HM Digital, AP-1). Nitrate levels were estimated using the acid reduction method (API, nitrate test kit). The pH of the water in each system was adjusted using a 33% sulfuric acid solution to maintain an optimal range of 6.5–7.0 (Blanchard et al., 2020). DCM and water were added to the system every five days from the start of the growing period to maintain the required TDS at each treatment. The water level in each treatment was regularly monitored, and any additions were recorded to maintain consistent conditions.

Evaluation of Plant Growth Parameters

Upon harvest, the perforated cup on each plant sample was gently removed, and the roots were lightly rinsed with water to remove the attached cocopeat and then drained. The fresh weight of each plant sample was quantitatively determined by separating the shoot and root portions and measuring their respective weight. Both portions of each plant sample were dried in an oven set at 80^{o}C within a 24-hour time frame, and subsequently, the weight of dried samples for plant biomass determination.

For the measurement of the brix level of leaves from each plant sample, first, the leaves were carefully separated from the stem using a clean knife. The separated leaves were then cut horizontally into small pieces and mixed thoroughly to ensure homogeneity. The cut leaves were grated using a grater with a mesh size of at least 1mm, placed in a fine mesh strainer (200 microns), and manually pressed to collect the plant juice. The collected juice sample was then placed in a handheld brix meter (Soonda, 0–32% model) to measure the Brix value of collected juice from romaine lettuce according to the manufacturer's instructions.

Physiochemical Analysis of Samples

The nutrient content of DCM and the produced liquid fertilizer were analyzed by an accredited laboratory (Regional Soils Laboratory of Department of Agriculture, Regional Field Office III, San Fernando City, Pampanga, Philippines). The moisture content was measured using the gravimetric method. The total nitrogen was determined using the Kjeldahl method. Total phosphorus and total potassium were determined using acid digestion, vanadomolybdate method, and flame atomic emission spectroscopy, respectively. Organic carbon and organic matter were measured using the Walkley-Black method (titrimetric).

Data Analysis

Data from plant growth parameters were displayed as mean ± standard error (SE) of the number of samples gathered at each treatment. Data analyses were performed with the α set at 0.05 (significantly different at a p-value less than 0.05) and were subjected to one-way ANOVA for comparison between treatments. Tukey's post hoc test determined the significant difference among treatments.

RESULTS AND DISCUSSION

Description of the Developed Small-scale Integrated Hydroponics-AWB

The small-scale integrated hydroponics-AWB was designed to combine two different technologies to create a sustainable and efficient method of producing food and managing animal waste (Figure 2). The system's operation involves converting animal waste into a valuable resource that can be used to sustainably produce crops. The system is a closed-loop cycle that benefits both the environment and the farmer by reducing waste and producing high-value crops.

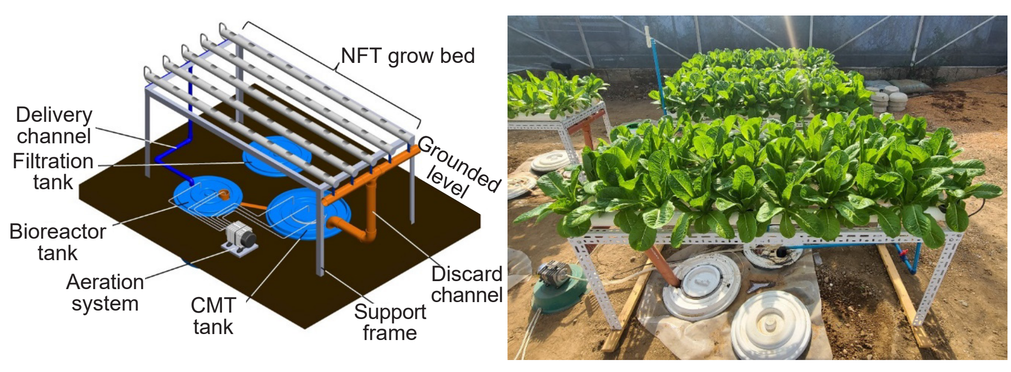

Figure 2. The design (left) and the experimental setup (right) of the small-scale integrated hydroponics-animal waste bioreactor for romaine lettuce production

The system has two main components: a hydroponics system and an AWB. The AWB operates on the principle of aerobic decomposition, where microorganisms use oxygen to break down organic matter in the DCM. The AWB is filled with a medium that provides a surface area for beneficial bacteria to grow. The bacteria feed on the organic matter present in the manure. These bacteria are essential for nitrification, which involves the conversion of ammonia present in DCM to produce a nitrate-rich CMT as a byproduct.

The CMT produced by the AWB is utilized in a hydroponic system, which provides plants with essential nutrients and water without using soil. The CMT is circulated throughout the hydroponic system, facilitating plant growth and development while allowing plants to absorb nutrients from the DCM. Table 1 shows the specification of the developed small-scale integrated hydroponics-AWB.

Environmental Conditions During the Production Period

Lettuce growth could be affected by temperature and relative humidity (RH). The temperature influences various aspects of plant development, such as photosynthesis, respiration, transpiration, and nutrient uptake (Hatfield & Prueger, 2015). The optimum

Table 1
Specification of the developed small-scale integrated hydroponics-AWB for romaine lettuce production

Specifications	Values
CMT capacity	105 Liters
Feedstock capacity	Up to 4 kg
Hydraulic retention period	7 to 14 days
Media-specific surface area (SSA)	500g of K1 media (with SSA of 500m^2/m^3)
Grow bed capacity	50 plants
Plant spacing	15 cm × 15 cm
Aeration System	Airflow rate: 65 Liters per minute
Power rating	65 watts (pump + aerator)

temperature for lettuce production ranges between 15°C to 25°C under greenhouse conditions (Ferrarezi & Testezlaf, 2016). In addition, relative humidity can affect the rate of transpiration, which is crucial for water and nutrient uptake, as well as plant growth and development (Chia & Lim, 2022). A study for lettuce growth and development under controlled humidity levels suggests that plants grown under 85% RH significantly have a faster growth rate compared to plants grown under 50% RH (Tibbitts & Bottenberg, 1976). The temperature and humidity in the greenhouse widely varied between 20.8°C (night) and 42.8°C (day) and 37.5% (day) to 96.2% (night), respectively, during the production period of the study (Figure 3). The values of temperature and RH were favorable during the nighttime compared to the daytime, which exceeded the recommended level.

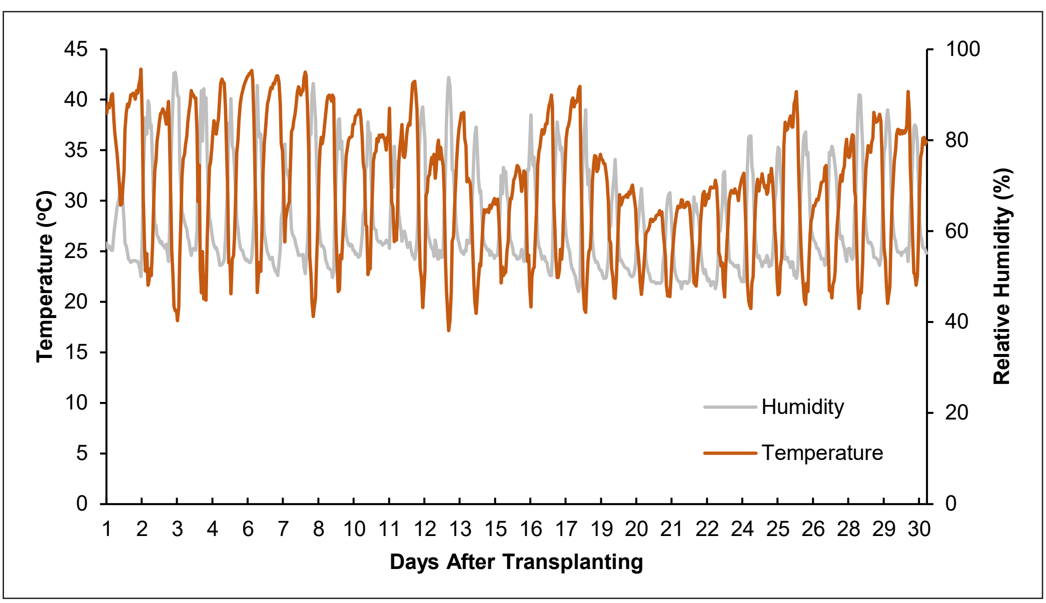

Figure 3. Temperature and relative humidity inside the greenhouse during the production period of romaine lettuce using the developed small-scale integrated hydroponics-AWB for romaine lettuce production

CMT Quality Parameters

Figure 4 shows the trend of dissolved oxygen levels in four different treatments of romaine lettuce over a growing period of 30 days using the developed small-scale integrated hydroponics-AWB. The DO levels observed in this study are critical for the success of the developed system. DO levels can impact the growth and survival of plants and microorganisms in the water (EPA, 2015). Overall, the mean DO level for each treatment remained within a relatively narrow range throughout the study period. Figure 4 shows that CNS had the highest DO levels, with an average of 6.92 mg/L over the 30 days compared to other treatment average DO levels (CMT1 = 6.64 mg/L, CMT2 = 6.19 mg/L and CMT3 = 5.83 mg/L). It suggests that the CNS had a better oxygen supply, possibly due to a lack of animal waste and associated microbial growth that could have consumed the oxygen in the system.

Applying organic fertilizers in hydroponic solutions has been reported to cause the decline of dissolved oxygen in a hydroponic system (Atkin & Nichols, 2004). In comparison with other studies on the utilization of waste as a nutrient source in hydroponic lettuce and cucumber production, the recorded DO level of liquid fertilizer derived from food waste ranges between 8.4 to 9.2 mg/L (Siddiqui et al., 2022). Lower dissolved oxygen levels in treatment with CMT compared to the CNS could be attributed to the higher organic loading rate in this treatment, which may have led to an increased demand for oxygen for the decomposition of organic matter in the DCM.

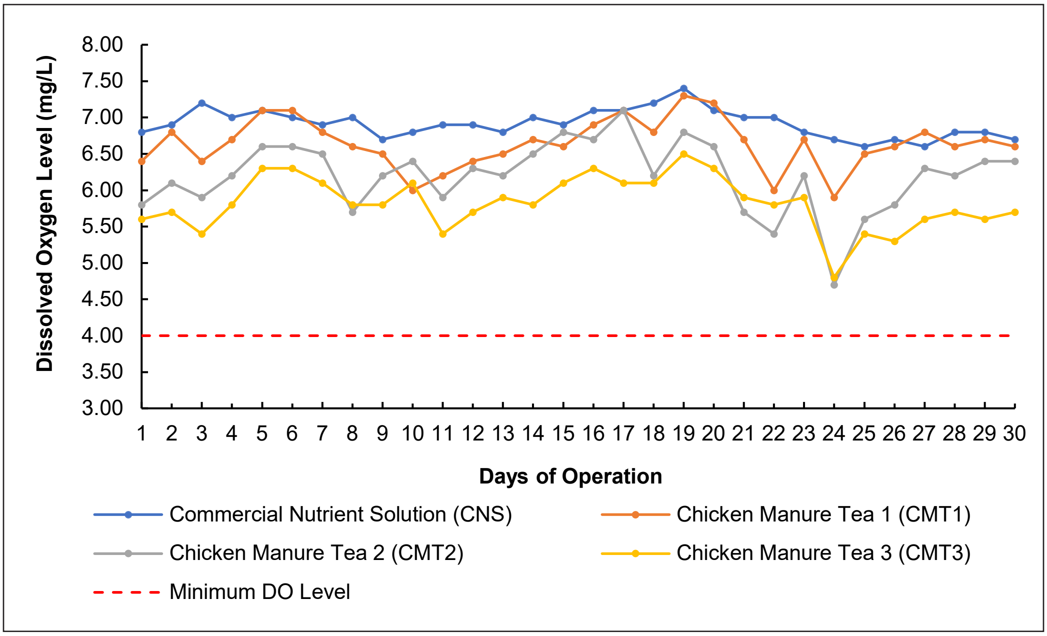

Figure 4. The dissolved oxygen level of chicken manure tea during the production period of romaine lettuce using the developed small-scale integrated hydroponics-AWB for romaine lettuce production

The DO levels in all treatment groups were generally within an optimal level for nitrifying bacteria to oxidize ammonia present in the water at 4 mg/mL and above (Stenstrom & Poduska, 1980). The minimum dissolved oxygen level observed during the study was 4.70 mg/L, considered the minimum acceptable level. Therefore, the dissolved oxygen levels observed in this study suggest that the integrated hydroponics-AWB system could support the growth of both plants and microorganisms without any significant negative impact on DO levels.

However, with the concern of food safety in lettuce production using the developed system, animal waste is regarded as a carrier of pathogenic microorganisms (Zhang et al., 2020). There could be similarities in diseases, such as those found in aquaponics and hydroponics systems; hydrophilic fungi or fungus-like protists are responsible for root or collar diseases (Stouvenakers et al., 2019). Maintaining an optimum level of dissolved oxygen could be a way to reduce the pathogens in the system. The study on biological aerobic treatment of wastewater from piggery suggests that pathogenic bacteria are lower in the products from this kind of treatment than in conventional systems (Béline et al., 2008). Furthermore, the oxidation of biosolids through aerobic digestion at specific durations and temperatures could potentially reduce pathogens (Al-Gheethi et al., 2018). Further studies are needed to ensure that the pathogenic bacteria in this system will be reduced or even eliminated during the bioprocessing period.

Table 2 presents the major nutrient content of CNS, CMT1, CMT2, and CMT3 under varying total dissolved solids using the developed small-scale integrated hydroponics-animal waste bioreactor for romaine lettuce production before and after the production period. There is an observed increase in the total NPK percentage in treatments 1, 2, and 3, while a decrease in NPK percentage is observed in the control. Although the study maintained the specific range of TDS levels on each treatment and control by adding DCM, nutrient solution, and water, there is an observable difference in the total NPK % before and after the production period.

Table 2
Nutrient content of the nutrient solution and chicken manure tea under varying total dissolved solids using a small-scale integrated hydroponics-animal waste bioreactor for romaine lettuce production

Nutrients	**Treatments**							
	CNS		**CMT1**		**CMT2**		**CMT3**	
	Before	After	Before	After	Before	After	Before	After
Total N, %	0.01	0.01	0.02	0.02	0.05	0.02	0.02	0.03
Total P, %	0.05	0.02	0.03	0.03	0.02	0.03	0.04	0.02
Total K, %	0.01	0.01	0.04	0.07	0.04	0.07	0.07	0.06
Total NPK, %	**0.07**	**0.04**	**0.09**	**0.11**	**0.011**	**0.12**	**0.13**	**0.11**

Note. CNS (Commercial Nutrient Solution), CMT 1 (Chicken Manure Tea 1), CMT 2 (Chicken Manure Tea 2), CMT 3 (Chicken Manure Tea 3), Total N (Total Nitrogen), Total P (Total Phosphorus), Total K (Total Potassium) and Total NPK (Total Nitrogen-Phosphorus-Potassium).

Compared with the previously conducted research on the optimum hydraulic retention period of CMT using AWB, there is an observed increasing trend of total NPK content during the production period of CMT (Torres et al., 2023). The slight increase of NPK content after the production period could be attributed to the continuous biodegradation of DCM recently added inside the AWB to maintain a specific range of TDS at each treatment. Based on the most recent research examining different chicken manure loadings on a lettuce-based bioponic system, residual nitrogen and phosphorus present in the system's liquid fertilizer after the production period was attributed to the process's incomplete microbial degradation and could also be utilized by the plant after the extended growing period (Wongkiew et al., 2021).

In the study, the developed integrated hydroponics-AWB could mineralize the organic nitrogen in the DCM into nitrate. The presence of nitrate was determined and monitored using the test kits. Throughout the growth period, the nitrate concentration of CNS was consistently high, ranging between 80 and 160 ppm. During the bioprocessing phase, the nitrate levels in CMT1 and CMT2 increased by up to 80 ppm. However, in CMT3, the nitrate level was below that of the other treatments, which ranged between 5 ppm and 10 ppm. The changes in nitrate levels of different concentrations of CMT could be attributed to the mineralization of organic forms of nitrogen present in DCM using the AWB during the bioprocessing and growth period of the study and absorption of plants with nutrients. Research on using organic fertilizers in hydroponics has found that cultured soil microorganisms could mineralize the organic nitrogen present in organic fertilizer through ammonification and nitrification into nitrates (Shinohara et al., 2011). Furthermore, the study found that excessive amounts of organic fertilizer could inhibit nitrification, which explains the nitrate production level in CMT3 (Shinohara et al., 2011).

During the growing period, both the nitrate levels of CMT1 and CMT2 decline to as low as 40 ppm after 10 days of the growing period. It was observed that after adding DCM to maintain the required TDS level of CMT in the integrated hydroponics-AWB, nitrate levels on both treatments increased by up to 80 ppm. It could be attributed to plant uptake and nitrate utilization for growth and development (Morgan & Connolly, 2013). In addition, microbial denitrification, a process by which bacteria convert nitrate to nitrogen gas (N2) and release it to the atmosphere, could be another reason for the decrease in nitrate level present in CMT (Albina et al., 2019).

Effect of Varying Concentration of CMT on the Growth Parameters of Romaine Lettuce

There is a significant difference in the growth parameters of romaine lettuce produced using conventional hydroponics and integrated hydroponics-AWB with varying CMT concentrations at $p < 0.05$, as shown in Table 3. The highest fresh lettuce weight produced

Table 3
Plant growth parameters of romaine lettuce produced using the nutrient solution and chicken manure tea under varying concentrations using a small-scale integrated hydroponics-animal waste bioreactor for romaine lettuce production

Growth Parameters	Treatment			
	CNS	CMT1	CMT2	CMT3
Plant fresh weight	153.7 ± 9.54a	136.3 ± 13.49ab	100.3 ± 8.98b	43.3 ± 3.45c
Plant dry weight	7.12 ± 0.32a	6.85 ± 0.77ab	4.90 ± 0.63b	2.34 ± 0.18c
Shoot fresh weight	132.3 ± 6.67a	112.7 ± 10.85a	79.8 ± 7.10b	34.7 ± 2.43c
Shoot dry weight	6.00 ± 0.26a	5.78 ± 0.66a	3.98 ± 0.52b	1.84 ± 0.14c
Root fresh weight	21.17 ± 2.65a	23.67 ± 2.33a	20.50 ± 1.33a	8.67 ± 3.20b
Root dry weight	1.115 ± 0.071a	1.042 ± 0.097a	0.930 ± 0.118a	0.502 ± 0.049b

Note. Values within the same row followed by different letters are significantly different at $p < 0.05$. Note: CNS (Commercial Nutrient Solution), CMT 1 (Chicken Manure Tea 1), CMT 2 (Chicken Manure Tea 2), CMT 3 (Chicken Manure Tea 3)

was observed in the CNS (153.7 ± 9.54 g), which is highly significant compared to CMT2 and CMT3. No significant difference was observed between CNS and CMT1 in terms of the fresh weight of romaine lettuce (Figure 5). CMT3 produced the lowest fresh lettuce weight (43.3 ± 3.45 g), significantly different among the treatments. The plant dry weight showed a similar tendency as the plant fresh weight, with the CNS having the highest plant dry weight (7.12 ± 0.32 g) and CMT3 having the lowest dry weight (2.34 ± 0.18 g). No significant difference was observed between the CNS and CMT1 in terms of plant dry weight.

The same result could be observed in both root and shoot growth. The shoot fresh weight and dry weight of romaine lettuce produced in CNS were highly significant in comparison to the shoot fresh weight and dry weight of romaine lettuce produced in CMT2 and CMT3 ($P < 0.01$). On the other hand, there is no significant difference between the CNS and CMT1 ($P > 0.05$). The

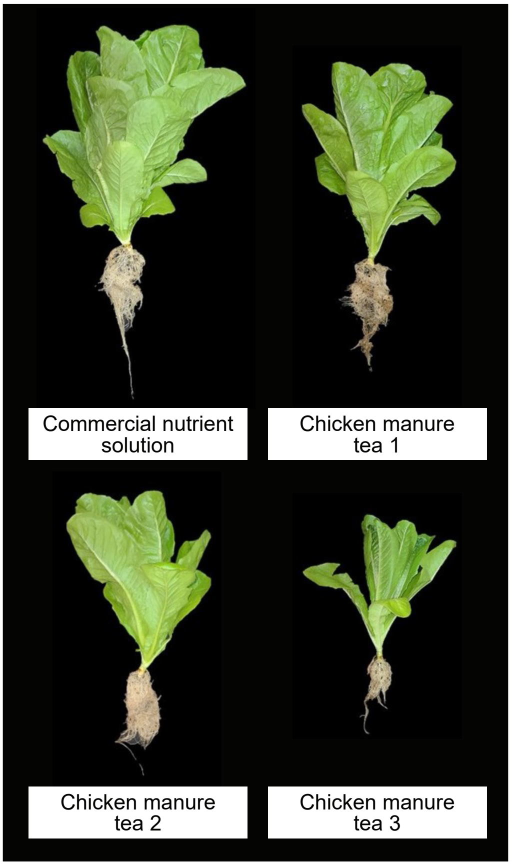

Figure 5. Romaine lettuce is produced using conventional hydroponics and small-scale integrated hydroponics-AWB

root fresh and dry weight of romaine lettuce produced in CNS were highly significant in comparison with CMT3 ($P < 0.01$). No significant difference was observed among CNS, CMT1, and CMT2 ($P > 0.05$).

There is a significant difference in the weight and mean of the number of romaine lettuce leaves produced using conventional hydroponics and integrated hydroponics-AWB with varying CMT concentrations at $p > 0.05$ (Figure 6). The mean number of leaves of romaine lettuce produced using the control, Treatments 1 and 2, was significantly higher than the mean number of leaves of romaine lettuce produced using Treatment 3. The leaf fresh weight was observed in the control, and this value decreased significantly with increasing concentrations of CMT. Treatment 3 showed the lowest fresh leaf weight, significantly different from the control.

In comparison between the CNS and CMT1, the result suggests that CMT1 could be a potential replacement for the CNS for romaine lettuce production using the developed integrated hydroponics-AWB. On the other hand, the result of the experiment also implies that using CMT at a high ppm level had a negative impact on the growth parameters of lettuce. The experiment's findings were in line with other research that showed a reduction in plant weight above ground when aerated chicken manure extract concentration was raised from 10 to 25 g/L and a permanent wilting of the plant at 50 g/L (Tikasz et al., 2019). In contrast, in a study of different chicken manure loadings (200 g, 300 g, and 400 g) on bioponic lettuce production, raising the manure loading rate increased plant yield (Wongkiew et al., 2021).

The observed decrease in values of growth parameters of romaine lettuce with higher ppm levels of CMT could be attributed to the toxic effects of organic nutrients in CMT

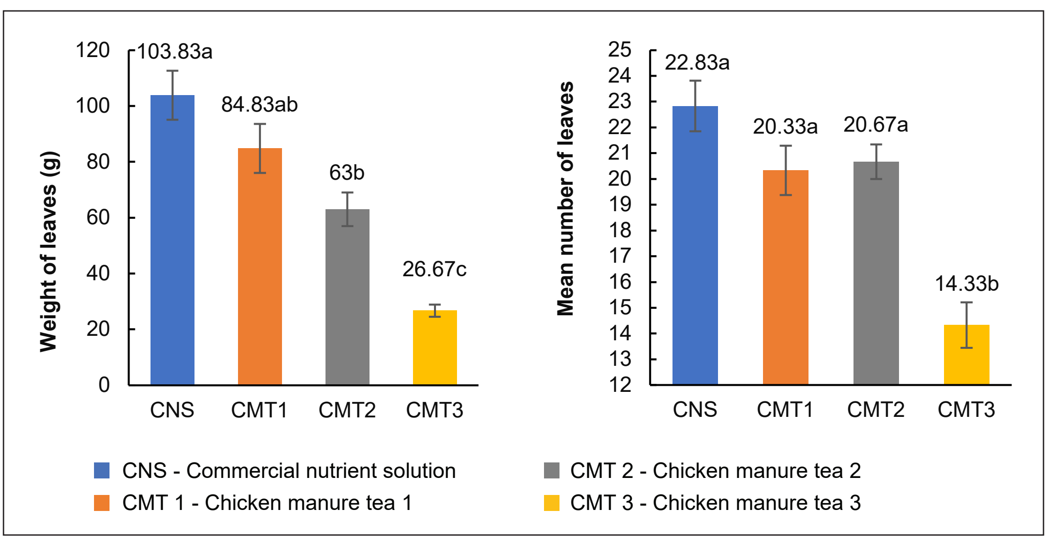

Figure 6. Influence of varying concentrations of chicken manure tea on the leaf weight (left) and number of leaves (right) of romaine lettuce produced using small-scale integrated hydroponics-AWB

on the plant. The quality of organic-based inputs for the growth of plants depends on mineralization rates that can be highly variable due to the quality of organic inputs and environmental conditions (Zandvakili et al., 2019). Unlike inorganic fertilizers, organic nutrients from animal waste are not readily available to the plant and must be converted to plant-available forms by microbes present in the system (Bi et al., 2010). Likely, higher ammonia levels in CMT3 and other organic nutrients could have resulted in stunted lettuce growth in weight and number of leaves. On the other hand, the values of growth parameters of romaine lettuce produced at lower concentration (CMT1) using the developed was comparable to the romaine lettuce produced in conventional hydroponics (CNS). It implies that the use of CMT at lower concentrations could be used for lettuce production using the developed system.

There was no significant difference in the moisture content and brix value of lettuce produced among each treatment using the integrated hydroponics-AWB at $p > 0.05$ (Figure 7). The moisture content of romaine lettuce plants is an important quality parameter as it affects the nutrient concentration present in the plant (Mou, 2012). The findings of the research on the comparison of the growth characteristics between hydroponically grown and soil-grown lettuce resulted in a lettuce moisture content of 94.23% and 92.94%, respectively (Lei & Engeseth, 2021). The dry matter of lettuce cultivated using typical hydroponics and decoupled aquaponics was 4.9% (95.1% MC) and 4.7% (95.3%), respectively (Monsees et al., 2019). The moisture content of the romaine lettuce produced using integrated hydroponics-AWB is comparable to that of the other studies conducted. The findings may indicate that the developed small-scale integrated hydroponics-animal waste bioreactor did not significantly impact the moisture content of the romaine lettuce plants.

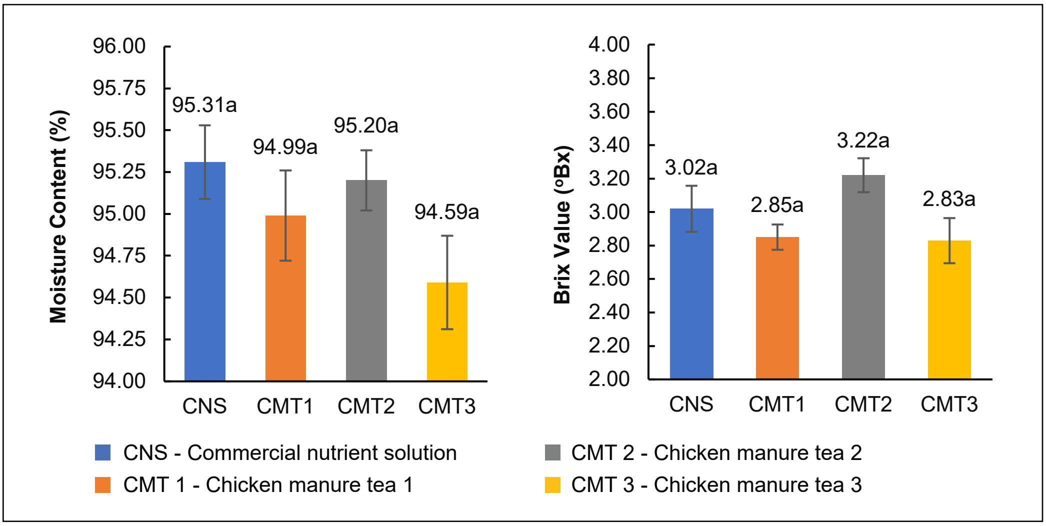

Figure 7. Influence of varying concentrations of chicken manure tea on the plant moisture content (left) and brix value (right) of romaine lettuce produced using small-scale integrated hydroponics-AWB

In terms of the brix value of the romaine lettuce produced using conventional hydroponics and integrated hydroponics-AWB with varying CMT concentrations, data suggests no significant differences in the Brix levels among the treatments. Brix value is used to express the level of soluble solids (sugars, pectin, organic acids, and amino acids) in a liquid, representing estimates of sugar content in fruits and vegetables (Kleinhenz & Bumgarner, 2012). In studying the effect of varying nutrient solutions on the brix value of lettuce produced in an NFT system, the results on romaine lettuce vary between 4.5 and 5.4 (Thakulla et al., 2021). Research on the use of biofertilizer from microalgae (chlorella vulgaris) in hydroponic lettuce production resulted in a brix value ranging between 3.7 and 4.5.

The brix value of romaine lettuce produced using integrated hydroponics-AWB was slightly lower than in the other literature. However, it is important to take into consideration that it could have been influenced by other factors, such as the specific cultivar of lettuce used in the experiment and the type of nutritional support for plants. The lack of significant differences in the brix value of romaine lettuce produced at conventional hydroponics and integrated hydroponics-AWB with varying CMT concentrations implies that the use of DCM as a nutrient source for hydroponically grown lettuce could not affect the quality of the produced lettuce.

CONCLUSION

The small-scale integrated hydroponics animal waste bioreactor was designed, fabricated, and tested to determine the ideal concentration of chicken manure tea as a source of nutrients for romaine lettuce in comparison with conventional hydroponics. The result of the study implies that the optimum concentration of chicken manure tea in the system should be between 1,000 ± 50 ppm. Within the optimum concentration of chicken manure tea in the system, the small-scale integrated hydroponics animal waste bioreactor produced romaine lettuce with growth parameters comparable to those produced on conventional hydroponics. Increasing the chicken manure tea concentration to 1,400 ± 50 ppm using the developed system negatively impacted the plant growth parameters of romaine lettuce in comparison to conventional hydroponics. The result of the study provides an alternative and sustainable method for hydroponic romaine lettuce production using the developed small-scale integrated hydroponics-animal waste bioreactor to bioprocess the chicken manure into nutrient-rich chicken manure tea as a plant nutrient source. Further studies are recommended to determine the scalability and techno-economic feasibility of the developed system for larger-scale hydroponic farming operations. In addition, it is also recommended to observe the secondary nutrient and micronutrient level of the chicken manure tea further to assess its impact on plant growth and development.

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