Jurnal Teknologi

Full Paper

PERFORMANCE OF BUBBLE COLUMN BIOREACTOR TO PRODUCE LIQUID ORGANIC FERTILIZER (LOF) FROM COW URINE AND SLURRY ORGANIC WASTE

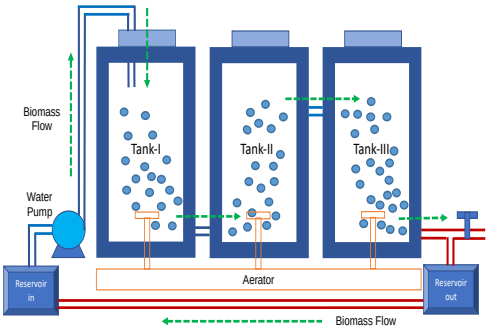
Yohanes Setiyo[a]*, Bambang Admadi Harsojuwono[b], Ida Bagus Wayan Gunam[b], Ida Bagus Putu Gunadnya[a], I Gusti Ngurah Apriadi Aviantara[a], I Gusti Ayu Lani Triani[b]

[a]Department of Agricultural and Biosystem Engineering, Faculty of Agriculture Technology, Udayana University, Badung 80361, Indonesia
[b]Department of Agroindustrial Technology, Faculty of Agricultural Technology, Udayana University, Badung 80361, Indonesia

Article history
Received
14 April 2022
Received in revised form
30 April 2023
Accepted
4 May 2023
Published Online
25 June 2023

*Corresponding author
yohanes@unud.ac.id

Graphical abstract



Abstract

This research aimed to evaluate the effectiveness of various diffusers in a bioreactor for producing liquid organic fertilizer (LOF) using cow urine and slurry organic waste as materials. LOF was produced by treating the organic waste with Urine-Single Orifice Diffuser (USOD), Urine-Coarse Bubble Diffuser (UCBD), Urine-Fine Bubble Diffuser (UFBD), Slurry-Single Orifice Diffuser (SSOD), Slurry-Coarse Bubble Diffuser (SCBD), and Slurry-Fine Bubble Diffuser (SFBD) in a bioreactor equipped with a water pump and an external aerator unit. The pump circulated and agitated the biomass at a discharge of 10 L/min. Meanwhile, the external aerator unit pumped air into the bioreactor, which the biomass absorbed at a discharge of 45 L/min. The discharge of biomass and water also caused agitation of the fermented biomass. Several variables were observed during the fermentation process, including temperature, acidity (pH), total dissolved solids (TDS), C/N ratio, electrical conductivity (EC), biological oxygen demand (BOD), microbial population, cation exchange capacity (CEC), and other chemical properties. LOF produced had a pH, temperature, total organic matter content, EC, TDS, BOD, microbial population, and CEC values of 6.3–6.8, 28.0–37.0°C, 20.77–22.46%, 54630–62369.24 µmS, 7543–11700 ppm, 1.2–2.6 mg/L, 3.2–7.5 log CFU, 12.3–14.1, and 29.98 me/100 g, respectively. The macro and micro nutrient contents of bio-urine were 12.72–12.96%, and 3.09–3.25%. Meanwhile, those of the bio-slurry were 12.35–12.99% and 2.39–2.72%. These characteristics of LOF were in accordance with the Indonesian National Standard (SNI).

Keywords: Organic waste slurry liquid organic fertilizer, bioreactor fermentation, and cow urine

1.0 INTRODUCTION

Using synthetic fertilizers continuously by farmers has detrimental effects on the soil, including a decrease in the number of micronutrients [1]. This leads to (1) a decrease in the population of soil biota that decomposes soil organic matter as well as insecticide and fungicide precipitates, (2) a decline

in soil material characteristics, such as porosity, water-holding capacity, water infiltration, and oxygen content, and (3) decrease in the micro-nutrients and organic matter [2]. Subsequently, it is advised for farmers to utilize solid organic fertilizer (SOF), namely compost. Farmers can also use liquid organic fertilizer (LOF) containing macro and micronutrients and non-pathogenic microbes. In horticultural cultivation in highland areas, LOF and SOF have the potential to reduce fertilizer usage by 40–60% [3].

LOF is a nutrient-rich solvent made from fermented organic material, made from various sources, such as plant residues, animal, and human wastes [4]. One of the methods used to produce LOF is using a mixture of cow urine with organic waste from traditional markets, which is then transformed into a slurry.

A mature cow is capable of producing 6 to 10 L/day. Despite its high nutrient content, cow urine is not commonly used as a fertilizer compared to its feces. Cow urine contains macronutrients such as phosphorus (P), nitrogen (N), and potassium (K) at concentration rates of 1.00%, 0.50%, and 1.50%, respectively. Furthermore, it contains several micro nutrients such as sodium (Na), calcium (Ca), magnesium (Mg), aluminium (Al), iron (Fe), sulfur (S), copper (Cu), and molybdenum (Mo) with water content of 95% [5]. Balinese farmers typically own 2-4 cows on average and are organized into Integrated Agricultural System (SIMANTRI) groups, each comprising 15–20 cows. This demonstrates the significant potential of farmers producing bio-urine from cattle for sustainable agriculture.

In Bali, traditional markets generate approximately 32 m^3 of waste every day, consisting of organic waste at 65.6% and non-organic waste at 34.4%. The organic waste mainly comprises vegetables, fruits, and food scraps, which account for ±1.6 tons per day. Solid organic waste from traditional markets has the primary components of fruits and vegetables, including fats, proteins, and carbohydrates. As stated by Paramita *et al.* [6], this waste typically has crude fiber (cellulose) at approximately 41–60%, fat (amino acids and protein) at 3-9%, ash at 4–20%, and water at 30–60%.

Cow urine and organic litter from traditional markets have the potential to undergo aerobic and anaerobic fermentation processes using microorganisms, resulting in simple minerals, energy, and water vapor [7]. Scientists in Bali, including Aritonang *et al.* [8], Suhartana *et al*. [9], Mudiarta *et al*. [10], Putra *et al*. [11], and Wirawan *et al.* [12], developed the fermentation process for organic matter on a laboratory scale (10–50 L capacity). They achieved this by adding molasses, urea, microbes, and oxygen to the fermentation process, which typically lasted 15–21 days [13]. The addition of these ingredients could increase the nutrient enrichment of the resulting liquid fertilizer by 3–8 times [14].

Laboratory-scale research has been conducted on producing LOF using biomass from livestock waste, including urine and other organic materials, in simple bioreactors. Nevertheless, to enable the implementation of this process on a more extensive range, particularly at the group of small-scale farmers with 2–4 cows or in the marketplace, additional studies are needed, particularly regarding the bioreactor performance during fermentation.

To facilitate aerobic fermentation of biomass, the bioreactor should possess certain capabilities, including the ability to (1) regulate the temperature within the range of 25 to 40°C, to enable mesophilic microbes to proliferate during the decomposition of the biomass organic material [15], (2) dissipate outsized heat to the circumstances [16], (3) ensure a consistent oxygen supply in the biomass, and (4) maintain the pH dynamics of the biomass within a range of 6 to 8, to create neutral pH conditions that promote microbial growth [17]. Accordingly, it is essential to equip the bioreactor with a continuous biomass flow pump, along with a 25-watt aeration pump that includes a bubble column for oxygen addition [18].

Additional components such as urea, molasses, or microbes are incorporated into the process. A continuous stream of biomass helps homogenize the fermented substrate. Even though farmers scaled up the laboratory-scale bioreactors to a capacity of 60–500 liters, they still experienced several weaknesses. The bioreactor performance is not optimal because (1) the fermentation time is more than 15 days, and (2) the C/N LOF value does not meet SNI standards.

The bioreactor lacks the necessary equipment for mixing the biomass and supplying oxygen to support aerobic fermentation, despite the addition of urea, sugar, and microbes at the start of the process. The study evaluated the result of implementing a bioreactor model that utilizes semi-continuous fermentation for fermenting biomass consisting of cow urine and slurry obtained from organic waste obtained from traditional markets. The bioreactor had a total capacity of 3300 L, consisting of three tanks, each weighing 1100 L. The primary objective was to investigate the ability of the bioreactor to facilitate aerobic fermentation of the aforementioned biomass and generate LOF that conforms to Indonesian National Standard (SNI) requirements.

2.0 METHODOLOGY

2.1 Material

The first primary component used was cow urine, which had electrical conductivity (EC), total dissolved solids (TDS), pH, organic matter content, and C/N values of 6.47, 9410±12 ppm, 54210±31 µmS, 12±0.7%, and 42±4, respectively. The second was slurry obtained from the decomposition of organic waste in a traditional market, which had pH, TDS, EC, and organic matter content values of 6.47±0.2,

9410±15 ppm, 54210±23 µmS, and 12±0.7%, respectively. The supporting materials used in the experiment included molasses, Nitrosomonas bacteria, potato dextrose agar, and distilled water for bacterial population analysis. In analyzing cation exchange capacity (CEC), K_2O, C-organic, N-organic, and P_2O_5, it was used several chemical ingredients, including distillate water, Fe_2SO_4, H_2SO_4, $K_2Cr_2O_7$, Na_2SO_4, $CuSO_4$, NaOH, NH_4OH, HCl, Na_2SO_5, $BaCl_2$, NH_4-acetate, and 80% alcohol.

The equipment utilized consisted of three fermentation tanks, each with a capacity of 1100 L, which were combined to form a semi-continuous bioreactor [19]. The bioreactor was prepared with a pump with a power of 186.5 watts, which was responsible for transporting biomass from tank 3 to 1. Biomass flowed from tank 1 to 2 and from 2 to 3 through a connecting pipe driven by gravity. This biomass flow played a role in the agitation of the biomass within the bioreactor [5].

Each of the three tanks (1, 2, and 3) was also equipped with an external pump unit and linked to a PVC diffuser pipe with a diameter of 25.4 mm. The function of this unit was to circulate air in the fermented biomass, thereby allowing the biomass to absorb oxygen.

Furthermore, the flow of air through the bioreactor was responsible for stirring the biomass [10, 12]. The external pump unit used had a power rating of 50 watts, an airflow of 45 L/min, and a pressure of 0.038 Mpa. This unit was capable of providing oxygen to the fermented biomass at a rate of 9.42 L/min.

A single orifice diffuser was used to produce a column of individual air bubbles. The diffuser was connected to the pump via a PVC pipe with a diameter of 25.4 mm. At the end of the PVC pipe, a plastic hose with an 8 mm diameter was attached [10].

The air bubble column generated by the diffuser moves rapidly from the diffuser surface to the biomass surface. Due to this movement, very coarse bubbles are unable to encounter the biomass for sufficient time to achieve a significant level of SOTE during aeration [20]. As a result, the amount of oxygen absorbed by the biomass was found to be 0.03% [10].

The coarse air bubble diffuser was made of PVC pipe with a diameter and length of 25.4 mm and 30 cm. The pipe was given 64 holes with a diameter 3 mm. When the air bubbles were released from the diffuser, they moved quickly from the surface of the diffuser to the biomass surface. However, because only 0.9% of the oxygen was absorbed by the biomass [10], the diffuser did not meet the SOTE criteria for aeration [20].

The fine bubble diffuser was constructed from ceramic stone with a diameter and thickness of 20 cm and 2 cm. It released bubbles with a diameter of 1 mm. The fine bubble diffuser caused the slurry to absorb up to 3.5% of the oxygen [10]. As a result, it met the Standard Oxygen Transfer Efficiency (SOTE) required during aeration [20], with an SOTE value of 2%.

The measuring instruments used included a TDS and EC meter that could measure a range of 0–5000 ppm and 0–9990 µmS. This was in addition to laboratory analysis equipment BOD meter, and pH meter, comprising of the volumetric flask, measuring glass, Erlenmeyer, dropper pipette, burette, and Kjeldahl flask.

2.2 Research Implementation

This study was performed with several treatments on the organic waste with Urine-Single Orifice Diffuser (USOD), Urine-Coarse Bubble Diffuser (UCBD), Urine-Fine Bubble Diffuser (UFBD), Slurry-Single Orifice Diffuser (SSOD), Slurry-Coarse Bubble Diffuser (SCBD), and Slurry-Fine Bubble Diffuser (SFBD) during the fermentation in a bioreactor. There are three replications in each treatment, subsequently, 18 experimental units were obtained.

2.3 Research Procedure

a) Biomass in the form of slurry was the outcome of organic waste from vegetables and fruits reduced and blended into a smooth and homogeneous product and mixed with water in a ratio of 1:1 by weight.
b) The cow urine biomass was obtained in the form of filtered cow urine.
c) During the preparation, nitrifying bacteria at 5%, urea at 2%, and molasses at 2% were supplemented to the biomass. The resulting mixture was then allocated to three separate bioreactor tanks based on the intended treatment.
d) The biomass was consistently circulated from tank 1 to 2, and from tank 2 to 3, through pipes by means of gravity in a continuous flow. However, a pipe was used to transport it from tank 3 to 1. The biomass underwent fermentation in a bioreactor for a period of 15 days.
e) Daily monitoring was carried out to observe and record various variables throughout the fermentation process. These included temperature, pH, BOD, and TDS by assuming flowing 1000 mL biomass samples.
f) Observation of carbon (C) and N content microbial population was carried out every 3 days by collecting 1000 mL samples from flowing biomass for analysis.
g) The content parameters of P_2O_5, K_2O, and other relevant substances were observed upon the completion of the fermentation process.

2.4 Observed Variables

The pH, temperature, EC, and TDS were measured by (1) placing the 20 mL sample taken from the fermenter into a 100 mL beaker. Furthermore, (2) the measuring tools, including a TDS meter, thermometer,

and pH meter, were installed into the sample in a beaker and allowed to stabilize. (3) Subsequently, the sample was diluted prior to conducting the analysis on the EC value.

The first stage for observing CEC variables, CEC, macronutrient content (C-organic, N, P_2O_5 dan K_2O), micronutrient content (Mn, Cu, Zn, Fe, B, and Co) and microbial population was collecting 1000 mL of biomass samples undergoing fermentation. These samples were randomly collected from five points in tanks 1, 2, and 3, where the biomass was being fermented in a bioreactor.

The CEC was observed using the washing method [21]. To perform this procedure, 10 mL of biomass was placed in a tube and mixed with 10 mL of NH_4OAc pH 7. The resulting mixture was vigorously shaken for 60 minutes and then subjected to centrifugation for 10 minutes. The solution filtration was performed through filter paper, and the filtrate collected in a container. The biomass was then washed by adding 10 mL of NH_4OAc pH 7 to the tube, centrifuged for 10 minutes, filtered, and the filtrate was accommodated back into the container. Furthermore, the biomass was washed again by adding 10 mL of NH_4OAc 1N pH 7 containing 1% NH_4Cl 1N to the tube and centrifuged for 10 min. After filtrating the resulting combination, the yield was collected and returned to the tube container.

The filtered sample in the tube was mixed with 10 mL of ethanol and centrifuged for 10 min. The resulting separated liquids were discarded, and this process was repeated up to three times. After the ethanol was washed out four times, it was also discarded. The remaining biomass sediment particles in the tube were then mixed with 10 mL of distilled water, transferred to a Kjeldahl tube, and added to 20 mL of 40% NaOH, ±50 mL of distilled water, and distilled immediately. The results were collected with 15 mL of 0.1N H_2SO_4, added with 3 drops of Conway indicator. The distillation process was halted once the reservoir reached a volume of approximately 50 mL. The collected distillate was titrated with NaOH of known normality. The NaOH volume required to change colour from red to green was recorded.

$$\text{CEC(me/100g)} = \text{mL Blank} - \text{mL Sample} \times \text{N NaOH} \times 100 \times \text{cf} \quad (1)$$

CEC = cation exchanged capacity, me/100 g
N = normality of the solution
cf = moisture content correction factor = 100/(100 - % moisture content)

The measurement on the c-organic parameter was carried out using the Walkley and Black method [22]. One gram of biomass sample was mixed with 10 mL of 1N $K_2Cr_2O_7$ in an Erlenmeyer flask., 20 mL of H_2SO_4 was added to the solution, which was then shaken to ensure homogeneity. The resulting mixture was left for 30 minutes. A blank sample without biomass was prepared in the same way. After 30 minutes, the mixture was diluted with 200 mL of H_2O, and 10 mL of 85% H_3PO_4 was added. Additionally, 30 drops of Diphenylamine indicator were supplemented to the solution, which was then titrated with $FeSO_4.7H_2O$ 1N using a burette. The titration was stopped when the colour changed from dark to light green.

$$\%\text{C - organic} = \frac{(\text{mL Blank} - \text{mL Sample}) \times 3 \times \text{cf}}{\text{mL Blank} \times \text{Weight Sample}} \quad (2)$$

% C-organic = content carbon-organic of biomass
cf = moisture content correction factor = 100/(100 - % moisture content)

The N-total was determined using the Kjeldahl method [23]. First, 25 mL of the biomass sample was placed into an Erlenmeyer flask, along with 1.9 g of a mixture of Se, $CuSO_4$, and Na_2SO_4. In addition, 5 mL of concentrated H_2SO_4 was added slowly to the flask and shaken. The sample was also added with 5 drops of liquid paraffin. The mixture was heated over low heat in an acid room, with the fire gradually increased until a green liquid was obtained. It was further heated for 15 minutes and allowed to cool. The sample was added with 50 mL of water, shaken briefly, and transferred quantitatively into the distillation flask. The liquid material in the distillation flask should not exceed ½ of the contents of the flask. Furthermore, 5 mL of 50% NaOH was put into the distillation flask, and the distillation stage was conducted. The resulting distillate was collected in a 125 mL Erlenmeyer flask that had been filled with a mixture of 100 mL of 4% H_3BO_4 and 5 drops of Conway indicator. The distillate titration was accomplished using standardized HCl until the colour changed from green to red. In addition, blanks were determined according to the procedure above.

$$\%\text{N} = \frac{\text{Vol HCl (Sample - Blank)} \times \text{N HCl} \times 1400}{\text{Weight Sample (mg)}} \quad (3)$$

% N = N content of biomass
N = normality of the solution

P-available and K-available were observed using the II method [24]. Firstly, a standard P solution with a concentration of 50 ppm was prepared and then diluted to 2, 4, 6, 8, and 10 ppm in 50 mL volumetric flasks. Subsequently, 1 mL of an 8-ppm P standard solution was introduced into a 10 mL volumetric flask, and then 9 mL of reagent solution was added subsequently. After allowing the solution to stand for 15 minutes, it was transferred to a UV-Vis cuvette, and its absorbance was calculated at a wavelength between 650–750 nm.

Seven 25 mL volumetric flasks were prepared, with flask 1 being filled with blanks, while flasks 2 to 7 comprised 1 mL of standard P solutions of 2, 4, 6, 8, and 10 ppm, respectively. 9 mL of reagent was added to each flask, and the solution was allowed to

stand for 15 minutes. The resulting solutions were then put into a cuvette, and the absorbance was estimated at the highest wavelength.

$$\%P = \text{ppm curve} \times \text{mL extract}/1000\ \text{mL} \times 100/\text{mg sample} \times \text{df} \times 0.32 \times \text{cf} \quad (4)$$

% P = content phosphate of biomass
ppm curve = sample rate acquired from the correlation regression curve between the levels of the standard series and their readings after deducting blanks
df = dilution factor
cf = moisture content correction factor = 100/(100 - % moisture content)

K levels in the sample were determined by preparing a standard K solution from 20 ppm K standard solution with variations of 2, 4, 6, 8, and 10 ppm. This was achieved by taking 1, 2, 3, 4, and 5 mL of the standard solution and adding distilled water to a 10 mL volumetric flask. The resulting solutions were then measured for their absorbance using AAS and plotted on a graph to obtain K calibration curve. Subsequently, 25 mL of biomass was added to a Kjeldahl flask, followed by the addition of 5 mL of HNO_3 and 0.5 mL of $HClO_4$. The mixture was shaken and left overnight before being heated gradually from 100°C to 200°C until a white vapor appeared. The remaining 0.5 mL of liquid in the flask was cooled, diluted with H_2O, and adjusted to 50 mL. The resulting mixture was thoroughly shaken until it was uniform and left to stand overnight, or filtered using W-41 filter paper to get a clear extract, referred to as "extract A". One milliliter of extract A was then transferred into a 25 mL volumetric flask, and distilled water was added to fill the flask up to the mark. The resulting mixture (extract B) was shaken until homogeneous, and the K levels were compared using the standard series.

$$\%K = \text{ppm curve} \times \text{mL extract}/1000\ \text{mL} \times 100/\text{mg sample} \times \text{cf} \quad (5)$$

% K = content Kalium of biomass
ppm curve = sample rate obtained from the correlation regression curve between the levels of the standard series and their readings after deducting blanks
cf = moisture content correction factor = 100/(100 - % moisture content)

The total plate count (TPC) method [25] was used to calculate the bacterial population. A dilution of the biomass sample ranging from 10^{-4}–10^{-9} was prepared, and 1 mL of this dilution was plated onto sterile Plate Count Agar (PCA) media. The diluted solution was evenly distributed across the agar surface using a sterile glass rod, and the agar plates were then incubated at room temperature for 48 hours. Bacterial colonies were counted for those numbering between 30 and 300.

2.5 Data Analysis

Fermentation process variable data such as temperature, pH, BOD, TDS, EC, and C/N were tested with ANOVA at a 5% or 1% confidence interval. When there was a substantial change between the treatments, then the smallest significant difference was continued with the Duncan test. In addition, the data were recorded in tables and graphs using the Excel program, and both were subjected to descriptive analysis to evaluate the fermentation process quality for each treatment.

The data collected on the quality of bio-slurry and bio-urine, including macro-nutrient, micro-nutrient, CEC, EC, BOD, pH, microbial population, C/N, and TDS contents at the end of a 15-day fermentation process were analyzed using ANOVA at either 5% or 1% confidence intervals. The smallest significant difference was determined using the Duncan test, assuming significant differences were found between the treatments. This statistical analysis aimed to evaluate and determine the best fermentation treatment.

2.6 Calculation of the Amount of Energy in the Fermentation Process

a) Heat to increase the temperature of biomass [16]

The equation used to calculate the heat required to increase the temperature in the bioreactor was dependent on several variables. These included the volume of biomass present, the mass density, the specific heat, and the change in temperature between the biomass on day h and the prior day. The heat to increase temperature was formulated using Equation (6).

$$Q_\theta = \rho_P \times V_P \times C_P \times \Delta\theta \quad (6)$$

Q_θ = the heat to increase temperature, J/kg
ρ_P = biomass mass density, g/mL
V_P = biomass volume, m^3
C_p = specific heat of biomass J/kg-°C
$\Delta\theta$ = difference temperature of biomass on day h and h-1, J/kg

$$\Delta\theta = \theta_h - \theta_{h-1} \quad (7)$$

$\Delta\theta$ = difference temperature of biomass on day h and h-1, °C
θ_h = temperature of biomass on day h, °C
θ_{h-1} = temperature of biomass on day h-1, °C
C_p = 4180 J/kg-°C
ρ = 1.01 g/mL

b) Heat lost to the environment [16]

The heat lost to the environment referred to the amount of heat lost from the bioreactor to the surroundings due to the change in temperature between the biomass in the bioreactor (T_b) and the average ambient temperature. This heat transfer was considered unsteady because the biomass's temperature in the bioreactor was constantly changing. The heat loss to the environment was determined based on the convection heat transfer coefficient on the inner wall of the bioreactor (h_d), the conduction heat transfer coefficient of the bioreactor wall (k), the thickness and the surface area of the bioreactor wall, and the temperature dissimilarity between the biomass inside the bioreactor and the ambient air temperature outside:

$$\Delta T = T_b - T_l \quad (8)$$

ΔT = the difference in air temperature between inside the bioreactor and outside the bioreactor, °C

T_b = air temperature outside the bioreactor, °C

T_l = air temperature outside the bioreactor, °C

$$Q_L = U \times A \times \Delta T \quad (9)$$

Q_L = heat lost to the environment, J/kg
U = combined heat transfer coefficient, J/(m² - °C)
A = surface area of the bioreactor wall, m²
ΔT = the difference in air temperature between inside the bioreactor and outside the bioreactor, °C

The heat resistance value was calculated by combining the conduction coefficient and convection heat transfer [26]. The score of h_d and h_l represent the convection heat transfer coefficient between the inner wall and the outer wall of the bioreactor, consecutively. The formula used to calculate the convection heat transfer coefficient on the bioreactor's inner wall was shown in Equation (10).

$$U = \frac{1}{h_d} + \frac{k}{\Delta x} + \frac{1}{h_L} \quad (10)$$

U = combined heat transfer coefficient, J/(m² - °C)
h_d = convection heat transfer coefficient on the inner wall of the bioreactor, J/(m² - °C)
Δx = bioreactor wall thickness, m
K = conduction heat transfer coefficient, J/(m - °C
h_l = convection heat transfer coefficient on the out wall of the bioreactor, J/(m² - °C)

The bioreactor tank was constructed using 3 mm thick plastic fiber material with thermal properties of k = 0.25 J/(m - °C), h_d =10 J/(m² - °C) [26], and h_l = 17 J/(m² - °C) [25]. The values of k, h_d, h_l and Δx were inputted in equation (5) to determine U, which had a value of 250 (m² - °C)/J.

c) Heat of fermentation [16]

The heat of fermentation was determined by combining the heat required to increase temperature and the heat lost to the environment. Since the bioreactor was closed, the heat utilized to vaporize biomass water could be neglected. The heat of the fermentation reaction was shown in Equation (9).

$$Q_R = Q_\theta + Q_L \quad (9)$$

Q_R = the heat of fermentation, J/kg
Q_θ = the heat to increase temperature, J/kg
Q_L = heat lost to the environment, J/kg

3.0 RESULTS AND DISCUSSION

3.1 The Aerobic Fermentation Reaction Temperature in a Bioreactor

Aerobic fermentation is the process of utilizing oxygen and microorganisms to convert substrates into a desired product while generating energy. The energy produced during fermentation was harnessed to raise the temperature of the fermented biomass, drive water evaporation from the biomass, and dissipate it into the surroundings. The outcomes were the heat of fermentation from USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments, which were 600–4800, 600–5400, 600–6500, 830–5700, 830–7600, and 830–9200 J, respectively. The heat of fermentation in UFBD and SFBD treatment was close to the findings of Setiyo *et al.* [16], where a heat insulator was used in the bioreactor. This indicates the aeration effectiveness that can be achieved by installing a fine bubble system that can substitute the insulating function in the bioreactor.

Fruit and vegetable waste slurries contain a more complex chemical content and structure than cow urine, resulting in a higher heat of fermentation. The chemical elements of fruit and vegetable slurry are more complete than those of cow urine. Therefore, the fruit and vegetable slurry fermentation process will produce more energy with higher biomass temperature, as shown in Figure 1.

The chemical content of vegetables contains compounds such as carbohydrates, proteins, lipids, organic acids, fiber, ash, K, P, Ca, Fe, Na, Mg, Mn, Cu, B, and Co in values of 5.3–5.6%, 1.7–1.9%, 0.2–0.22%, 0.1–0.13%, 0.7–0.72%, 0.9–0.92%, 640–654 ppm, 260–272 ppm, 621–640 ppm, 687–700 ppm, 789–800 ppm, 300–311 ppm, 321–333 ppm, 312–332 ppm, 598–609 ppm, and 19.46–19.54 ppm, respectively. This is in addition to vitamins such as A, B complex, C, and E [25]. The chemical content of fruits are carbohydrates, proteins, lipids, organic acids, fiber, ash, K, P, Ca, Fe, Na, Mg, Zn, Cu, B, and Co in values of 32.16–33.80%, 2.80–2.92%, 3.36–4.61%, 0.1–0.14%,

21.81–26.31%, 11.39–12.45%, 830–1620 ppm, 260–311 ppm, 660–684 ppm, 520–905 ppm, 680–698 ppm, 520–905 ppm, 922–4558 ppm, 221–231 ppm, 372–384 ppm, 613–621 ppm, and 15.42–15.66 ppm, respectively.

The chemical content of cow urine contains C, N, P, K, Fe, Al, Cu, Mn, Cu, Zn, B, Co, and water in values of 10.86–10.93%, 0.92–1.00%, 500–515 ppm, 1500–1523 ppm, 775–786 ppm, 654–663 ppm, 254–261 ppm, 312–342 ppm, 231–239 ppm, 286–291 ppm, 456–459 ppm, 23.42–23.78 ppm, and 95%, respectively [27]. Both cow urine and vegetable/fruit slurries contain carbohydrates, which are chemical compounds made up of functional hydroxyl groups. In addition, they both contain proteins, which consist of polypeptide chains made from the combination of various amino acids through peptide bonds [28].

The fat or fatty acids contained in fruit and vegetable slurries are unbranched in shape and have an even number of C atoms. Metals in fruit and vegetable slurries and cow urine are bound by compounds or molecules with metal bonds. Other minerals are bound by compounds or molecules with ionic bonds.

The carbohydrates in vegetable and fruit slurries have more complex chemical bonds than those in cow urine. This can impact the fermentation reaction, producing more energy and increasing the temperature of the biomass in the bioreactor, as shown in Figure 1.

The slurry of fruit and vegetable waste contains higher amounts of carbohydrates, proteins, and fats than cow urine. This leads to a greater heat of fermentation and higher fermentation temperature, as shown in Figure 1.

Physically the slurry of vegetable and fruit waste has a specific gravity, viscosity, and water content values of 1.12 g/mL, 3.6±0.2 cP, and 80–87%, respectively. Cow urine has specific gravity, viscosity, and water content values of 1.008 g/mL, 2.0±0.1 cP, and 95% [30]. The slower flow of air bubbles in the slurry compared to cow urine is due to the higher specific gravity and viscosity of the slurry. This resulted in less oxygen absorption and a faster fermentation reaction with a higher heat of reaction in the slurry than in cow urine. Consequently, the slurry temperature during the fermentation process in treatment SOUD, SCBD, and SFBD is higher than in USOD, UCBD, and UFBD, as shown in Figure 1.

The temperature of the biomass during the fermentation process (Figure 1) also describes the level of oxygen uptake. The order of oxygen absorption by biomass from lowest to highest was USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatment. These observations suggest that the size of air bubbles is inversely proportional to the oxygen uptake by the biomass in both slurry and cow urine.

The inclusion of molasses, aeration, and urea can expedite the process of fermentation, specifically, the decomposition of organic material, reducing the usual timeframe of 14 days to a more rapid range of 6 to 9 days [29]. Figure 1 shows the changes in biomass temperature at the fermentation stage.

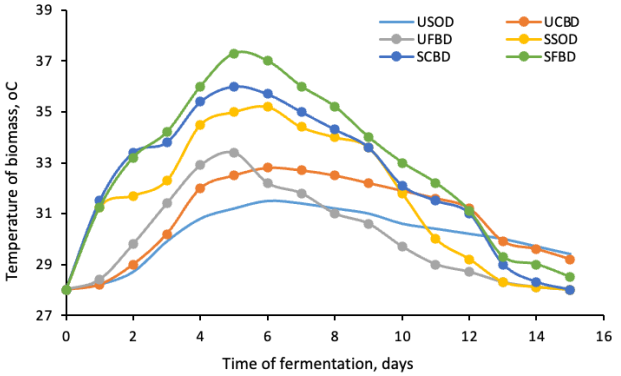


Figure 1 The association between fermentation time and temperature

The temperature increases in the fermentation process when $Q_R > Q_L$ on days 0 to 6, while the temperature in the biomass decreases from days 6 to 15 when $Q_R < Q_L$. The heat for increasing the biomass temperature for USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments was 2300±32 J, 2432±43 J, 2546±54 J, 2868±42 J, 3090±22 J, and 3456±37 J. The quantity of thermal energy required to raise the temperature aligns with the findings presented in the research conducted by Liu *et al*. [15]. Figure 1 illustrates the temperature dynamics of the biomass in the bioreactor over a 15-day fermentation period for the six treatments investigated, namely USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatment are 31.5°C, 32.0°C, 32.2°C, 35.2°C, 35.7°C, and 37.0°C, respectively, for the respective treatments.

The peak temperature observed during the fermentation reaction of cow urine with single bubble slurry treatment was lower compared to the treatments with the course and fine bubbles. Notably, the fine bubble fermentation treatment resulted in the highest temperature. Generally, there was a temperature variance of 0.13–1.15°C between a single bubble and a course bubble of 0.13–1.15°C, while the temperature difference between the treatment of a coarse bubble and a fine bubble was 0.03–1.1°C. The type of bubbles used in the fermentation process influenced the oxygen adequacy factor in the biomass [28]. Therefore, the bioreactor employing a fine bubble fermentation reaction exhibits enhanced oxygenation compared to treatments with coarse or single bubbles, resulting in more complete oxygenation of the biomass. Moreover, fine bubble treatment also evenly distributed oxygen in the biomass, promoting more active microbial growth in aerobic fermentation. Bubbles in the bioreactor can result in the utilization of approximately 86% of the available oxygen for the fermentation reaction [30].

The dissimilarity in temperature among the cow urine and slurry treatments ranged from 0.33°C to 4.96°C. This is primarily influenced by the more

complex chemical compounds in the slurry, leading to a higher energy production and biomass temperature. Meanwhile, the heat lost to the environment is 12 to 23% or for the treatment of USOD, UCBD, UFBD, SSOD, SCBD, and SFBD are 72.21–1104 J, 72.21–1242 J, 72.21–1945 J, 99.46–1311 J, 99.46–5852 J, and 99.46–2116 J, respectively. The semi-continuous fermentation process in the bioreactor is deemed effective based on the minimal heat loss to the environment.

Referring to the ANOVA test, the temperature of the fermentation between the treatments was significantly different. This indicated that the chemical composition, structure, and physical properties of the biomass as well as the type of bubble used for fermentation (single, coarse, or fine) had a significant impact on the process. The Duncan test revealed that the SFBD treatment was the most effective.

3.2 Acidity (pH)

Figure 2 shows the pH values of biomass fermented with various treatments, namely USOD, UCBD, UFBD, SSOD, SCBD, and SFBD. During the 15-day fermentation process of urine and slurry, the pH of the biomass initially increased from days 0 to 4, followed by a decrease from days 4 to 15. The increase in pH during the first stage indicates a demineralization process, whereby organic matter is broken down into cations and anions. Due to the dominance of cations, the pH rises. While the decrease in pH at the latest stage corresponds to the formation of organic acids.

The pH of the biomass showed a significant increase on the fourth day of the fermentation process due to the demineralization of microelements such as K^+, Mg^{2+}, Ca^{2+}, Na^+, Fe^{2+}, and Al^+. These cations attach to the acids produced in the fermentation, leading to an increase in the pH of the biomass process [31].

The drop in pH observed during the fermentation was attributed to the breakdown of organic substances by microorganisms into simpler compounds. This included organic acids, during the molasses decomposition and LOF ripening [6]. As a result, the release of cations reduced while the production of acids increased, which led to a decrease in pH. The release of alkaline NH_3 gas was also observed, as reported in [32].

The pH difference between urine and slurry biomass during the fermentation process ranges from 0.1 to 0.55, while that between the coarse and single bubble treatments is between 0.05 to 0.15. Furthermore, the disparity in pH among the fine and coarse bubble treatments is relatively 0 to 0.2. The variation in pH can be attributed to the changes in the chemical composition of the slurry and urine. Slurry typically conceives higher metal nutrients than cow urine, resulting in a pH closer to neutral. The differences in pH among fermentation treatments using single, coarse, and fine bubbles are related to the availability and distribution of oxygen in the biomass. This affects the complete chemical reactions in the fermentation.

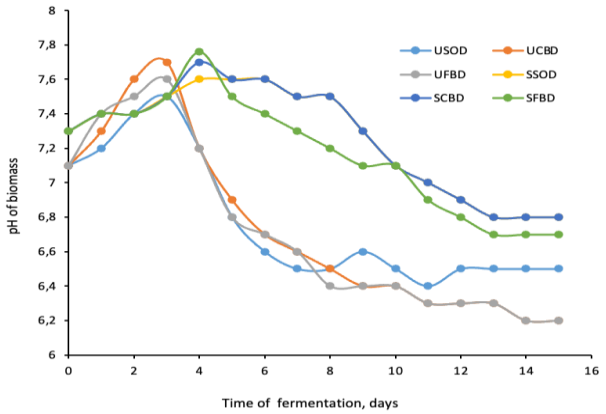


Figure 2 The relationship between fermentation time and pH

The pH values of biomass were subjected to a 5% confidence interval ANOVA test to decide whether there were significant changes among the USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments. The results revealed a significant difference between these treatments. Furthermore, Duncan test was used to identify the best treatment, which was found to be UFBD based on the pH values of the biomass during and after the fermentation processes.

In accordance with the Indonesian Ministry of Agriculture No: 261/KPTS/SR. 310/M/4/2019 [33], the pH values of bio-slurry and bio-urine obtained from the fermentation using USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments still complies with the SNI standard for LOF pH, which ranges from four to nine (Table 1). Specifically, the pH values of bio-urine and bio-slurry falls within the range of 6.3 to 6.9, and 6.7 to 6.8, respectively.

It can be observed from the transformations in the pH of the biomass that the bioreactor is capable of effectively producing urine and slurry fermentation. As a result, the end product obtained in the form of LOF complies with the SNI standards, with a pH ranging from 6.2 to 6.8 [33].

3.3 Total Dissolved Solid (TDS)

TDS increase rate of slurry fermented with USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments was observed to be within 31 to 888 ppm/day, 11 to 976 ppm/day, 43 to 1578 ppm/day, 20 to 1165 ppm/day, 10 to 2368 ppm/day, and 22 to 2147 ppm/day respectively [34]. During the zeroth to seventh day of biomass, the TDS value increased at a rate of 462±27, 525±12, 652±16, 761±27, 970±17, and 1050±28 ppm/day, for each treatment, respectively. Meanwhile, for days 8 to 15, the TDS increase rate on USOD, UCBD, UFBD, SSOD, SCBD, and SFBD were 171±8, 140±12, 52±6, 182±5, 75±7, and 47±8 ppm/day respectively [35]. The rise in Total Dissolved Solids (TDS) value is significantly impacted by the chemical

composition of the raw materials sourced from both slurry and urine. The dissimilarity in the average TDS value between the slurry biomass of traditional market organic waste and cow urine is 189±12 ppm/day. Whereas, the difference between the single and coarse bubble treatments was 25±3 ppm/day, and that between fine and coarse bubble treatments was 53±2 ppm/day. TDS value increased by 2.82 to 2.95 times in the treatment of cow urine fermentation, whereas for the bio-slurry organic waste in traditional markets, it increased from 3.32 to 4.05 times, which was better than the results obtained by Paramita *et al.* [28] and Marickar *et al.* [36].

On day six of the treatments, the highest elevation in Total Dissolved Solids (TDS) values was noted, which aligned with the initial fermentation process that resulted in the dissolution of micro-element minerals such as Al^{+}, K^{+}, Mg^{2+} Fe^{2+}, Ca^{2+}, and Na^{+}, in the biomass. The TDS expansion was attributed to the supplementation of molasses, as research showed that its use in the aerobic fermentation process of organic matter increased the TDS value due to the presence of micro and macronutrients in the biomass (Figure 3).

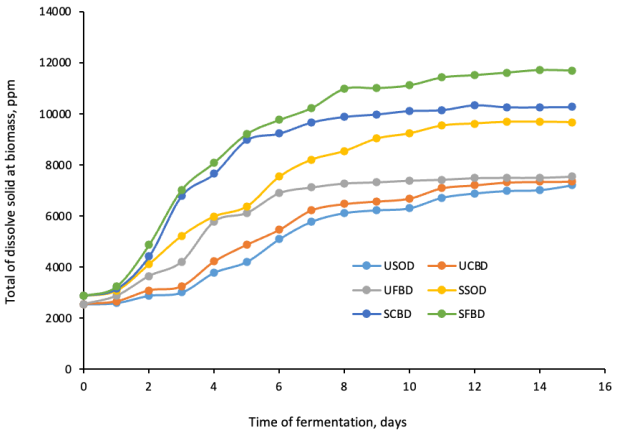


Figure 3 The association between fermentation time and total dissolved solid

The ANOVA test results showed a significant difference in TDS value of the treatments (USOD, UCBD, UFBD, SSOD, SCBD, and SFBD) during the fermentation and at the end of the process. Duncan test results revealed that SFBD was the best treatment for TDS value. The bioreactor was found to be efficient in the aerobic fermentation process of biomass [34].

3.4 Electrical Conductivity (EC)

The variation in the Electrical Conductivity (EC) value of the biomass throughout the fermentation process is regarded as a pivotal indicator of the nutrient in the fermented material. The EC value is primarily composed of positively and negatively charged ions dissolved in water, as stated by Marickar *et al.* [36]. The shifts in the EC value of the biomass during aerobic fermentation with treatments such as 3118±45 µmS/day were similar to the pattern observed in the TDS value.

During the first seven days, there was a rapid increase in the EC value shown in Figure 4, followed by a slight increase from days 8 to 15. The average increase in EC values for the following fermentation treatments USOD, UCBD, UFBD, SSOD, SCBD, and SFBD were 3118±45, 3185±35, 3551±15, 3631±17, and 3707±21 µmS/day, consecutively. The increase in EC values for each treatment during the first seven days were 6625±42, 6768±15, 6825±15, 7545±37, and 7878±21 µmS/day [37].

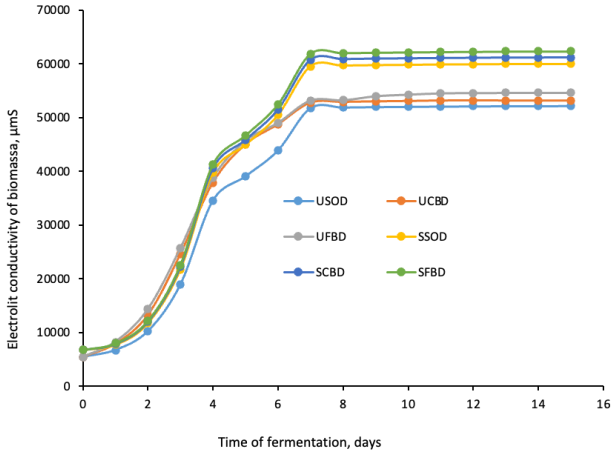


Figure 4 The relationship between fermentation time and electrical conductivity

For each treatment, the ripening mass in LOF showed an increase in EC values: 170±25, 89±8, 70±15, 67±15, and 62±45 µmS/day. The chemical composition of the raw materials from slurry and urine strongly influenced the increase in EC value [17, 36, 38], with a difference of 435±15 mS/day between the two. The difference in the average increase in EC value between single and coarse bubble treatments was 73±3 µmS/day, while that between fine and coarse bubble treatments was 86±5 µmS/day.

The change in the EC value of biomass treated with USOD, UCBD, UFBD, SSOD, SCBD, and SFBD during the fermentation process was similar to that observed in the TDS value. The ANOVA test results revealed that the EC value during the fermentation process and the final outcomes of the USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments were highly significant. The best treatments based on the TDS value was SFBD, which indicated the bioreactor functioned optimally in the aerobic fermentation process of biomass.

3.5 Biomass BOD

Shifts in BOD value reflect the quantity of oxygen consumed by microorganisms during the decomposition of organic matter in biomass, resulting in the formation of simpler compounds and energy

[28]. In aerobic fermentation with limited oxygen availability, microorganisms exhibit increased efficiency in breaking down organic matter to synthesize cells and achieve optimal growth [31, 37]. Figure 5 illustrates the dynamic changes in Biological Oxygen Demand (BOD) values during the fermentation process of cow urine and slurry derived from traditional market organic waste.

During the fermentation process, the conversion of complex compounds in biomass into simpler ones is indicated by the changes in oxygen levels, or the BOD values. The BOD values of USOD, UCBD, UFBD, SSOD, SCBD, and SFBD fermentation treatments exhibited an average decrease of 0.42±0.03, 0.43±0.03, 0.43±0.01, 0.39±0.01, 0.47±0.03, and 0.49±0.02 mg/L, each.

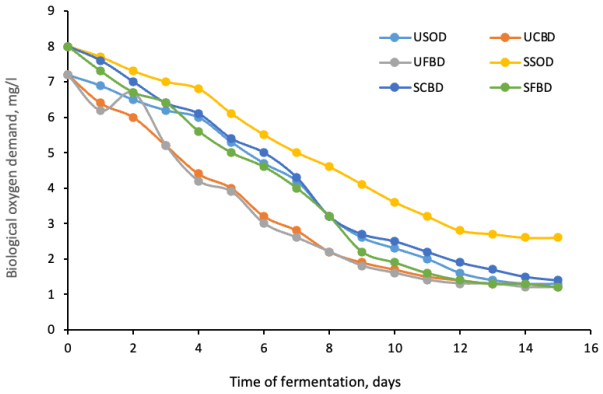


Figure 5 The association between fermentation time and biological oxygen demand

Bubble columns are commonly used in diverse endeavors such as chemical, petrochemical, biochemical, and metallurgical as efficient contactors and reactors [18, 39]. The chemical make-up of the raw materials, which include slurry and urine, has a significant impact on the reduction in BOD value during the fermentation process. The average decrease in BOD value is 0.89±0.02 mg/L, which differs between the two raw materials. The difference in BOD value reduction between the treatment of the single and coarse bubbles is 0.74±0.03 mg/L, while that between the fine and coarse bubble treatments is 0.18±0.02 mg/L.

The ANOVA test results showed that the BOD values during the fermentation process and the final outcomes of the USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments were highly significant. Based on the Duncan test categorization of urine and slurry fermentation from traditional market organic waste, the SFBD treatment was found to be the most effective. This indicated that the bioreactor was capable of operating efficiently in the aerobic fermentation process of these two types of biomasses in question [18].

3.6 Microbial Population

The graph in Figure 6 shows the mean growth of microbial population during the fermentation of slurry and cow urine using different treatments: USOD, UCBD, UFBD, SSOD, SCBD, and SFBD. Microbial growth is significantly influenced by biomass's oxygen and nutrient availability [28]. Therefore, the single air bubble column had the lowest microbial population compared to other fermentation treatments. The fermentation treatment with a fine air bubble column had the highest microbial population, as shown in Figure 6.

During the fermentation process, the microbial population initially increased from day 0 to 9 and decreased thereafter. The highest microbial populations recorded for each treatment were 5±0.3, 5.4±0.3, 6.2±0.2, 5.9±0.1, 6.5±0.3, and 7.3±0.2 log CFU. The type of bubble used and the biomass had an impact on the availability of oxygen, and the nutrient content, leading to differences in the microbial population during the fermentation for USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments, ranging from 3.2-5.0, 3.2–5.4, 3.2–6.2, 3.6–5.9, 3.6–6.5, and 3.6–7.3 log CFU each.

The ANOVA test results showed significant differences in the microbial population among the various biomass fermentation treatments. The SFBD treatment showed the best results according to the Duncan test.

The presence of nutrients and suitable environmental conditions strongly influence microbial populations [40]. In the context of traditional market slurry organic waste biomass and cow urine, there was a difference of 0.69±0.03 log CFU in microbial population [41]. The size of the air bubbles produced by single, coarse, and fine bubble diffusers affects their impact. The diameter of the air bubbles produced by each diffuser is 8 mm, 3 mm, and 1 mm. Air bubbles with a smaller diameter (1 mm) are more efficient in delivering oxygen to the fermented biomass and supporting microbial reproduction [39]. The microbial population showed a difference of 0.25±0.02 log CFU between the single and coarse bubbles, while that between the coarse and fine bubbles was 0.45±0.02 log CFU.

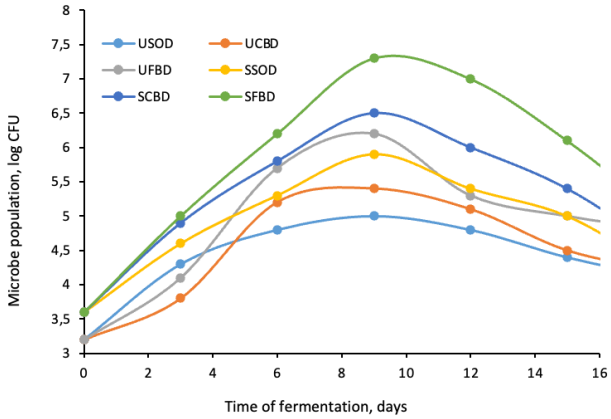


Figure 6 The association between fermentation time and microbial population

The population of non-pathogenic microbes observed at the end of the fermentation process for organic slurry raw materials and cow urine (Figure 6) exceeds the SNI standard shown in Table 1. In the same vein, the number of pathogenic microbes is lower than what is permitted by the SNI standard, as shown in Table 1 [33].

Several factors strongly influence the rate of microbial population development, during the bioreactor fermentation of cow urine and organic waste slurry. These include the initial C/N ratio of the substrate, pH of the biomass, fermentation temperature, and dissolved oxygen availability in the biomass. Among these factors, the fine bubble treatment yields the highest microbial population. This is due to the abundant and evenly distributed oxygen supply to the bioreactor. However, controlling variables such as oxygen and moisture contents and temperature during the fermentation process is challenging, which are crucial for supporting the growth of biomass-decomposing microbes in producing high-quality LOF [41].

3.7 C/N Ratio

Figure 7 shows the C/N shifts in cow urine and slurry biomass for six fermentation treatments, namely USOD, UCBD, UFBD, SSOD, SCBD, and SFBD. The C/N value of the slurry biomass and cow urine dropped from 42 to 14.3±0.2, and 32 to 13.9±0.1, respectively on day 9. From days 9 to 15, the C/N values for USB fermentation treatment, UCBD, UFBD, SSOD, SCBD, and SFBD remained stable at 13.6±0.2, 13.4±0.1, 12.6±0.2, 14.1±0.2, 13.2±0.2, and 12.4±0.1, respectively.

An ANOVA test was conducted to examine the effect of USOD, UCBD, UFBD, SSOD, SCBD, and SFBD on C/N values during the fermentation process. The test results indicated significant differences between the treatments. In addition, Duncan test revealed that UFBD was the most effective treatment.

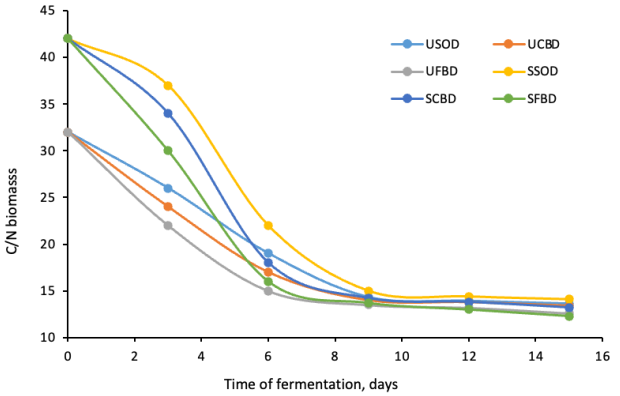


Figure 7 The association between fermentation time and C/N ratio

According to Garcia-Ochoa and Gomez [43], the primary factor for the successful fermentation of biomass is oxygen availability, which is facilitated by aeration using bubbles. The role of column bubbles is to ensure even distribution and absorption of oxygen by the biomass, thereby supporting the complete fermentation reaction of organic liquid fertilizer. A perfect fermentation reaction results in a smaller C/N value at the end of the process and a rapid decrease on days 0 to 9 [10].

Fermenting cow urine in a bioreactor without an aerator yields a C/N ratio of 1.96–2.05/day [12]. On the other hand, the rate of change in C/N value of the biomass for fermenting cow urine and slurry in a bioreactor with an aerator is between 2.04–2.11/day and 3.10–3.14/day from days 0 to 9. The C/N value of biomass for fermenting cow urine and slurry at a bioreactor with an aerator from day nine to 15 is lower, ranging from 0.6–0.9/day and 0.9–1.4/day, respectively.

The data presented in Figure 7 shows that the average C/N values underwent a change over time for the various fermentation treatments, including USOD, UCBD, UFBD, SSOD, SCBD, and SFBD. Specifically, the average C/N values changed at a rate of 1.23, 1.24, 1.29, 1.86, 1.92, and 1.98/day, in order [41].

The variance in the rate of decrease in C/N values can be attributed to the higher viscosity of slurry in comparison to urine. This disparity affects the dissolution of oxygen in urine and its availability in the biomass, ultimately impacting the fermentation reaction rate.

The dissimilarities in the reduction of C/N values are due to the sufficient supply of oxygen during urine fermentation. This is enabled by the bubble column present in the bioreactor, which supplies oxygen and influences the hydrodynamics of the biomass, allowing for a steady temperature range of 28 to 34°C [43, 44]. Contrarily, aerobic fermentation needs 192 g of oxygen to oxidize 1 mole of glucose (which decomposes into CO_2 and H_2O). Nitrosomonas and Nitrobacter catalyze the oxidation of NH^{4+} to NO^2 and NO^3, which breaks down the protein in cow urine. The oxygen demand for biomass decomposition decreased within 9 to 15 days, leading to a C/N value reduction of 0.1 to 0.15 each day. The decrease in C/N value for organic slurry is higher ranging from 0.15 to 0.23 each day. This drop in C/N is caused by a decline in the number of microbes, an increase in N from nitrification, and a drop in C from CO_2 gas loss from biomass.

3.8 Nutrient Content of Bio-Urine and Bio-Slurry

Table 1 displays the nutritional value of organic waste slurry from the traditional market and fermented cow urine. Generally, the fermentation process in a bioreactor with USOD, UCBD, UFBD, SSOD, SCBD, and SFBD treatments can produce LOF. Additionally, it has a total organic matter content, CEC, EC, TDS, BOD, and pH values of 12.85–22.46%, 26.96–29.98 me/100g, 52190–62369.24 µmS, 7211–11700 ppm, 1.2–2.6 mg/L and near to neutral or 6.3–6.8, respectively [45-47].

Table 1 Nutrient content in bio-urine and bio-slurry

	Variable of LOF	A Single Bubble	A Coarse Bubble	A Fine Bubble	SNI Standard
Bio-urine	Content of carbon, %	10.86 ± 0.18[a]	10.85 ± 0.11[a]	10,33 ± 0.06[b]	> 10
	Content of nitrogen, %	0.8 ± 0.02[b]	0.81 ± 0.06[b]	0.82 ± 0.08[a]	> 0,5
	Content of P_2O_5, ppm	786 ± 17[a]	774 ± 11[b]	792 ± 18[a]	Total of (N + of P_2O_5 + K_2O,) is 2 to 6 %
	Content of K_2O, ppm	500 ± 13[c]	652 ± 19[b]	6786 ± 17[a]	Total of (N + of P_2O_5 + K_2O,) is 2 to 6%
	Content of Mn, ppm	279 ± 12[c]	283 ± 17[b]	299 ± 19[a]	250–500
	Content of Cu, ppm	178,2 ± 27[b]	185,4 ± 11[b]	188,8 ± 11[b]	250–500
	Content of Zn, ppm	254 ± 11[d]	278 ± 8[c]	286 ± 14[c]	250–500
	Content of Fe, ppm	420 ± 17[e]	483 ± 12[d]	621 ± 18[c]	90–900
	Content of B, ppm	383 ± 11[c]	387 ± 17[c]	442 ± 15[b]	125–2500
	Content of Co, ppm	12.85 ± 1.2[b]	13.35 ± 1.4[b]	19.46 ± 2.2[a]	2–20
	CEC, me/100 g	27.7 ± 1.2[b]	28.9 ± 2.0[b]	29.98 ± 2.1[a]	> 25
	EC, µmS	52190 ± 132[c]	53200 ± 1632[b]	54630 ± 112[a]	>5000
	TDS, ppm	7211 ± 54[b]	7356 ± 32[b]	7543 ± 44[b]	>5000
	BOD, mg/L	1.3 ± 0.08[b]	1.2 ± 0.13[b]	1.2 ± 0.03[b]	<2,0
	pH	6.5 ± 0.12[a]	6.3 ± 0.17[b]	6.3 ± 0.11[b]	4–9
	C-N	13.6 ± 0.03[c]	13.4 ± 0.04[b]	12.6 ± 0.02[a]	<15
	Population microbe non pathogen, CFU	4.1 x $10^{5\,b}$	4.2 x $10^{5\,b}$	4.8 x $10^{5\,a}$	>5.0 x 10^3
	Population microbe pathogen, CFU	not detected	not detected	not detected	<1.0 x 10^2
	Variable of LOF	Single Bubble	Coarse Bubble	Fine Bubble	SNI Standard
Bio-Slurry	Content of carbon, %	10,72 ± 0.02[c]	10,16 ± 0.01[d]	10,71 ± 0.03[c]	> 10
	Content of nitrogen, %	0,76 ± 0.03[b]	0,77 ± 0.01[b]	0,82 ± 0.03[a]	> 0,5
	Content of P_2O_5, ppm	722 ± 12[b]	781 ± 13[b]	792 ± 18[b]	Total of (N + of P_2O_5 + K_2O,) is 2 to 6%
	Content of K_2O, ppm	611 ± 32[c]	641 ± 12[c]	671 ± 15[b]	Total of (N + of P_2O_5 + K_2O,) is 2 to 6%
	Content of Mn, ppm	299 ± 32[a]	307 ± 32[a]	321 ± 32[a]	250–500
	Content of Cu, ppm	318 ± 2[a]	326 ± 1,6[a]	322 ± 2,2[a]	250–500
	Content of Zn, ppm	286 ± 32[c]	334 ± 17[b]	420 ± 32[a]	250–500
	Content of Fe, ppm	876 ± 35[b]	898 ± 32[a]	896 ± 27[a]	90–900
	Content of B, ppm	642 ± 32[a]	613 ± 37[a]	609 ± 22[a]	125–2500
	Content of Co, ppm	17.46 ± 2.2[a]	19.61 ± 2.0[a]	19.77 ± 2.4[a]	2–20
	CEC, me/100 g	26.96 ± 2.6[c]	28.23 ± 2.7[a]	29.69 ± 2.3[a]	> 25
	EC, µmS	60018 ± 38[a]	61218 ± 25[a]	6236 ± 15[a]	>5000
	TDS, ppm	9670 ± 15[a]	10276 ± 37[a]	11700 ± 21[a]	>5000
	BOD, mg/L	1.6 ± 0.12[a]	1.4 ± 0.12[a]	1.2 ± 0.2[b]	<2,0
	pH	6.8 ± 0.25[a]	6.8 ± 0.22[a]	6.7 ± 0.17[a]	4–9
	C-N	14.1±0.19[a]	13.2±0.17[a]	12.3±0.22[b]	<15
	Population microbe non pathogen, CFU	4,2 x $10^{5\,b}$	4,7 x $10^{5\,a}$	5,8 x $10^{5\,a}$	>5,0 x 10^3
	Population microbe pathogen, CFU	not detected	not detected	not detected	>1,0 x 10^2

Note: LOF SNI Standard [32]; Different letters in the same row and variable indicate differences in the value of these variables between treatments

When cow urine and slurry from conventional market organic waste are fermented using a fine bubble treatment, LOF is created that is superior to other types and meets SNI standards. The value of macro nutrient (Mg, C, P, N, Ca, S, and K) and micro nutrient (Al, Co, B, Fe, Cu, Mn, and Zn) for bio-urine were 5.882±0.27% and 0.135±0.02%. While those for bio-slurry were 6.51±0.23% and 0.14±0.015% [48]. The macro and micro nutrients content in the liquid organic fertilizer produced through a single bubble,

coarse bubble, and fine bubble treatment is higher than the SNI standard for organic fertilizer [33], [49].

The amount of oxygen in cow urine and organic waste slurry when they are combined in a bioreactor with various aerator types, namely single bubble, coarse bubble, and fine bubble, directly affects the fermentation effectiveness and microbe growth. This, in turn, affects the bioreactor's productivity as a tool for producing LOF-containing micro and macro nutrients. The amount of micro and macronutrients in the LOF increases with the oxygen content of the biomass. Therefore, the fine bubble treatment is preferred over the coarse bubble and single bubble diffuser treatments for fermenting cow urine and traditional market organic waste slurries, as it produces LOF with higher nutrient content. Imperfect aerobic reactions during fermentation may produce gases such as CH_3S, H_2S, S_2, and $(CH_3)_2$, gases, aliphatic acids, as well as CO [421], leading to unpleasant odors and reduced nutrition in LOF.

4.0 CONCLUSION

In conclusion, the pH, temperature, TDS, EC, BOD, microbial population, and C/N variables indicated that the bioreactor's UFBD and SFBD fermentation treatments were successful in producing high-quality LOF. The resulting LOF had total organic matter content, CEC, EC, TDS, BOD, and pH values of 20.77–22.46%, 29.98 me/100 g, 54630–62369.24 µmS, 7543–11700 ppm, 1.2–2.6 mg/L, and 6.3–6.8, respectively. The bio-urine and bio-slurry both had high macro-nutrient content of 12.72 to 12.96% and 12.35–12.99%, with micro-nutrient content of around 2–3%. Among the treatments, the fermentation of cow urine with a column of fine air bubbles produced by a 250-watt aeration pump was the most effective. However, all treatments were able to produce LOF that met the SNI standards.

Conflicts of Interest

The author(s) declare(s) that there is no conflict of interest regarding the publication of this paper.

Acknowledgment

Gratitudes are expressed to Udayana University for funding this research.

References

[1] Li, D. P. and Z. J. Wu. 2018. Impact of Chemical Fertilizers Application on Soil Ecological Environment. *J. Ying Yong Sheng Tai Xue Bao*. 19(5): 1158-1165.

[2] Pahalvi, H. N., L. R. Majeed, S. Rashid, and B. Nizar. 2021. Chemical Fertilizers and Their Impact on Soil Health. *Microbiota and Biofertilizers*. 2: 1–20. Doi: 10.1007/978-3-030-61010-4_1.

[3] Kilande G., J. S. Tenywa, M. C. Rwakaikara-Silver, and A. A. Katushabe. 2016. Cattle Urine as a Fertilizer: Micro-biochemical Changes in Fermenting Cattle Urine and Implications on Plant Nutrient Conservation. *British Microbiology Research Journal*. 11(2): 1-10.

[4] Sastro, Y. and B. Bakri. 2018. The Effect of Fermentation Method, Microbes Inoculation and Carbon Source Proportion on the Quality of Organic Fertilizer Made from Liquid Wastes of Chicken Slaughterhouse. *Journal of the Indonesian Tropical Animal Agriculture*. 38(4): 257-263 Doi: 10.14710/jitaa.38.4.257-263.

[5] Pramana, K. S., Y. Setiyo, I G. N. A. Aviantara. 2019. Optimalisation of Cow Urine Fermentation Process. *Jurnal BETA* 7(1): 153-158. https://doi.org/10.24843/JBETA.2019.v07.i01.p05.

[6] Paramita, P., M. Shovitri, and N. D. Kuswytasari. 2012. Market Organic Waste Biodegradation Using Natural Microorganisms in Septic Tanks. *ITS Science and Arts Journal*. 1: E23-E26,

[7] Dalzell, H. W., A. J. Riddleston, K. R. Gray, and K. Thuenrairajan. 1987. Soil Managemen Compost Production and Use in Tropical and Subtropical Environment. FAO. Rome.

[8] Aritonang, M., Y. Setiyo, I. B. P. Gunadnya. 2013. Optimalisation Cow Urine Fermentation Process Become to Bio-urine. *Jurnal BETA*. 1: 268-279.

[9] Suhartana, I P. G., Y. Setiyo, and I W. Widia. 2015. Study of Sludge Cow Manure Fermentation Process. *Jurnal BETA*. 5(1): 12-22.

[10] Mudiarta, I. M., Y. Setiyo, and I. W. Widia, 2018. Study Bioslurry of Bio-slurry from Cow Manure Fermentation Process with Adition Mollases. *JITP Agrotechno*. 3(1): 277-283. https://doi.org/10.24843/JITPA.2018.v03.i01.p03.

[11] Putra J. K. G., Y. Setiyo, I. N. Sucipta. 2022. Impact of Adition Nitrosomonas Bacterial at Cow Urine Fermentation to Produce Liquid Organic Fertilizer. *Jurnal BETA*. 10(1): 12-22. Doi: https://doi.org/10.24843/JBETA.2022.v10.i01.p02.

[12] Wirawan I. W. E., Y. Setiyo, I. A. G. B. Madrini. 2021. Study of Vegetable and Fruit from Traditional Marked with Fermentation. *Jurnal BETA*. 9(2): 268-279. Doi: https://doi.org/10.24843/JBETA.2021.v09.i02.p14.

[13] Mohanty I., M. R. Senapati, D. Jena, and S. Paalai. 2014. Diversified Uses of Cow Urine. *International Journal of Pharmacy and Pharmaceutical Sciences*. 6(3): 20-22.

[14] Nuraini Y., and Asgianingrum. 2017. Improving the Quality of Cattle Biourin with the Addition of Biofertilizers and Molasses and their Effect on Growth and Productivity of Pakchoy. *Journal of Indonesian Horticulture*. 8(3): 183-191. Doi: 10.29244/jhi.8.3.183-191.

[15] Liu, C., Y. Sun, N. Li, B. Zhang, and F. Zhen. 2019. Impact of Temperature Fluctuation on Anaerobic Fermentation Process of Upgrading Bioreactor under Solar Radiant Heating. *Applied Thermal Engineering*. 156: 382-391. https://doi.org/10.1016/j.applthermaleng.2019.04.092.

[16] Setiyo, Y., N. L. Yulianti, P. B Sanjaya, and I. B. W Gunam. 2021. The Impact of Calorage Changes on Bio-urine Quality from Aerobic and Anaerobic Fermentation Process in a Bioreactor. *Journal of Global Biosciences*. 10(4): 8512–8529.

[17] Jiang, Y., J. Meiting, L. Weizun, R. Qingbin, L. Liu, Y. Chen, Q. Yang, Q. Hou, and Y. Liu. 2015. Rapid Production of Organic Fertilizer by Dynamic High-temperature Aerobic Fermentation (DHAF) of Food Waste. *Biores. Technology*. 197: 7-14. Doi: 10.1016/j.biortech.2015.08.053.

[18] Kantarcia, N., F. Borak, and K. O. Ulge. 2005. Bubble Column Reactors. *Process Biochemistry*. 40: 2263-2283. https://doi.org/10.1016/j.procbio.2004.10.004.

[19] Dunn, I. J. 2003. *Biological Reaction Engineering: Dynamic Modelling Fundamentals with Simulation Examples*. Weinheim: Wiley-VCH Verlag. 508. Doi: 10.1002/3527603050

[20] Crush, F. C. A. Marioucos, and A. M. Bilton. 2022. Experimental Characterization of an Oxygen Transfer Model of a Fine Pore Diffuser Aerator. *Aquacultural Engineering*. 98. Doi: 10.1016/j.aquaeng.2022.102259.
[21] Silva, S., C. Baffi, S. Spalla, C. Cassinari, and P. Lodigiani. 2010. Method for the Determination of CEC and Exchangeable Bases in Calcareous Soils. *Agrochimica*. 54(2): 103-114.
[22] Anon. 2009. Food and Agricultural Organization of United Nations. Standard Operating Procedure for Soil Organic Carbon. Global Soil Partnership, Manila, Philippines.
[23] Anon. 2018. Nitrogen Determination by Kjeldahl Method. PanReac, AppliChem, ITW Reagen. https://www.itwreagents.com/uploads/20180114/A173_EN.pdf.
[24] Sherrell, C. G. 1970. Comparison of Chemical Extraction Methods for the Determination of "Available" Phosphate in Soils. *New Zealand Journal of Agricultural Research*. 13(3): 481-493. Doi: 10.1080/00288233.1970.10421597.
[25] Chouchan, S. 2015. Enumeration and Identification of Standard Plate Count Bacteria in Raw Water Supplies. *OSR Journal of Environmental Science, Toxicology and Food Technology*. 9(2): 67-73. Doi: 10.9790/2402-09226773.
[26] Holman, J. P. 1997. *Transfer of Heat*. Sixth Edition, Interpretation: E, Jasjfi. Erlangga, Jakarta.
[27] Butnairu, M., and A. Butu. 2015. Chemical Composition of Vegetables and Their Products. *Handbook of Food Chemistry*. 3: 627-692. Doi: 10.1007/978-3-642-36605-5_17.
[28] Romelle, F. B., A.P Rani, and R. S. Manohar. 2016. Chemical Composition of Some Selected Fruit Peels. *European Journal of Food Science and Technology*. 4(4): 12-21.
[29] Safitri, A., A. Roosdiana, A. Srihardyastutie, and Masruri. 2018. Fermentation of Cow Urine Collected from Ngabab Village, Malang: Its Potential as Liquid Fertilizer. *IOP Conf. Series: Earth and Environmental Science*. 239(2019): 012029. IOP Publishing. Doi: 10.1088/1755-1315/239/1/012029.
[30] Lübbert, A. 1987. Bubble Columns and Airlift Bioreactors Biotechnology, Bubble Columns and Airlift Bioreactors. https://www.eolss.net/Sample-Chapters/C17/E6-58-04-19.pdfhttps://www.eolss.net/Sample-Chapters/C17/E6-58-04-19.pdf.
[31] Hosseini, M., S. A. Shojaosadati, and J. Thofighi. 2003. Application of a Bubble-column Reactor for the Production of a Single-cell Protein from Cheese Whey. *Industrial & Engineering Chemistry Research*. 42(4): 764-766. Doi: 10.1021/ie020254o.
[32] Ali, J., R. P. Singh, and V. Durgapal. 2016. Biogas Production from Different Organic Biomass Materials by Anaerobic Batch Fermentation. *American Journal of Biomass and Bioenergy*. 5(1): 43-56. Doi: 10.7726/ajbb.2016.1004.
[33] LOF SNI Standar. 2019. Decree of the Minister of Agriculture of the Republic of Indonesia No: 261/KPTS/SR. 310/M/4/2019.
[34] Gwon, B. G. and J. K. Kim. 2012. Feasibility Study on Production of Liquid Fertilizer in a 1 m^3 Reactor using Fishmeal Wastewater for Commercialization. *Environ. Eng. Res.* 17(1): 3-8. http://dx.doi.org/10.4491/eer.2012.17.1.003.
[35] Kalpesh, C. Ashara and Ketan V. Shah. 2016. Cow's Urine: An Incredible Aqueous Phase. *Global Journal of Biotechnology & Biochemistry*. 11(2): 145-152. Doi: 10.5829/idosi.gjbb.2016.11.02.104149.
[36] Marickar, F. 2010. Electrical Conductivity and Total Dissolved Solids in Urine. *Urological Research*. 38: 233-235. Doi: 10.1007/s00240-009-0228-y.
[37] Gazinsky, E., and F. Boege. 2002. Urine Screening with the UF-series Analyzers: The Use of Urine Conductivity as a Surrogate Marker of Urine Osmolality and Renal Diuresis. *Sysmex J. Int.* 12: 76-79.
[38] Degaleesan, S., M. Dudukovic, Y. Pan. 2001. Experimental Study of Gasinduced Liquid-flow Structures in Bubble Columns. *AIChE Journal*. 47: 1913-1931. https://doi.org/10.1002/aic.690470904.
[39] Setiyo, Y., I. B. W. Gunam, I. B. P. Gunadnya, K. A. Yuliadhi. 2018. Application of Chicken Manure Compost as Organic Fertilizer to Improve the Quality and Productivity of Potato (Solanum tuberosum L.). *Ecol Environ Conserv*. 24: 23-34.
[40] Setiyo, Y., I. B. W. Gunam, I. B. P. Gunadnya, K. B. Susrusa. 2017. The implementation of Low External Input Sustainable Agriculture System to Increase Productivity of Potatoes (*Solanum tuberosum* L.). *J Food Agric Environ*. 15(2): 62-67.
[41] Shoemaker, W. R., S. E. Jones, M. E. Muscarella, M. G. Behringer, B. K. Lehmkuhl, and J. T. Lennon. 2021. Microbial Population Dynamics and Evolutionary Outcomes under Extreme Energy Limitation. *Proc. Natl. Acad. Sci.* 118(33). https://doi.org/10.1073/pnas.2101691118.
[42] Bristow, A. W., D. C. Whitehead, J. E. Cockburn. 2006. Nitrogenous Constituents in the Urine of Cattle, Sheep and Goats. *J. Sci. Food Agri*. 59: 387-394. https://doi.org/10.1002/jsfa.2740590316.
[43] Garcia-Ochoa, F., and E. Gomez. 2009. Bioreactor Scale-up and Oxygen Transfer Rate in Microbial Processes: An Overview. *Biotechnology Advances*. 27: 153-176. https://doi.org/10.1016/j.biotechadv.2008.10.006.
[44] Paek, K. Y., D. Chakrabarty, E. J. Hahn. 2005. Application of Bioreactor Systems for Large Scale Production of Horticultural and Medicinal Plants. *Plant Cell, Tissue and Organ Culture*. 81: 287-300.
[45] Miah, N. A., R. U. Miah, and M. Z. Alam. 2017. Determining Chemical Composition of Cattle Urine and Indigenous Plant Extracts. *International Annals of Science*. 3(1): 23-26. Doi: 10.21467/ias.3.1.23–26.
[46] Nasir, M., M. Ghazi, R. Omar, and A. Idris. 2014. Bioreactor Performance in the Anaerobic Digestion of Cattle Manure. *Energy Sources, Part A*. 36: 1476-1483.
[47] Raden, I., S. Syarif, Fathiliah, M. Fadli, and Suyadi. 2017. Nutrient Content of Liquid Organic Fertilizer (LOF) by Various Bio-activator and Soaking Time. *Nusantara Bioscience*. 9(2): 209-213. Doi: 10.13057/nusbiosci/n090217.
[48] Rosenfeld, P. E., and C. L. Henry. 2000. Wood and Ash Control of Odor from Biosolids Application. *J. Environ. Qual.* 29: 1662-1668. Doi: 10.2134/jeq2000.00472425002900050038x.
[49] Setiyo Y., I. B. P. Gunadnya, I. B. W. Gunam, I. G. A. Lani Triani, P. Budisanjaya, and N. L. Yulianti. 2023. The Impact of Implementation of the Leisa System on the Conservation and Land Restoration of Citrus Cultivation in Bali, Indonesia. *International Jurnal of Agriculture and Biology*. 29: 181-192. Doi: 10.17957/IJAB/15.2018.