

## Physiochemical and microbial changes during organic liquid fertilizer preparation using animal and plant materials under anaerobic conditions

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**Abstract-** The recycling of organic wastes from biological fermentation processes for agricultural production is a fast development of bioconversion strategy. Microbes play a major role on physiochemical changes during Organic Liquid Fertilizer (OLF) preparation. Hence, this study investigated the Physiochemical changes such as pH, EC, TDS, nutrient availability and microbial count. The OLF was produced prepared compost, banana peels, wild sunflower leaves, and goat manure at the rates of 80g:50g:80g:80g (w/w) respectively. Also, brown sugar (25g/l of water) was added as a fermenting agent. The anaerobic fermentation was allowed up to 30 days. The results significantly ( $P < 0.05$ ) showed that, maximum EC (9.93 dS/m), TDS (6200 mg/L), lower pH (4.6), higher total nitrogen (513.4 mg/L), and total phosphorous (588.19 mg/L) at 15 DAF than at 30 DAF. Anyhow, significantly higher total potassium (6.42 mg/L) was reported at 30 DAF than at 15 DAF. Further, the increased number of microbes did not significantly influence on physiochemical changes of OLF, but s bacteria ( $14.09 \pm 0.3$ ), Actinomycets ( $16.1 \pm 0.2$ ) and fungus count ( $11 \pm 0.3$ ) were significantly increased at 30DAF. It is concluded that except for potassium level, parameters such as EC, pH, TDS, N, P and microbial performance were highly recorded at 15DAF. Hence, a short fermentation time period (15 Days) would be recommended to achieve the best physiochemical parameters during the OLF preparation.

**Keywords:** Anaerobic fermentation, wild sunflower leaves, banana peel, goat manure, organic liquid fertilizer, microbial count

### I. INTRODUCTION

The least quantity of nitrogen, phosphate and potassium (NPK) are found in organic liquid fertilizers (OLF), also depending on the nature of

biomass and methods of production, the nutrient concentration is different (Hartz *et al.*, 2010). The crop's final yield and quality are influenced by the OLFs concentration and nutritional content (Porkherl *et al.*, 2015; Succop and Newman, 2004). In several research, OLF was said to have the ability to boost plant output in numerous trails (Anila, 2009). However, it has been proposed that liquid fertilizer derived from animal sources may be used in greenhouse settings in place of chemical fertilizer (Guajardo-Rios *et al.*, 2018). However, the OLF might be created using a variety of additional organic matter sources (Shinohara *et al.*, 2011). Compost, biogas discharge, soluble fish waste, and maize steep fluid are just a few examples of the byproducts of liquid organic waste. However, some researchers mix the elements from plants and animals. Once these components have undergone anaerobic digestion, they can produce more ammonia and potassium in the liquid. However, choosing the right crop species and applying the right amount of organic fertilizer are essential for obtaining excellent yields from crops when using gardening (Chapagain *et al.*, 2010).

Thanaporn and Nuntavun, (2019) proposed that the liquid fertilizers are produced using straightforward fermentation processes employing organic waste can be used as carbon input. Microorganisms are essential to the fermentation process because they break down the substrates. Plant growth stimulants, organic acids, phytohormones, and plant growth promoters are all present in the liquid organic fertilizers (Osman *et al.*, 2009). Further, Recent study of plant-microbes relations and microorganisms that encourage plant growth have attracted more attention. These microorganisms boost growth by increasing nutrient uptake, enhancing the plant's capacity to withstand biotic and abiotic pressure, and promoting plant growth stages (Lamont *et al.*, 2017).

Anyhow, during the OLF preparation pH, Electric conductivity (EC), total dissolved solids (TDS), nutrient in the suspension and microbial counts are changed based on the different plant and animal material used. These parameters are considered as physiochemical properties of OLF. Hence, this study was designed to examine the Physiochemical changes and microbial action during OLF preparation using goat manure, banana peel, natural organic compost and wild sunflower under anaerobic microbial fermentation.

## II. MATERIALS AND METHODS

### A. Location of the study

The research was conducted in a plant net house facility at Agrotech Park in Malwatta, Faculty of Technology, South Eastern University of Sri Lanka, from January 2022 to December 2022. The experimental site is located at a latitude of 7.29° N and a longitude of 81.82° E, at a mean sea level of 20.00 m (Mubarak *et al.*, 2022). It links to the agro ecological zone of the low country dry zone, (DL2b) in Sri Lanka (Natural Resource Management Centre, Department of Agriculture, 2017).

### B. Formulation of liquid organic fertilizer

Initially, the compost was prepared with the ratios of hay: cow dung: albizia leaves (*Albizia julibrissin*) (1:3:1 v/v w/w), and the red earthworm (*Eisenia foetida*) was introduced at a rate of 150 g / 25 kg of cow dung (Abul Hashem *et al.*, 2020) then it was allowed to decompose for three months. Subsequently, the natural compost was sieved to obtain homogenous particles and used for the preparation of liquid organic fertilizer, as mentioned below.

In addition to natural compost, banana peels, wild sunflower leaves, and goat manure (80g: 50g: 80g: 80g) (w/w) were used. Banana peels were cut into 2 cm pieces and allowed for five days of sun drying, and goat manure also was subjected to five days of sun drying and sieved until it homogenous particles. These materials were fermented at ambient temperature for 30 days in cleaned 25-liter plastic containers. One third of containers were left without media. The brown sugar was used as a fermenting agent in a quantity of 25 g/L. The mixture was stirred using a wooden pole 3 times per day during the fermentation process to make a homogenous mixture. The OLF subsamples were filtered twice by using a 0.45 micron filter at 15 days after fermentation (DAF) and 30 DAF. Then samples were subjected to physiochemical and nutrient analysis.

### C. Measurement of Physio- chemical properties of organic novel liquid fertilizer

The physiochemical parameters such as pH, electrical conductivity (EC), and total dissolved solids (TDS) in liquid organic fertilizer were measured using a pH meter (OHAUS, starter 2100) and an EC meter (Mettler Toledo, Five Easy FE30-1), respectively. Total

nitrogen, total phosphorus, and total potassium were analyzed using the Kjeldahl method, wet digestion, spectrophotometer (420 nm), and flame photometer methods, respectively (Phibunwatthanawong and Riddech, 2019).

### D. Determinations of microbes count in liquid organic fertilizer

Microbial count was done for nutrient solubilizing bacteria, fungus and for Actinomycetes. Solubilizing bacteria were counted using Ashby's nitrogen free medium (Jiménez *et al.*, 2011), Actinomycete colonies counts were determined using Pochon medium method (Javoreková *et al.*, 2019) and Fungus was count using Dichloran Rose Bengal Chloramphenicol (DRBC) agar medium (Nafaa *et al.*, 2023).

### E. Data collection

The physicochemical properties, including pH, electrical conductivity (EC), and total dissolved solids (TDS), were monitored at five-day intervals over a period of one month. Additionally, the concentrations of total nitrogen, phosphorus and potassium were determined at two specific time points: 15 and 30 days after the start of the fermentation process. Sampling was carried out randomly from the fertilizer container. To ensure the reliability of the results, four replicates were performed consistently throughout the research period, both for nutrient analysis and physicochemical property measurements.

### F. Statistical method

An analysis of variance (ANOVA) was performed using Minitab 18 (Minitab HK Limited, Hong Kong) to analyze the data and a Turkey post-hock test was performed to find out the significance between the treatments at  $P < 0.05$ .

## III. RESULTS AND DISCUSSION

### A. Changes of nutrient solubilizing microbes counts

Microorganisms is an important assets that play a vital role of dissolving nutrients from insoluble minerals (Hu *et al.*, 2018). A various group of microbes, such as fungi, bacteria and Actinomycetes, have the ability to solubilize minerals by excretion of organic acids (Sarikhani *et al.*, 2018). In this study, Bacteria inclusive of K, P and N solubilizing, Actinomycetes and Fungus count were significantly increased with time interval. Higher count of those microbes were recorded in 30 days after fermentation (DAF) than 15 DAF. The microbial counts were changed from (14.09±0.3 - 21.87±0.9), (16.1±0.2 - 24.86±0.6) and (11±0.3 - 17.9±0.8) for Bacteria, Actinomycetes and fungus respectively (Table 01). This results reveled that with 30 days' time interval microbe's population were increased. Mainly microbe's population was related with carbon source use for fermentation. According to the research of

Wang *et al* ( 2023) more carbon in the sources offers a proper environment for the existence of microorganisms and has increase the microbial community, while increased number of microbes was not an indication to release of the nutrient in the effluent. The study of (Hastuti *et al.*, 2022) explain that the microorganism passes three steps such as adaptation, multiplication and balancing the numbers.

These steps involved in amount of carbon source in the material used for organic liquid fertilizer. During the balancing stage of microbes, producing microbes cell same as microbes dead. Hence in this stage even microbe’s population high and causes toxic to the environment consequently microbe’s activities becoming low.

Table 01: Changes Microbial counts during organic liquid fertilizer production

Time interval for microbial fermentation	Nutrient solubilizing microbes		
	Bacteria count *10 <sup>6</sup> (CFU/ml)	Actinomycetes count* 10 <sup>4</sup> (CFU/ml)	Fungus count* 10 <sup>3</sup> (CFU/ml)
15DAF	14.09±0.05 <sup>b</sup>	16.1±0.03 <sup>b</sup>	11±0.05 <sup>b</sup>
30DAF	21.87±0.16 <sup>a</sup>	24.86±0.1 <sup>a</sup>	17.9±0.13 <sup>a</sup>

Mean values ±SE. DAF-Days after fermentation Mean values followed by the same letter were not significantly different at the 0.05 probability level based on one-way ANOVAs followed by Turkey post-hock test within the same column, CFU-Colony forming unit, N=06.

*B. Changes of Electric conductivity (EC)*

Table 2 shows the changes in electrolysis properties of EC, pH, and TDS. The EC showed significantly increasing trends up to 15 days after fermentation (DAF), and then it tended to decrease towards 30 DAF. The maximum EC (9.93±0.009) was recorded at 15 DAF, and a lower EC was (7.42±0.009) observed at 30 DAF. It reveals that more ions are present during 15 DAF (Nemali and van Iersel, 2004) indicating, the number of ions available to plants root zone is proportional to the EC of the nutrient solution. The optimum EC varies based on the crop (Sonneveld *et al.*, 2009), and a higher EC hinder the nutrient uptake by accumulating high osmotic pressure (Signore *et al.*, 2016). Anyhow, Bustomi Rosadi *et al* ( 2014) find that the threshold hole EC of plant was 3 dSm<sup>-1</sup>.

(3603±0.7). The TDS and EC showed the same phenomenon because (Grace, 2016) found that TDS was proportionally related to the EC value of the liquid solution.

*D. pH Changes*

Putra *et al.*, 2015; Marschner *et al.*, (2011) pointed out that the plant growth and dry matter are influenced by pH of the nutrient solution. In this study, pH showed significant (P<0.05) variation to the fermentation time interval (Table 02). The acidity of organic liquid fertilizer (OLF) was significantly increased, along with a fermentation time interval of up to 15 DAF. At 15 DAF, it showed maximum acidity (pH= 4.6±0.007) and mild lower acidity was recorded at 30 DAF (5.7±0.008). Acidity was recorded due to the decomposition of organic material content and time period of fermentation by the microbial activities. Higher carbon levels in the organic materials also lead to high acidity during OLF preparation. Thanaporn *et al.*, (2019) stated that the pH of a liquid organic fertilizer depends on the carbon in the organic material used to prepare the OLF. More carbon could lead to a higher pH due to the accumulation of lactic acid and acetic acid in OLF.

*C. TDS Changes* The total dissolved solids (TDS), are charged minerals consisting of cations and anions. In this study, the peak TDS (6200.22±0.6) was recorded at 15DAF then it showed a significant reduction towards 30DAF. Lower TDS has been recorded at 30 DAF

Table 02: Changes of electrolysis properties of organic liquid fertilizer

Physiochemical properties	Days after fermentation (DAF)					
	5DAF	10DAF	15DAF	20DAF	25DAF	30DAF
EC (ds/m)	9.9±0.004 <sup>b</sup>	9.9±0.004 <sup>b</sup>	9.93±0.01 <sup>a</sup>	9.6±0.007 <sup>c</sup>	9.3±0.007 <sup>d</sup>	7.42±0.01 <sup>e</sup>
pH	5.8±0.008 <sup>a</sup>	5.51±0.005 <sup>d</sup>	4.6±0.01 <sup>e</sup>	5.5±0.007 <sup>d</sup>	5.6±0.008 <sup>c</sup>	5.7±0.01 <sup>b</sup>
TDS (mg/L)	5367.6±1.14 <sup>c</sup>	5631.7±0.9 <sup>b</sup>	6200.22±0.6 <sup>a</sup>	4971.5±0.7 <sup>d</sup>	4674±0.7 <sup>e</sup>	3603±0.7 <sup>f</sup>

Mean values ± SE. DAF- Days after fermentation. TDS- Total dissolved solids. Mean values followed by the same letter were not significantly different at the 0.05 probability level based on one-way ANOVAs followed by Turkey post-hock test within the same column, N-06

*E.. Changes of total nitrogen content*

During OLF preparation, a significant (P<0.05) variation in total nitrogen concentration was observed with time interval. At 15 DAF, higher total nitrogen (513.4 ± 0.74) was recorded than at 30 DAF (329.1±1.81) in the prepared OLF. (Table 3). According to Tantrip *et al.*, (2016), total nitrogen availability in OLF is influenced by the organic carbon material used to prepare the OLF. Also, the performance of nitrifying bacteria (*Rhizobium mayense* sp) depends on the organic material available in OLF. Further, according to Indriani, (2011) finding, fermentation time affects the quality of OLF, and (Thoyib, 2016) proved that higher nitrogen content was recorded at 17 days after fermentation during OLF preparation.

*F. Changes of total phosphorous content in OLF* The total available phosphorous in OLF was significantly decreased with the fermentation time interval. The peak amount was achieved at 15 DAF (588.19±0.6), while a significantly reduced the amount of phosphors was recorded at 30 DAF (64.53±0.53). This result suggests that phosphorous-solubilizing bacteria were actively performing at 15 days of fermentation. The dissolution of phosphate bacteria was high due to the availability of carbon in OLF (Suresh Kumar *et al.*, 2013). After 30 days of fermentation, bacteria might reduce their activity due to a lack of carbon. Surtinah, (2013) reported that high carbon content is needed for maximum performance of phosphorus solubilizing bacteria. Further, Santi, (2008) summarized that lower

phosphorus was recorded due to the low carbon material during OLF preparation. Hence, due to peak organic material degradation, more phosphorus was produced at 15 DAF.

*G. Changes of total available potassium concentration*

Unlike total nitrogen and phosphorous, the total potassium was significantly higher at 30 DAF (6.42±0.02) than at 15 DAF (4.24±0.04). This study revealed that increasing the fermentation time may increase the number of potassium dissolving microbes (mucilaginous) present under organic carbon.

This was verified by Amanillah, (2011) that once OLF has undergone a lengthy fermentation process, the potassium production in OLF enhances the metabolism of the microbes, leading to further increases in the amount of potassium in OLF.

Table 03 : Time course changes of total nitrogen phosphorous and potassium contents in the novel organic liquid fertilizer

Original fertilizer concentration	Liquid	Total Nitrogen (mg/L)		Total phosphorous (mg/L)	Total potassium (mg/L)		
		15DAF	30DAF		15DAF	30DAF	
Original fertilizer (1000ppm)		513.4±0.12 <sup>b</sup>	329.1±0.3 <sup>a</sup>	588.19±0. <sup>b</sup>	64.53±0.08 <sup>b</sup>	4.24±0.01 <sup>b</sup>	6.42±0.003 <sup>b</sup>
Initial solution		616.3±0.1 <sup>a</sup>	492.06±0.01 <sup>a</sup>	705.6±0.05 <sup>a</sup>	84.06±0.01 <sup>a</sup>	5.05±0.01 <sup>a</sup>	7.69±0.001 <sup>a</sup>
Adding 200ppm		102.2±0.04 <sup>c</sup>	82.1±0.02 <sup>b</sup>	117.3±0.04 <sup>c</sup>	14.1±0.01 <sup>c</sup>	0.41±0.003 <sup>c</sup>	1.3±0.001 <sup>c</sup>

Mean values ± SE. DAF-Days after fermentation Mean values followed by the same letter were not significantly different at the 0.05 probability level based on one-way ANOVAs followed by Turkey post-hock test within the same column, N=6.

IV. CONCLUSIONS

Organic liquid fertilizer showed significantly higher physicochemical values, microbial count and nutrient content (pH, EC, TDS, total N and total P) at 15 days

after fermentation (DAF) than at 30 DAF. However, high total potassium availability (total K) was found at 30 DAF than at 15 DAF. Higher microbial counts were also recorded at 30 DAF, but there was significantly

higher microbial decomposition performance at 15 DAF than at 30 DAF.

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