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Development of a Bench Scale Biodigester for the Production of Bio-fertilizer using Cow Dung and Watermelon Peels

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Abstract
In order to reduce environmental pollution arising from watermelon waste and cow dung, conversion of these wastes into bio-fertilizer is an effective mechanism of waste management. As a result a study was carried out for the development of a bench scale batch anaerobic bio-digester (23 L) for the production of bio-fertilizer. Five kilograms (5 kg) each of cow dung and pre-treated watermelon peels were mixed with water in a ratio of 1:1 w/v to obtained 18 L of slurry. The slurry was then charged into the developed bio-digester and allowed for 35 days at a temperature of 26-31˚C. Bio-fertilizer yield of 83% and biogas yield of 17% were obtained. Plants macronutrients (i.e. NPK) content were substantially increased in the digestates by 79%, 89% and 85% respectively. The presence of Clostridium (a nitrogen fixer bio-fertilizer), Bacillus and Pseudomonas (phosphate solubilizing bio-fertilizers) in the digestate indicated its suitability as bio-fertilizer.
Keywords: Bio-fertilizer; Biodigester, Anaerobic digestion; Retention time; Cow dung; Watermelon peels

1. Introduction

Bio-fertilizers are environmentally friendly fertilizers that not only prevent damages to natural sources but help, to some extent in cleaning the nature from precipitated chemical fertilizers (FAO, 2008). In addition, bio-fertilizers are one of the best modern tools for agriculture and it is a gift of our modern agricultural science. Bio-fertilizers are applied in the agricultural field as a replacement to our chemical fertilizers because they maintain the natural habitat of the soil, provides protection against soil borne diseases and they are cost effective (Owamah *et al*., 2014).

The term 'bio-fertilizer' denotes nutrient supplement inputs for plant growth which are in biological origin. Bio-fertilizers accelerate certain microbial processes in the soil which augment the extent of availability of nutrients in a form easily assimilated by plants and also mobilizing nutritive elements from non- usable form to usable form through biological processes. The role of bio-fertilizers in agricultural production assumes special significance, particularly in the present context of expensive chemical fertilizers. Moreover, it provides the farmers with a new strategy which is helpful for achieving the targeted goal of food security in Nigeria by increasing high productivity yield of food grains (FAO, 2012).

The most popular way of producing bio-fertilizer is through anaerobic digestion. Anaerobic digestion involves the controlled degradation of organic waste in the absence of oxygen and in the presence of anaerobic microorganisms (Alfa *et al*., 2013). This process generates a product called biogas that is primarily composed of methane, carbon dioxide and digestate bio-fertilizer suitable as soil conditioners (Owamah *et al*., 2014). However, the biochemical reaction involved in the digestion process is shown in Equation (1);

$$Biomass \xrightarrow{Anaerobic\ digestion} CH_4 + CO_2 + H_2 + NH_3 + Digestate \quad (1)$$

Furthermore, the advantage of using anaerobic digestion in an urban areas is to treat organic waste as opposed to composting, anaerobic digestion produces biogas with a high percentage of methane which can be used as fuel whereas composting produces mostly carbon dioxide which could not be used as fuel and the demand for bio-fertilizer is dependent on compliance with quality standards (Alfa *et al*., 2013).

The use of digestate bio-fertilizer to increase agricultural food production and soil improvement has been established by previous researchers, but its safety as determined by the amount of pathogens contained is still of public health concern to end users (Owamah *et al*., 2014) and reports on the design of a bench scale biodigester for the production of bio-fertilizer using cow dung and food wastes are scanty in literature, despite the large volume of literature available on biogas production from various substrates (Alfa *et al*., 2013). Moreover, researches on biogas and bio-fertilizers in Nigeria have focused on the utilization of animal dung, human excreta, chicken droppings, and kitchen wastes as substrates while the use of plant wastes such as peels have been limited to water hyacinth, cassava peels, *cymbopogon citrus* and peels of water lettuce (Alfa *et al*., 2013). Watermelon (*Citrus latanus*) originated from western Kalahari region of Namibia and Bostwana in Africa is now found in most tropical countries and Nigeria is one of the world's largest producers with over 347,000 metric tons in the year 2002 (Schippers, 2002).

Survey carried out in Bauchi Metropolis – Nigeria indicates that one in every twenty watermelon in the fruit market is lost to microbial attacked which translate to 5%. In addition, the peels are disposed after consumption although, it is used as animal feed but its availability and sustainability is assured.

The objective of this study is to develop a bench scale bio-digester for the production of bio-fertilizer and to assess the quality parameters, yields of the digestate and biogas obtained from the co-digestion of cow dung and watermelon peels. The development of the bench scale bio-digester will open way for the scale-up to a commercial scale of production which has several advantages (e.g. industrialization, creation of jobs, revenue generation etc.).

2. MATERIALS AND METHODS

2.1 Biodigester design

A bench scale biodigester capable of handling 20 L of feedstock for the production of bio-fertilizer was designed by adopting Ajoy karki's biogas model (Karki, 2002) with some modifications. For instance, separate floating gas holder system consisting of water jacket with inverted measuring cylinder was incorporated into the design for ease of measuring biogas volume. Biodigester volume was based on the substrate input quantity and chosen retention time. Cylindrical shape was adopted for better mixing, stainless steel was chosen as material for construction due to the nature of raw material (highly corrosive to metals).

2.2 Biodigester design consideration

The operating volume of the biodigester (V_o) was determined based on the substrates input quantity (S_d) and the chosen retention time (RT) Equation (2) (Omprakash & Yasabie, 2013).

$$V_O = S_d \times RT\ [=]\ m^3 \quad (2)$$

The retention time is the interval of time during which the biomass remains to decompose in the digester. Normally, the retention time for anaerobic digestion of food wastes and cow dung at mesophilic temperature is between 20-40 days (Omprakash & Yasabie, 2013) and thus 30 days was adopted in the design of biodigester.

Substrate input is given as:

$$S_d = Biomass\ (B) + Water\ (W)\ [=]\ m^3/day \quad (3)$$

Usually the total volume of the biogas is always higher than the operating volume. Therefore, operating volume of 80% of the total volume was chosen (Omprakash & Yasabie, 2013). This is to give room for the expansion in volume of the slurry during fermentation. Therefore, the total volume is given as:

$$V_T = \frac{V_0}{0.8}\ [=]\ m^3 \quad (4)$$

In addition, the bio-fertilizer yield was given by Equation (5):

$$\text{Biofertilizer yield} = \frac{\text{mass of the digestates biofertilizer produced}}{\text{mass input of the substrate}} \times 100\% \quad (5)$$

2.3 Fabrication of biodigester

An airtight batch anaerobic bio-digester was fabricated using the appropriate machines and hand tools at the Mechanical Engineering workshop of the Technology Incubation Centre (TIC) Bauchi, Nigeria. The theory behind was simply downward delivery of biogas and upward displacement of water. The biodigester consists of digestion chamber, inlet from the top cover, digestates outlet pipe, sampling point and a stirrer as shown in the isometric drawing in Plate I. The fabrication was carried out by stick compliance with the design parameters calculated.

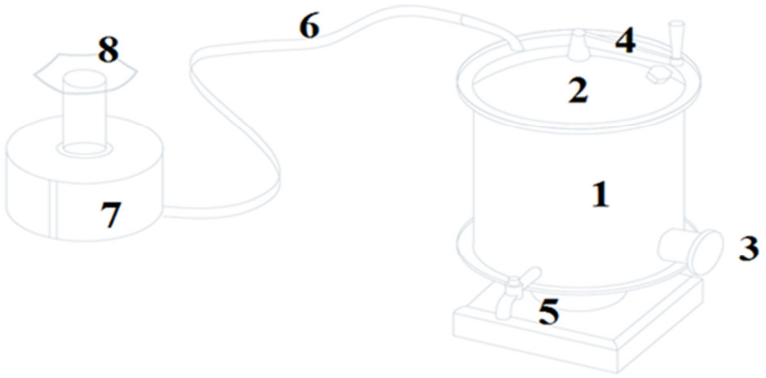

Plate I: Isometric projection of the bench scale bio-digester (1. Bio-digester body; 2. Top cover; 3. Outlet pipe; 4. Stirrer; 5. Tap head; 6. Rubber hose; 7. Water jacket; 8. Measuring cylinder*)*

2.4 METHODS

2.4.1 Bio-fertilizer Production

Cow dung and watermelon peels were collected in a water proof sack from cattle market (Kasuwan Shanu) and Muda lawal market, Bauchi-Nigeria respectively. The raw materials were pretreated by removal of the unwanted

non-biodegradable materials and watermelon peels washed with distilled water in order to remove impurities. In addition, cow dung was sun dried for 3 days at an average temperature of 28.5℃, watermelon peels were blended using BLG-401-18N kitchen blender. The elemental compositions of cow dung and watermelon peels were determined using DR/890 colorimeter and atomic absorption spectrophotometer.

Five kilograms (5 kg) each of the respective biomass (cow dung and watermelon peels) were measured using digital weighing balance and thoroughly mixed. The resulting mixture (i.e. 10 kg of biomass) was mixed with equal amount of water to obtained the feedstock (1:1 w/v). The dilution of the biomass was to enable bacteria move freely in the bio-digester. The feedstock was homogenized for easy digestion and its physicochemical and microbiological parameters were analyzed before charging into the bio-digester.

Twenty kilograms (20 kg) of the feedstock were fed into the bio-digester through the top opening cover (Plate 1) and closed after charging: airtight condition of the digestion process was ensured. The feedstock occupied 79% of the bio-digester volume leaving a clear space of 21% for expansion/biogas production. The digester and its content was allowed to ferment for 35 days at atmospheric conditions (Omprakash & Yasabie, 2013). The physicochemical and microbiological analyses of feedstock and digestate before and after digestion were carried out respectively, the temperature of the bio-digester was taken three times daily, volume of biogas produced was measured on daily basis, pH of the bio-digester content was taken daily, and weekly collection of samples for physicochemical and microbiological analyses. All analyses were carried out in triplicate.

2.5 Physicochemical and Microbiological Parameters of Feedstock and Digestate

2.5.1 Physicochemical parameters

The physicochemical parameters of the feedstock and digestate were evaluated using standard procedures (APHA, 2012). Parameters analyzed includes biochemical oxygen demand (BOD), chemical oxygen demand (COD), total solids (TS), total suspended solids (TSS), organic carbon, nitrogen content, carbon/nitrogen ratio, P_2O_5 , K_2O and pH respectively.

2.5.2 Microbiological parameters

Microbial counts

Microbial population in the biodigester feedstock and digestate produced were enumerated by standard plate count technique using 0.5 ml aliquots of appropriate dilution pour plated onto nutrient agar, MacConkey agar, Eosin methylene blue agar and fastidious anaerobic agar for bacteria. Potato dextrose agar (PDA) plus chloramphenicol was used for fungal isolation and enumeration. Nutrient agar, MacConkey and Eosin methylene blue agar plates were incubated at 37℃ for 48 h, Potato dextrose agar plates were incubated at 25℃ for 5 days while fastidious anaerobic agar plates were incubated in an anaerobic jar (oxoid) containing a moistened pack of gas generating kit (Bio-oxoid) at 37℃ for 7 days.

Microbial isolation and identification

Purification and identification of individual colonies were carried out by morphological and biochemical tests while for fungi isolates, the microscopic and macroscopic features of the hyphal mass, morphology of cells and spores, nature of fruiting bodies were used for identification (Tsuneo, 2010). The colonies that appeared from the isolates were gram-stained, wire loops were sterilized using flame and they were allowed to cool. In addition, the portion of the colony was picked, emulsified and allowed to dry in the glass-slide. A drop of grams iodine and crystal violet were introduced in the slide and allowed to stay for 1 min. The slide was decolorized with 70% alcohol and flooded with water and it was further counter stained with safranin and allowed to stay for 1 min and further washed with water. The gram positive and gram negative organisms were identified by blue crystal and neutral red respectively. More so, the slide was placed in the microscope and observed under x100 objective lens with a drop of oil emulsion for bacterial identification while fungi were identified at x40 objective lens with a drop of lacto phenol cotton blue.

3. RESULTS AND DISCUSSION

3.1 Biodigester Design

Summary of design parameters for the development of 23 L per batch of bio-fertilizer is presented in Table 1.

Table 1: Summary of design and operating parameters of a bench scale bio-digester

Design parameters	Capacity/Description
Digester Configuration	Cylindrical
Basis of bio-digester design	20 kg/batch substrate
Mixing ratio	1:1
Substrate input quantity (S_d)	0.018 (m^3/day)
Operating Volume (V_O)	0.018 (m^3)
Total Volume of the digester (V_T)	0.023 (m^3)
Height to radius ratio ($h_d : r_d$)	3:1
Height of bio-digester (h_d)	0.432 m (43.2 cm)
Radius of the bio-digester (r_d)	0.144 m (14.4 cm)
Digester diameter	0.288 m (28.8 cm)
Material of construction	Stainless steel

The geometry of the bio digester was chosen as cylindrical for ease of operation, maintenance and to promote good mixing of the content. In addition, stainless steel was used as material of construction for fabrication of the bio digester because it is strong to withstand the weight and pressures of the substrates, good tensile strength and durability in an acidic and basic environment. As it is known, some of the components of biogas such as carbon (IV) oxide (CO_2) and hydrogen sulphide (H_2S) are corrosive.

In addition, based on the design parameters calculated as presented in Table 1, a detailed diagram was drawn for the developed bio-digestion unit as shown in Plate II. The fabricated bio digester is as shown in Plate III.

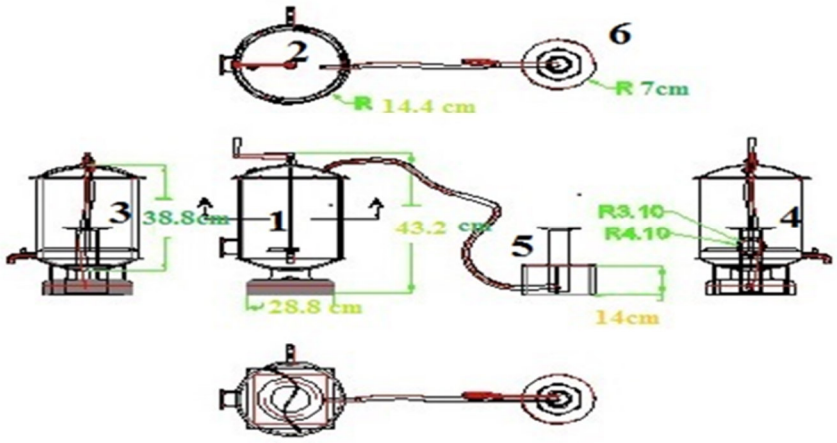

Plate II: Detailed drawing of the developed bio-digester (1. Front elevation of bio-digester; 2. Plan; 3. Left elevation; Right elevation; 5. Front elevation of gas holder; Plan view of gas holder)

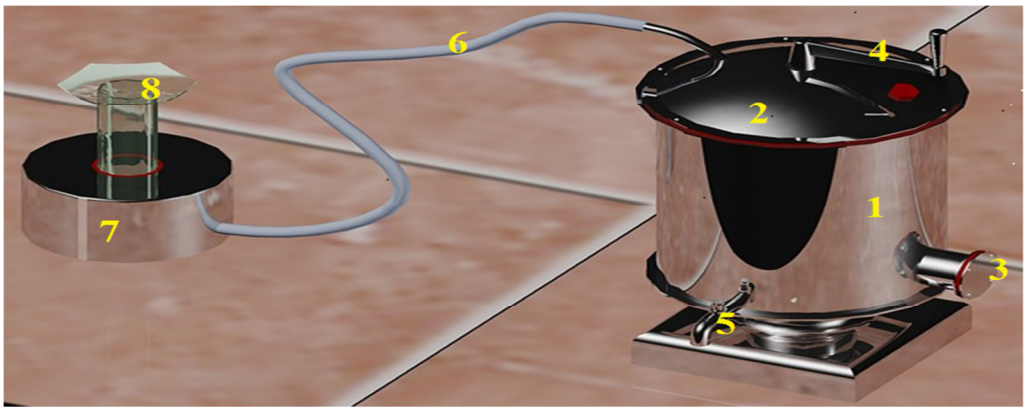

Plate III: Pictorial representation of the developed bench scale biodigester (1. Biodigester body, 2. Top cover, 3. Outlet pipe, 4. Stirrer, 5. Tap head, 6. Rubber hose, 7. Water jacket and 8. Measuring cylinder)

3.2 Bio-fertilizer Production

3.2.1 Analyses of feedstocks

The elemental analyses of cow dung and watermelon peels presented in Table 2 revealed that cow dung had high nitrogen, P_2O_5, K_2O, and micronutrients contents than the watermelon peels. This might be due to high organic

matter present in the cow dung (Alfa *et al*., 2013). In contrast, watermelon peels had higher moisture content than cow dung. This observation can be linked to higher moisture content of watermelon (Dahunsi *et al*., 2015) and this could be an added advantage for microbial activities in the digester.

Table 2: Elemental analyses of cow dung and watermelon peels

Parameter	Cow dung	Watermelon
Nitrogen (mg/L)	9.86 ± 0.13	3.21 ± 0.06
P_2O_5 (mg/L)	1.76 ± 0.01	0.92 ± 0.01
K_2O (mg/L)	1.53 ± 0.83	0.53 ± 0.40
Calcium (mg/L)	0.18 ± 0.01	0.02 ± 0.01
Sodium (mg/L)	2.13 ± 0.04	1.60 ± 0.02
Sodium (mg/L)	2.13 ± 0.04	1.60 ± 0.02
Zn (mg/L)	4.16 ± 0.01	2.10 ± 0.01
pH	6.30 ± 0.01	7.40 ± 0.20
Moisture content (%)	33.41	68.53

3.2.2 Products yield

In this study, the two main products are the digestate or slurry bio-fertilizer and the crude biogas. Results obtained showed that at an average bio-digester temperature of 26.5^{o}C, a bio-fertilizer yield of 83.35% (16.67 kg) digestate and 16.65% biogas were obtained after 35 days of continuous digestion. This agrees with the finding Al-saedi *et al.* (Al saedi *et al*., 2006). The pictorial representation of the digestate is presented in plate IV. This digestate was later cured by drying at ambient temperature for fifteen days at an average temperature of 29.5^{o}C, the product obtained (i.e. the cured digestate) is as shown in Plate V.

Plate IV: Digestate produced from co-digestion of cow dung and watermelon peels

Plate V: Cured digestate produced from co-digestion of cow dung and watermelon peels

The yield of biogas generated was presented on cumulative basis for the retention period (35 days). The

cumulative biogas generation of 592.67 cm^3 per 10 kg of feedstock (i.e. average of 59.3 cm^3/kg) was obtained. This biogas production is less compared with similar study as presented in Table 3. This could be as a result in the difference in their initial carbon to nitrogen ratio, type of feedstock and wider temperature range variation in this study compared with Abdulsalam *et al*. (Abdulsalam *et al*., 2012). In addition, bio-digesters used in (Abdulsalam *et al*., 2012) were painted black to minimise loss of heat to the surroundings. Moreover, the weekly mixing of the content of the biodigester in the present study could affect the activity of the microorganisms and hence the low biogas yield obtained. Although, similar approach was adopted in the collection of biogas by Abdulsalam *et al*. (Abdulsalam *et al*., 2012) and the present study (i.e. the downward displacement) but losses of biogas were inevitable with could probably account for the low generation.

Table 3: Comparison of biogas generation in this study with simular study

Substrate	OPT (°C)	RT (day)	Ave. OP pH	Ave. Biogas Generated (cm^3/kg)	Reference
Cow dung	26 – 28	33	7.6±1	69.07	(Abdulsalam *et al*.,2012)
Elephant dung	26 – 28	33	7.6±1	79.40	(Abdulsalam *et al*.,2012)
Cow & Elephant dung	26 – 28	33	7.6±1	96.07	(Abdulsalam *et al*.,2012)
Cow dung&Watermelon	24 – 30	35	6.5±0	59.30	Present study

OPT: operating temperature; RT: retention time; OP pH: operating pH

Figure 1 presents the variations in biogas volume, operating temperature and ambient temperature with retention time. From this figure, it can be seen that the operating and ambient temperatures have strong correlation; that is, as the operating temperature increase, the ambient temperature increased and vice-versa thus the ambient temperature was relatively higher than the operating temperature. It could also be seen that biogas generation started on the 4th day and increased until it attained a maximum value on the 12th day (50.8 cm^3) which coincided with the period of maximum operating temperature (30.5°C). Thereafter, there was a fall in biogas generation between the 12th and 15th day which resulted due to fall in operating temperature (from 30.5°C to 25°C) by the influence of fall in ambient temperature (from 30.8°C to 26.2°C). As the operating and ambient temperatures increased there was a corresponding increase in biogas generation between the 16th and 20th day. After the 20th day a steady decline in the volume of biogas generated was observed and from the 33rd to 35th day there was no biogas generation.

From the above, it is seen that operating temperature had great influence on the generation of biogas and the ambient temperature greatly influence the operating temperature and hence the volume of biogas generation. It is therefore, recommended that the developed biodigester should be lagged or insulated to prevent or minimize the loss of heat to the environment and heating element be incorporated in order to be able to operate within optimum conditions (FAO, 2008): temperature (30-35°C), pH (6.8-7.5); solid content (7-9%).

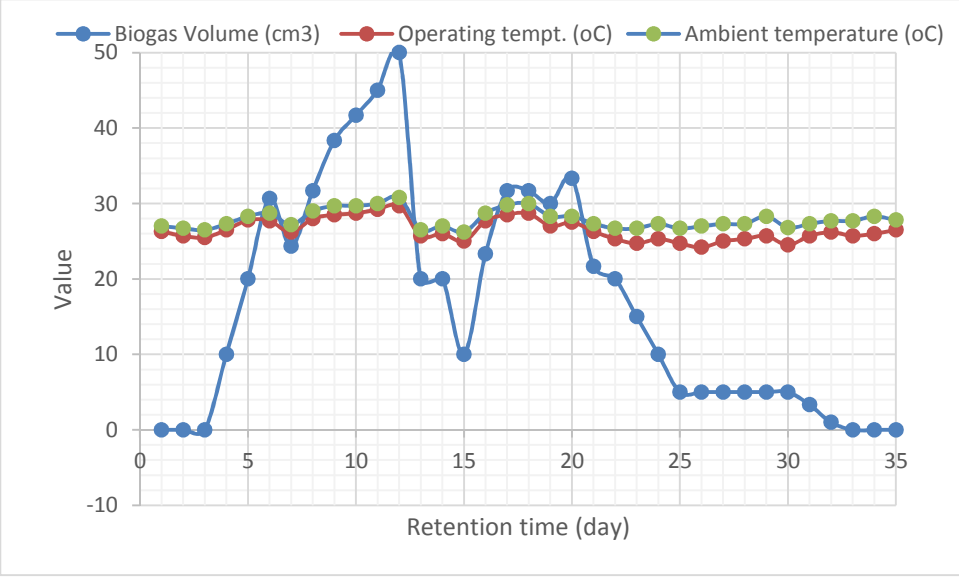

Figure 1: Variation in biogas volume, operating and ambient temperatures with retention time

3.2.3 Physicochemical parameters of digestate

The physicochemical parameters of the bio-fertilizer produced were presented in Table 4 and the weekly variations of these parameters are presented in Figures 2 and 3. In addition, the World Health Organization (WHO, 2006) standard and Nigerian Environmental Standards and Regulations Enforcement Agency (NESREA, 2011) guidelines for discharge of effluent into water bodies or irrigation land as shown in Table 4 were used as bases for comparison of the digestate produced.

From Table 4, it can be seen that the biological oxygen demand (BOD), total solids (TS) and organic carbon (OC) decreased as would be expected (Owamah *et al*., 2014) while the chemical oxygen demand (COD), total suspended solids (TSS), nitrogen (N), P_2O_5, K_2O and pH increased (Owamah *et al*., 2014). The weekly variations in physicochemical parameters (Figures 2 and 3) depict same trend from week to week.

Table 4: Physicochemical and microbiological parameters before and after digestion

Parameter	Before digestion	After digestion	±%	WHO limit	NESREA limit
BOD (mg/L)	88.29± 0.02	0.14± 0.01	99.84	50	30
COD (mg/L)	196.96±0.09	329.24± 0.25	67.16	250-1000	80
Total solids (mg/L)	111.64± 0.98	104.70± 0.27	6.22	500	500
TSS (mg/L)	67.76± 0.98	126.04± 0.03	86.01	-	-
Organic C (mg/L)	590± 0.01	240± 0.00	59.32	-	-
Nitrogen (mg/L)	14± 0.001	25±0.001	78.57	10-30	-
P_2O_5	2.75±0.01	5.2± 0.06	89.09	1-10	-
K_2O	2.6± 0.26	4.8 ± 0.04	84.62	1-10	-
C:N	42.76:1	9.73:1	77.25	-	-
pH	5.8±0.00	7.2± 0.00	24.14	6.5 - 8.5	6.0 - 9.0
Temperature (^oC)	28.5			20 – 32	< 40
Coliform count	$4.84\times10^5\pm0.02$			10^5-10^6	-
Fungal count	$2.30\times10^3\pm0.10$			10^3-10^6	-

Sources: (WHO, 2006; NESREA, 2011)

From Table 4, it could be seen that all the physicochemical parameters for the digestate bio-fertilizer produced met the WHO and NESREA standards except COD that had value above the maximum limit of 80 mg/L stipulated by NESREA but fell within the WHO standard.

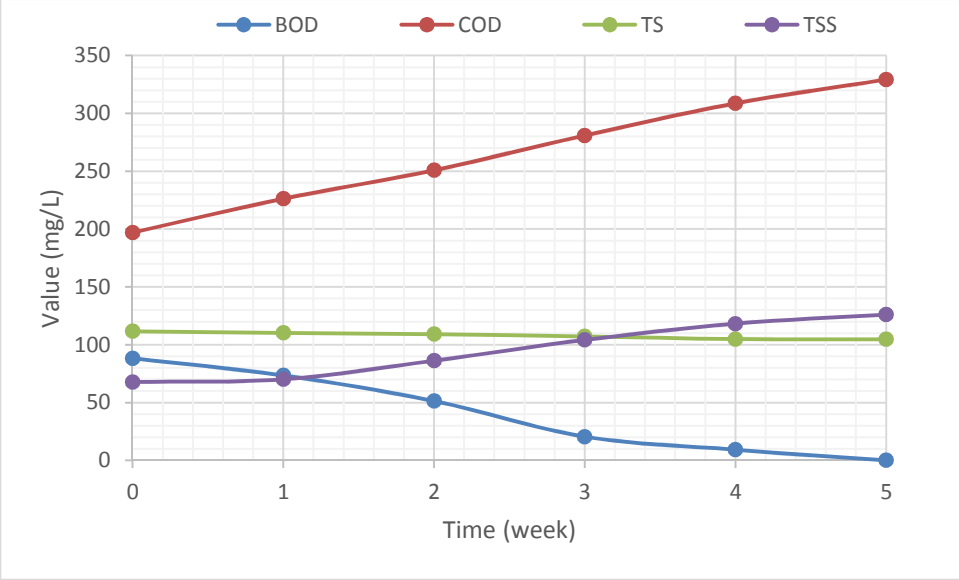

Figure 2:

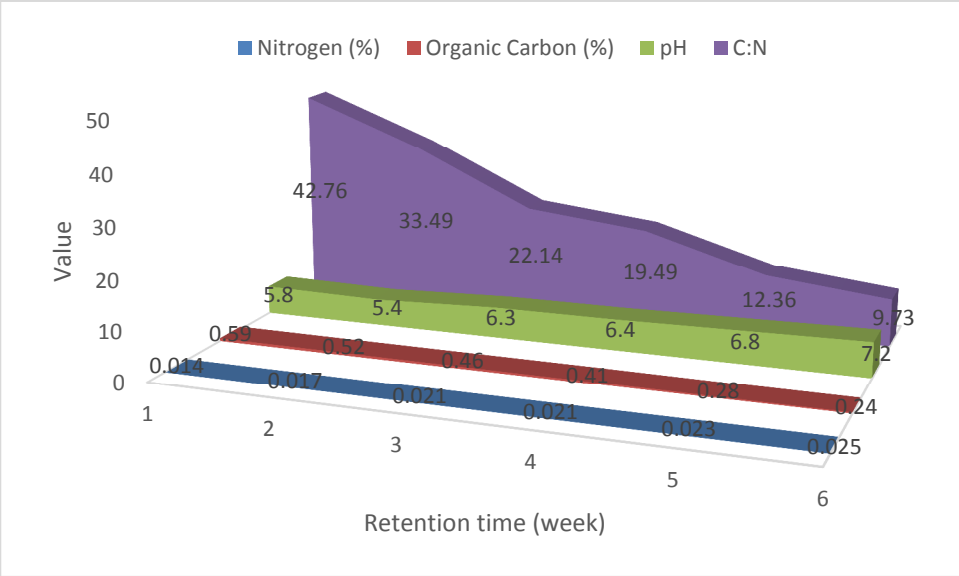

Figure 3:

The study indicates 99.84% reduction in BOD of the digestate with respect to the feedstock and the results revealed that BOD of undigested feedstock was higher than that of the one anaerobically digested (bio-fertilizer). This could be due to the fact that the soluble BOD was readily degradable by healthy microbes in a medium while the insoluble BOD requires longer retention time to degrade (Taylor & Wilkie, 2014). The reduction was higher in the first three weeks (77%) which could be attributed to the rapid deletion in the amount of oxygen needed by microorganisms at the operating temperature and then declined at low rate between weeks 3 and 5.

However, the COD content of the digestate increased by 67.16% when compared with the feedstock. This might be due to increased in the organic content in the digestate as a result of biodegradation of the organic matter in the substrates by the activities of mesophilic micro-organisms at anaerobic condition (Smith *et al*., 2007). Weekly analysis of COD in the biodigester revealed a somewhat linear increase throughout the retention period (Figure 2).

The reduction in total solids by 6.22% and increase in total suspended solids by 86.01% in the digestate when compared to the feedstock. The magnitudes of decrease in TS and increase in TSS are low compared with similar studies (Owamah *et al*., 2014), this could be due to the very low solid content associated with water melon peels. Although, the trend of increase and decrease in TS and TSS were in conformity with similar findings (Alfa *et al*., 2013). Furthermore, the removal of organic carbon in form of CH_4 and CO_2 during the digestion process accounted for the reduction in TS in the digestate.

Weekly analysis of TS showed a linear decrease. On the other hand, TSS increase was slow between weeks “0” and “1” (i.e. 3.57% per week) which could be as a result of low activities of the microorganisms as they get used to the new environment. After acclimatization, microbial activities increased and TSS increased exponentially between weeks “1” and “4” (23.64% per week) and then between weeks “4” and “5” a slower rate of increased in TSS was observed (11.56% per week).

The reduction in carbon/nitrogen ratio was from 42.14:1 (before digestion) to 9.6:1 (after anaerobic digestion) showed that carbon was highly consumed in the digestion process. Furthermore, the C/N ratio of the digestate does not exceed the acceptable limit of 20:1 for biogas and bio-fertilizer production as reported by Owamah *et al.* (2014).

The study revealed 78.57% increase in nitrogen content of the digestate when compared to the bio-digester feedstock and the increased in nitrogen content from 14 mg/L in the feedstock to 25 mg/L in the digestate agrees with similar findings (Monnet, 2003). The increased in nitrogen content after the anaerobic digestion process might be due to high organic matter present in the cow dung as reported by (Smith *et al*., 2007). The P_2O_5 content of the feedstock was 2.75 mg/L and after the digestion, the content increased to 5.2 mg/L which showed 89.09% increase. This increase might be due to the released of organically bound phosphorous in watermelon peels and cow dung during the fermentation process as reported by (Smith *et al*., 2007). Moreover, the study showed 84.62% increase in K_2O content of the digestate when compared with the feedstock. The increase observed in K_2O content from 2.6 mg/L in the feedstock to 4.8 mg/ L in the digestate agrees with similar findings (Al saedi *et al*., 2006, Smith *et al*., 2007).

The order of contribution of macronutrients in the digestate was N > P > K which agrees with similar findings [18] and that of the general purpose fertilizers. The high content of total nitrogen in the digestates might

be due to high nitrogen content of the cow dung and the P > K in the digestate might be due to the less K_2O content in both substrates when compared with P_2O_5.

3.2.4 Elemental analysis of compost bio-fertilizer

The elemental composition of the compost prepared from the digestate was shown in Table 12. The study revealed that the constituents of the cured compost compositions were organic carbon (24.36%), Nitrogen (2.67%), P_2O_5 (2.43%), Sulfate (0.18%), moisture content (18.75%), Nitrate (0.026%), Nitrite (0.053%), Carbon/Nitrogen ratio (9.12:1), and K_2O (2.10%). The study revealed that nitrogen, phosphate, K_2O contents of the resulting compost to be higher than those obtained in previous works (Owamah *et al*., 2014, Schippers, 2002). The N.P.K ratio of the resulting compost corresponds to the specification standard of NPK fertilizer ratio of 1:1:1.

3.2.5 Microbilogical characteristics

The results presented in Table 5 indicates a significant reduction in TAPC of the feedstock from $2.73\times10^8 \pm0.02$ to $1.34\times10^4\pm0.05$ in the digestate. This agrees with (Alfa *et al*., 2013) that TAPC decrease during the digestion process and possibly due to the decrease in carbon/nitrogen ratio which leads to the supply of low nutrients for micro-aerophilic organisms. In addition, the study revealed a significant reduction in total coliform in the digestate as against the value in the feedstock and within the retention period. This agrees with (Owamah *et al*., 2014, Shu-Hsien *et al*., 2007) that microbial population has a tendency to decrease within the period of digestion at mesophilic temperature. Though anaerobic digestates can be used to efficiently improve soil fertility and boost crop production, its safety still remains a source of concern to end users due to pathogens (Alfa *et al*., 2013). The reduction in fungal counts from $4.12\times10^6 \pm0.07$ in the feedstock to $2.30\times10^3 \pm0.10$ in the digestate might be due to the operating temperature (28.5^{o}C) which was above the surviving fungi temperature (21-25^{o}C) in anaerobic digestion.This agrees with similar findings [2, 5]. These colony counts were within the limit specified by both WHO (2006) and NESREA (2011) as shown in Table 4.

Table 5: Microbiological characteristics of the digestate biofertilizer

Period	TAPC (CFU/100 ml)	Coliform count	Fungal count	Species of organisms isolaled
Before	$2.73 \times 10^8 \pm0.02$	$4.73 \times 10^8 \pm0.06$	$4.12 \times 10^6 \pm0.07$	Bacteria *Escherichia coli, Citrobacter, Proteus, Bacillus, Pseudomonas, Entrobacter, Clostridium, Bacteroides, Staphylococcus,* *Streptococcus, Salmonella, Shigella and Klebsiella.* Fungi *Rhizopus, Penicillum, Mucor and Aspergillus.*
After	$1.34 \times 10^4\pm0.05$	$4.84 \times 10^5\pm0.02$	$2.30 \times 10^3 \pm0.10$	Bacteria *Proteus, Bacillus, Bacteroides, Pseudomonas, Clostridium and Salmonella.* Fungi *Penicillum, Aspergillus and Rhizopus*

The biochemical tests revealed several species of bacteria and fungi were found and isolated as presented in Table 5. Species of bacteria and fungi isolated from the digestate includes; *Bacillus, Pseudomonas, Penicillum, Clostridium, Bacteroides, Rhizopus, Aspergillus, Proteus and Salmonella*. *Clostridium* is known to be free-living nitrogen fixing bio-fertilizer (TNAU, 2014) and the digestates would enhance the fertility of soil for crop production. *Bacillus* and *Pseudomonas* are phosphate solubilizing bio-fertilizers and furthermore, *Bacillus* act as solubilizers for trace elements like silicates and zinc as well as plant growth promoters (TNAU, 2008). Species of *Aspergillus* and *Penicillum* are also phosphate solubilizing fungi (Alfa *et al*., 2013).

In the feedstock, four pathogenic microorganisms were detected (*E. Coli, Shigella, Salmonella* and *Klebsiella)* which reduced to only one *(Salmonella)* after digestion, this might be due to the significance decrease in carbon/nitrogen ratio and high increase in plant macronutrient of the bio-digester content because these pathogens could not survive at very low carbon/nitrogen and at high macronutrient contents. Furthermore, the presence of *salmonella* in the digestate calls for concern in its use on plant that can be eaten raw, since it is pathogenic and could be transmitted to man and animals via contaminated food (TNAU, 2008). This findings is an improvement over previous studies as only one pathogenic microorganism was left in the digestate.

3.3 Cost estimate of the developed bio-digester

The detailed cost estimates of materials and cost of labour for the fabrication of the bench scale anaerobic bio-

digester is presented in Table 6. The estimated cost for the developed bench scale anaerobic bio-digester was approximately three hundred and thirty four dollars naira per 23 liter capacity ($334/23L). However, the basis for carrying out the cost estimate is to give a clue for the development of a pilot plant.

Table 6: Cost estimates for the developed bench scale bio-digester

Component	Material	Unit	Quantity	Price ($)
Digester/water jacket body	1 mm stainless steel	m^2	½ sheet	198.00
Stirrer	Ø15mm stainless steel rod	m	1	6.00
Measuring cylinder	Plastic	cm^3	1	9.00
Gas hose	Ø10mm rubber hose	m	2	2.40
Tap valve	1/3 inch stainless steel	-	1	7.20
Gasket	Water seal	-	1	2.40
ABRO silicon sealant	-	-	-	6.00
Bolt & nuts	Size 8, stainless steel	-	10	0.60
Rolling	-	-	-	18.00
Gas welding	Brass	-	-	2.40
Thermometer	Glass	-	-	9.60
Workmanship	-	-	-	36.00
Miscellaneous	-	-	-	6.00
Sub-total	-	-	-	303.60
Add Contingency (10%)	-	-	-	30.36
Total	-	-	-	$333.96

4. CONCLUSION

The study has shown that nitrogen fixer bio-fertilizer could be produced through the anaerobic digestion of cow dung and watermelon peels at mesophilic temperature. The developed bio-digester was effective for the production of bio-fertilizer with yield of 83% (N: 25 mg/L; P_2O_5: 5.2 mg/L and K_2O: 4.8 mg/ L), after curing the macronutrient composition were N (2.67%), P_2O_5 (2.43%) and K_2O (2.10%). *Clostridium*, *Bacillus and Pseudomonas* are important microorganism in bio-fertilizer. *Salmonella* in the digestate is a major health concern, and needs to be addressed in further work. In addition, the approximate cost estimate for the developed bio-digester was $334.

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