Anaerobic co – digestion of pig and poultry droppings with elephant grass for the production of biogas

Okewale, A. O. [1,*] and Babayemi, K. A.[2]

[1]Department of Chemical Engineering, Federal University of Petroleum Resources, Effurun P.M.B 1221, Effurun, Delta State, Nigeria.
[2]Chukwuemeka Odumegwu Ojukwu University, Uli, Chemical Engineering Department
Effurun, P.M.B 02 Uli, Anambra State, Nigeria
***Corresponding author's email:** okewale.akindele@fupre.edu.ng
*Received: 20/01/2018, Accepted: 05/04/201*8

Abstract

Co – digestion of elephant grass, poultry and pig droppings anaerobically for laboratory scale production of biogas was under taken. The pH and temperature ranges for this study were 5.5 – 7.1 and 25^{o}C – 30^{o}C respectively within the hydraulic retention time of 52 days. 9.10% total solid concentration was used in each of the digesters. The water displacement method was used to estimate the biogas produced. The percentage weight ratio distribution of poultry droppings to pig droppings were; (100:0), (75:25), (50:50), (25:75), and (0:100) for digesters A, B, C, D and E respectively. Digester B gave the maximum biogas yields of 301 $cm^3CH4/ g – VS_{added}$ at the end of 52 days of fermentation after which there was no further production. It is suggested that the presence of polycyclic aromatic hydrocarbon, alkanes, SP^3 and methyl functional group in all these substrates used as shown by the Fourier transform infrared spectroscopy carried out make these materials be good for biogas production. The GC analysis on the biogas produced in digester B had maximum production showed 69.43 %v/v and 23.22 %v/v for methane (CH_4) and carbon dioxide (CO_2) respectively. The experimental data fitted well with the linear kinetic model which indicated that there was an increase in the yield of biogas as the retention time increases. The net performance of the digesters were, digester B > digester C > digester A > digester D > digester E. X – RF analysis carried out on the substrates showed that poultry dropping has more Fe_2O_3, CaO, P_2O_5, K_2O, and Mn_2O_3 essential elements required for enzymes and microbial metabolisms in anaerobic digestion. This makes poultry droppings a very viable substrate for biogas production compared to the other two substrates. The overall power generations were 6.54, 9.57, 7.8, 5.4, and 1.89 watt in digesters A, B, C, D, and E respectively.

Keywords:
Biomethanisation, fermentation, power, energy production, kinetics, digester methane, biogas

1. Introduction

One of the most important factors to global prosperity is energy of which its importance cannot be over emphasized, ranging from domestic purposes (heat energy for cooking food and heating water), industrial use (for heating furnaces and running electric motors) and transport purposes. It is also important because it is the cornerstone of economic and social development (El-saeidy, 2004). The need for exploring and exploiting new sources of energy which are renewable, sustainable as well as eco-friendly is inevitable in today's energy demanding lifestyle of the world. Waste from animals such as; poultry droppings, cow dung, and swine (pig) droppings usually constitutes environmental problems for the people living around the area where such wastes are dumped due to obnoxious odours generated from these wastes. These animal wastes have been found to consist of exploitable gas and energy which can be obtained by a process called bio-menthanisation and the gas produced can be used as a source of energy or if burnt directly could be used for heat effect (Dupont and

Accorsi, 2006). The most promising form of renewable energy source is the use of biomass. Biomass sources for renewable energy have a great potential for meeting the future energy demands. In the present moment biogas energy has proven to be a reliable, easily available and economically feasible source of alternative and renewable source which can be managed by using locally available resources and simple technology for rural villages (Mshadete and Parawira, 2009). Anaerobic digestion is the controlled degradation of organic waste in the absence of oxygen and in the presence of anaerobic micro-organisms (Ojolo et al., 2007). The anaerobic digestion process is characterized by a series of biochemical transformations brought about by microbial consortia which convert complex macromolecules into low molecular weight compounds; biomethane, carbon dioxide, water, and ammonia (Mudhoo and Kumar, 2013).

The process of anaerobic digestion usually takes place in the following four stages viz; hydrolysis, acidogenesis, acetogenesis, and methanogenesis. In the first stage of hydrolysis or liquefaction, fermentative bacteria convert the insoluble complex organic matter into simpler soluble molecules. The complex polymeric matter is hydrolysed to monomers, e.g., cellulose to sugars or alcohols and proteins to peptides or amino acids, by hydrolytic enzymes, (lipases, proteases, cellulases, amylases, etc.) secreted by microbes (Karki et al., 2005). Acidogenesis is the acid-forming phase, this is the process whereby acidogenic bacteria turn the products of hydrolysis into simple organic compounds, mostly short chain (volatile) acids (e.g., propionic, formic, lactic, butyric, or succinic acids), ketones (e.g., ethanol, methanol, glycerol, and acetone) and alcohols (Ostream, 2004). Acetogenesis occurs through carbohydrate fermentation, in which acetate is the main product, and other metabolic processes (Themelis and Verma, 2004). The methanogenesis stage produces biomethane resulting from the action of methanogenic bacteria from hydrogen, carbon dioxide, acetic acid, and other resulting substrates in which methanol and formic acid constitute the most significant parts in the methanogenesis stage. The simultaneous digestion of more than one type of wastes in the same unit is referred to as co – digestion. Advantages of co – digestion include better digestibility, enhanced biogas production / methane yield arising from the availability of additional nutrients, as well as a more efficient utilization of equipment and cost sharing (Parawira et al., 2004; Mshandete et al., 2009). Results of co – digestion of food waste and dairy manure in a two – phase digestion system conducted at laboratory scale showed that the biogas production rate of co – digestion was enhanced by 0.8 – 5.5 times as compared to the digestion with dairy manure alone (El – Mashad et al., 2007). This study focused on anaerobic co - digestion of pig and poultry droppings with elephant grass for the production of biogas. The work was aimed at finding the proportion of poultry and pig droppings that will be co – digested by keeping the elephant grass weight constant to achieve a maximum biogas (CH_4) production. Furthermore, the kinetic parameters from the linear mathematical model concerning the rate of biogas production for the bio-digester in batch mode of operation were studied.

2. Materials and Methods

2.1 Materials

The elephant grass was obtained from the Ugbomro community, Effurun and Delta State while pig and poultry droppings were procured from piggeries and poultry farms respectively in Okuokoko, Effurun, and Delta State. Conical flasks (500 ml), mercury in glass thermometer (range between -10 ^oC – 100 ^oC, with an accuracy of ±0.1 ^oC), digital pH meter (HANNA model pH – 211), delivery tubes, corks, measuring cylinders (200 mL), muffle furnace, Oven (Genlab oven model, Mino/75/f), connecting tubes, mortar and pestle, weighing balance (model BH 600) with an accuracy of 0.01 g, sodium chloride (NaCl), tetra oxo sulphate (VI) acid (H_2SO_4), Buckner flasks (500 ml), and distilled water which was procured from the Department of Chemistry Laboratory, Federal University of Petroleum Resources, Effurun were used for the biogas production.

2.2 Methods

2.2.1 Pre-treatment of sample and sample characterization

The elephant grass was shredded, poultry and pig droppings were sun dried for two days and thereafter dried in the oven at 110°C for a period of 6 hours. The particle size range of 0.800mm was used for the biogas study.

2.2.2 Fourier Transform Infrared Spectroscopy

Elephant grass, poultry, and pig droppings of 0.8mm particle size were observed with FTIR spectroscopy (Buck Scientific model 530) with the range 650 - 4000 cm^{-1} (wavelength).

2.2.3 X – Ray Fluorescence Analysis

The elemental and chemical analyses of the substrates were investigated to identify the elements present in it. The substrates were examined using a Philip (PW1606) X-ray fluorescence spectrometer model.

2.2.4 Gas Chromatography (GC) Analysis

The biogas produced was analysed using an Agilent GC analyzer model (7890) equipped with TCD. The operating pressure is 109.5 kPa at a flowrate of 25 mL/min, temperature of the injector is 130ºC, and 140ºC was the detector temperature while 120ºC is the temperature of the column used. Helium was the carrier gas used for the analysis.

2.2.5 Determination of pH

5g of the sample slurry was poured into a beaker. The slurry was agitated and left for 24 hours to stand at room temperature. The pH of the slurry was then measured using the pH meter (HANNA model pH – 211) (ASTM, 1996).

2.2.6 Determination of moisture content

The moisture content was determined using standard test ASTMD 2867 – 91 (ASTM, 1991).
5 g each of the pre-treated sample was weighed in a petri dish (initial weight) which was then placed in an oven at 110 ºC (Genlab oven, model Mino/75/f). The weight was taken after every 10 minutes until a constant weight was obtained (final weight). The moisture content was subsequently determined by using equation 1 below;

$$\%\text{Moisture content} = \frac{\text{Initial weight} - \text{final weight}}{\text{Initial weight}} \times 100 \quad (1)$$

2.2.7 Determination of volatile matter

5 g of each of the samples were weighed and placed in a crucible (initial weight), transferred to a muffle furnace that has been pre – heated to 600 ºC for 4 hours. The samples were moved to a desiccator and re – weighed again. The weight lost is now the volatile matter present in the samples calculated using equation (2) below;

$$\%\text{Volatile Matter} = \frac{\text{Initial weight of wet sample+Crucible} - \text{final weight after heating+Crucible}}{\text{final weight after heating+Crucible} - \text{Initial weight of the crucible}} \times 100 \quad (2)$$

2.2.8 Determination of the ash content

5 g of each sample was weighed into a porcelain crucible and placed in a furnace that was preheated to 600 ^{0}C for 2 hours. Thereafter, the crucible was transferred to the desiccator to cool. The final weight was measured after cooling. Ash content was determined by using equation (3);

$$Ash\ content\ (\%) = \frac{final\ weight}{initial\ weight} \times 100 \quad (3)$$

2.2.9 Determination of Energy produced

The energy production was estimated using the method of Reungsang et al., (2012). Energy production from CH_4 (kJ/g – VS_{added}) is estimated by multiplying the relative density of methane (0.72mg CH_4/cm^3 – CH_4) and the heating values of methane (55.6J/mg – CH_4) with the methane yield (cm^3CH4/g – VS) (Sittijunda, 2015).

2.2.10 Determination of Carbon and Nitrogen

Carbon and nitrogen present in the substrates were estimated using the standard method of APHA, (1995).

2.2.11 Estimation of power generation

The power generation from anaerobic co – digestion of elephant grass, pig and poultry droppings was calculated using equation (4) below;

$$Power\ (watt) = \frac{Overall\ energy\ production\ (J)}{Overall\ time\ of\ fermentation\ (hr)} \quad (4)$$

2.2.12 Experimental procedure

The apparatus used were properly washed with a soap solution, distilled water, and allowed to dry overnight in the laboratory. Buckner flasks (500mL) were used as digesters for each of the sample. Another set of Buckner flasks (500mL) which contained an acidified brine water solution, was connected to each of the digester by means of a connecting tube and also, on the other side, connected to a conical flask by means of a connecting tube. Thus, the biogas produced in the digester by the fermented slurry (samples) passed through the connecting tube to the Buckner flask containing an acidified brine solution. The acidified brine water solution was displaced on the other side of the conical flask by the pressure of the biogas produced. The amount of water displaced was then measured as the volume of biogas produced. The digester was operated at ambient temperatures. The total solid concentration of 9.10% was used in each of the digesters.

Digester A which consists of elephant grass, poultry droppings and water were mixed together by mass ratio 10g: 15g: 250g (1:1.5:0) respectively. Digester B which consists of elephant grass, poultry and pig droppings, and water were mixed together by mass ratio 10g: 11.25g: 3.75g: 250g respectively. Digester C consists of elephant grass, poultry and pig droppings, and water were mixed together by mass ratio 10g: 7.5g: 7.5g: 250g respectively. Digester D was made up of elephant grass, poultry and pig droppings, and water were mixed together by mass ratio 10g: 3.75g: 11.25g: 250g respectively. Digester E consists of elephant grass, pig droppings, and water were mixed together by mass ratio 10g: 15g: 250g respectively. The ratio of the percentage distribution of poultry droppings to pig droppings were (100 wt. %:0 wt. %), (75 wt. %:25 wt. %), (50 wt. %:50 wt. %), (25 wt. %:75 wt. %), and (0 wt. %:100 wt. %) for digesters A, B, C, D and E respectively. The experimental set – up is shown in fig. 1.

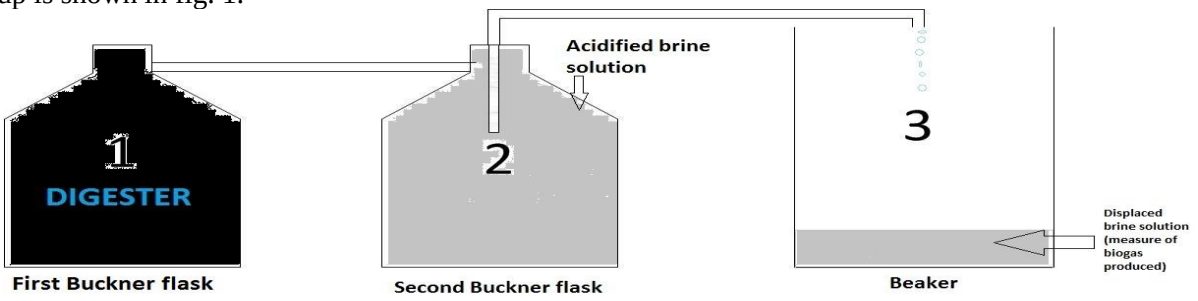

Fig. 1 Experimental Set – up for biogas Production.

2.2.13 Kinetic model for biogas production

Biogas production rate from co – digestion of elephant grass, poultry and pig droppings, was simulated using linear plot. The ascending and descending limbs could be expressed in this model (Kumar et al., 2004) reported that ascending and descending limbs of biogas production rate can be expressed by the linear equation (5) below;

$$y = a + bt \quad (5)$$

Where y is biogas production rate in ml/gm/day, t is time in days for the digestion, a (ml/gm/day) and b (ml/gm/day) were constant obtained from the intercept and slope of the plot of y against t in ml/gm/day.

3. Results and Discussion

3.1 Characterization of the substrates

Table 1 Characterization of elephant grass, pig and poultry droppings.

Parameters	Elephant Grass	Pig Dropping	Poultry Dropping
pH (initial – final)	5.72 – 6.94	5.8 – 6.8	5.5 - 7.1
Particle size (mm)	0.800	0.800	0.800
Carbon Content (%)	15.5	14.3	27.25
Nitrogen Content (%)	0.62	0.65	2.19
C:N	25	22	12.44
Moisture content (%)	0.2	1.0	7.30
Ash Content (%)	5.3	22.80	35.55
Volatile Solid (%)	94.7	67.95	64.45

As seen in Table 1, poultry dropping had the least volatile solid in comparison with the other substrates. It was reported by El – Mashad and Zhang, (2010), that biogas production increases with a decrease in volatile solids. They opined that methanogenic consortium microorganisms acclimatized very well and this leads to easy digestion of volatile solid in anaerobic condition. The lower C/N ratio of poultry droppings makes the poultry droppings produce more biogas compared to pig droppings. This also corroborates the assertion made by Adelekan and Bamgboye, (2009), those substrates with very high C/N ratio would produce very low biogas. The pH range of 7.00 – 8.00 was suitable for obtaining high biogas production and degradation of Volatile solids (Zhai et al., 2015). It has been reported that methanogenic bacteria perform well within a pH range of 6.80 – 7.20 while a drop in pH below 6.60 might inhibit methanogens (Chandra et al., 2012). This study shows that the final pH range of 6.94 – 7.1 is appropriate for methanogens.

Table 2 Ratio of Substrates used for the biogas production.

Digesters	Elephant Grass	Poultry dropping	Pig Dropping	Cumulative biogas yield (cm^3 $CH4/g – VS_{added}$)
A	1	-	1.5	204
B	1	1.125	0.375	301
C	1	0.75	0.75	243
D	1	0.35	1.125	169
E	1	-	1.5	59

As seen in Table 2, both poultry and pig droppings were varied in order to determine their effects on biogas (CH_4) production. It can be observed that maximum biogas yields of 301 $cm^3CH4/ g – VS_{added}$ was obtained at a mixing ratio of 1:1.125:0.375 which implied that a suitable C/N ratio was provided, both microbial growth and substrate utilization were improved thus enhanced high methane yield (Yokoi et al., 2001; Prapinnagsorn et al., 2017). A decrease in the ratio of poultry dropping resulted in low biogas yield which had adverse effects on CH_4 production (Parkin and Owen, 1986).

The low biogas yield might be explained by the imbalance between hydrolytic, fermentative, and acetogenic bacteria, and methanogenic archea (Brown and Li, 2013; Prapinnagsorn et al., 2017). These imbalances might be caused by an unsuitable low pH, substrate ratio, high total ammonia – nitrogen, free ammonia content, and accumulation of organic acids (Li et al., 2013). Digester C had the second highest methane yield at a mixing ratio of 1:0.75:0.75. This shows that the microorganisms supplied by these manures were very close to the requisite C/N ratio for the methanogenesis phase. The low methane yield observed in the mixing ratio 1:1.5 for digester E can be explained as a result of the high amount of nitrogen content present in pig dropping during the fermentation process. It means that a low amount of carbon content inhibited the methanogen leading to accumulation of free ammonia concentration from the pig dropping. This free ammonia concentration can be attributed to the differences in acclimation period, and environmental conditions of the microorganisms. This low methane yield in digester E can also be due to the structure of the elephant grass with more complex lignin, hemicellulose and crystalline cellulose. The accession of microorganisms present in the fermentation system for the grass was more difficult to digest (Prapinnagsorn et al., 2017). A mixing ratio of 1:1:0.375 is noted to be the most suitable mixing ratio due to the highest methane yield obtained.

3.2 Elemental composition of substrates

Tables 3 – 5 depict the elemental compositions of the substrate used in the fermentation process. A trace level of these elements is required for the activation and /or functioning of many enzymes and co – enzymes during anaerobic digestion (Mudhoo and Kumar, 2013; Bayer et al., 2007; Cirne et al., 2007; Mata – Alvarez et al., 2000). These elements form part of the enzymes that are essential in driving anaerobic fermentation reactions. Iron has been reported to be essential for the growth of almost all microorganisms. The basic physiological function of iron is a cofactor for some proteins, most of which are related to energy metabolism (Mudhoo and Kumar, 2013). The nutrient requirement is a major concern for the stable operation of the methane fermentation process (Mathew et al., 2014). The growth of methanogens is dependent on many ions such as sodium, nickel, cobalt, iron, zinc, magnesium, calcium and potassium cations and molybdate or tungstate and phosphate anions (Ramansu et al., 2016). With the exception of sodium which is required for coupling methanogenesis with ADP phosphorylation, all the other ions are required for the synthesis of enzymes, prosthetic groups, and coenzymes (Kaster et al., 2011; Hattori et al., 2009). The presence of an iron element in all the substrates used suggest these materials to be good for biogas production since iron is required in methanogenesis by almost every metalloenzyme involved in the methanogenesis pathway (Rao et al., 2011). The magnesium pathway uses the synthetase and kinase enzymes complexes of ATP and ADP with Mg^{2+} as substrates and products, this (Mg^{2+}) is predicted to be taken up by the MgtE system (Rao et al., 2011). It has been reported that methane formation in cell suspensions of microorganisms is simulated by the gradient of Ca^{2+} ions which is driven by membrane – associated Ca^{2+} ATPase (Kaster et al., 2011). (Ca^{2+}) calcium ions are required for the synthesis of the enzyme Mch and a membrane bound Ca^{2+} ATPase (Zhang et al., 2008; Qiang et al., 2012). The majority of the methanogenic enzymes function optimally only at a high concentration of K^{+} ions (Ramansu et al., 2016). Ramansu et al., (2016), reported that potassium ions are not directly involved in methanogenesis from CO_2 and H_2O. The presence of the potassium ions in these substrates suggests that methanogenic bacteria will be able to withstand various environmental stresses they may be subjected to. The preponderance of these essential elements in poultry droppings makes them a viable substrate for biogas production compared to the other substrates. This is even evidenced in Digester B that has 75% poultry droppings with 25% pig droppings producing maximum biogas during the 52 day retention period while digester E that has 100% pig droppings had the least yield in biogas.

Table 3 Elemental Compositions of elephant grass.

Element	Na_2O	MgO	Al_2O_3	SiO_2	P_2O_5	K_2O	CaO	TiO_2	Fe_2O_3	Mn_2O_3
Concentrations (wt. %)	0.00	1.001	4.151	40.947	5.675	23.138	11.664	0.254	1.602	0.441

Table 4 Elemental Compositions of pig dropping

Element	Na_2O	MgO	Al_2O_3	SiO_2	P_2O_5	K_2O	CaO	TiO_2	Fe_2O_3	Mn_2O_3
Concentrations (wt. %)	0.00	0.904	6.936	53.159	7.396	2.353	10.118	1.343	5.603	0.118

Table 5 Elemental Compositions of poultry dropping

Element	Na_2O	MgO	Al_2O_3	SiO_2	P_2O_5	K_2O	CaO	TiO_2	Fe_2O_3	Mn_2O_3
Concentrations (wt. %)	1.684	3.391	2.766	11.930	16.913	11.533	40.414	0.231	2.126	0.583

3.3 Fourier transform infrared spectroscopy (FTIR) interpretation

From fig. 2, the broad band with frequency (3283.8cm^{-1}) exhibited RO – H (Alcohol) wide rounded band showing the presence of alcohol. The broadband with frequency (2922.2cm^{-1}) shows the presence of the functional group; SP^3 (saturated hydrocarbon), (C – H) methyl ($-CH_3$). Alkenyl (C = C) stretch was seen at frequency of broadband (1640.0cm^{-1}). The broad band with frequency (1423.8cm^{-1}) indicates the presence of vinyl C-H plane bend. The functional group (O – H) phenol or tertiary alcohol OH band was exhibited at frequency (1364.2 – 1319.5cm^{-1}). The broad band (1244.9cm^{-1}) represents the aromatic ether aryl –O while broad band (1155.5cm^{-1}) represents SP^2, C-O group. The broadband with frequency (1028.7cm^{-1}) suggests the presence of primary amine, CN stretch. Vinyl C=H out of plane band was revealed at the broad band with frequency (898.3cm^{-1}) (Coates, 2000). The presence of the SP^3 (saturated) functional group in the elephant grass suggests it to be a good material for biogas production.

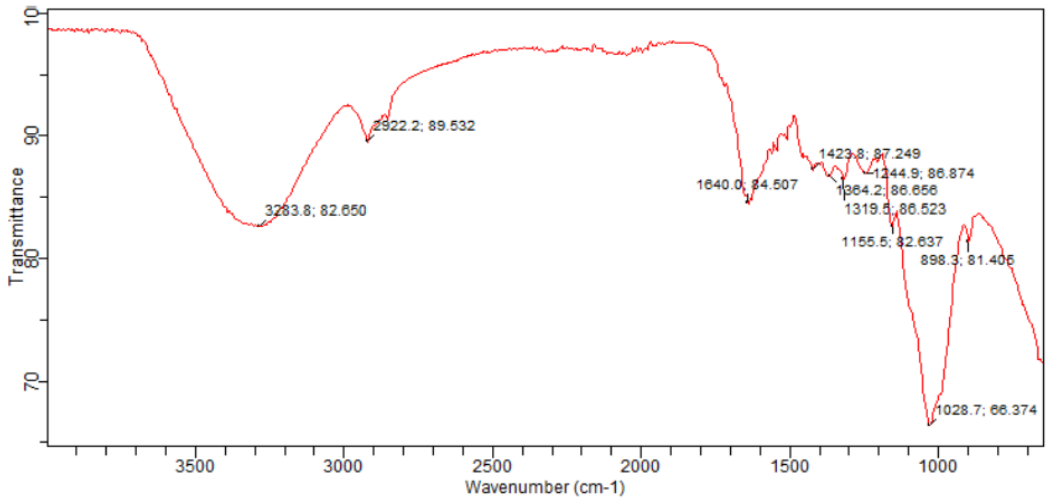

Fig. 2 FTIR spectra for elephant grass.

As seen in fig. 3, the broadband (2851.4 - 2922.2cm^{-1}) exhibited the major functional group present; SP^3, (C – H) tallest C – H band. The broad band with frequency (3291.2cm^{-1}) showed the presence of RO – H (Alcohol) wide rounded band showing the presence of alcohol. (C = C) alkenyl stretch was noticed at frequency (1636.3cm^{-1}). The broadband frequency of 1461.1cm^{-1} revealed the presence of methylene C = H bend. The broadband frequency of 1036.2cm^{-1} suggests the presence of

primary amine, CN stretch. The presence of SP^3 (saturated functional group), C-H group and methyl C-H group as the groups with the highest transmittance indicates that pig droppings are a viable substrate for biogas production.

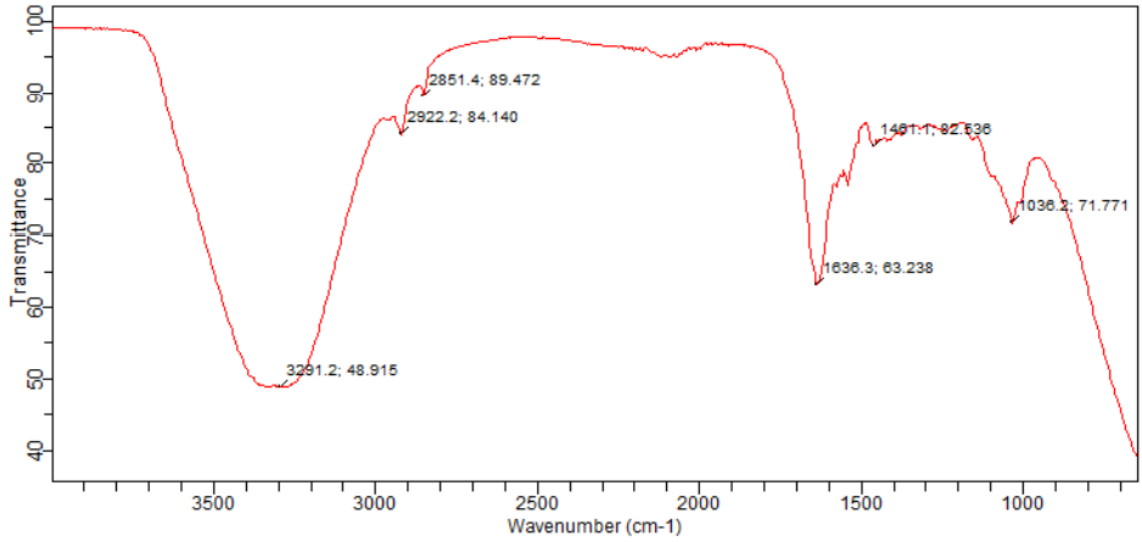

Fig. 3 FTIR spectra for pig dropping.

As shown in fig. 4, the frequency (1028.7cm^{-1}) suggests the presence of primary amine, CN stretch. The broad band with frequency (3280.1cm^{-1}) exhibited RO – H (Alcohol) wide rounded band showing the presence of alcohol while the broadband (1636.3cm^{-1}) exhibited (C = C) alkenyl stretch. The main functional group (O – H) phenol or tertiary alcohol OH band was seen at broadband (1408.9 – 1319.5cm^{-1}). The presence of alkene and phenol makes poultry droppings a very good substrates for biogas generation.

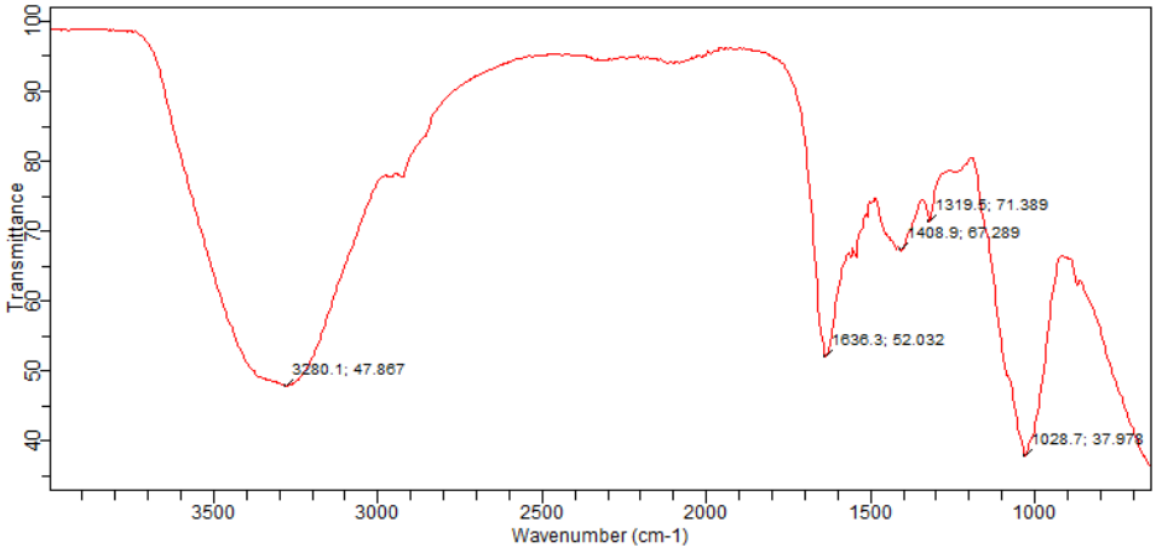

Fig. 4 FTIR spectra for poultry dropping.

3.4 Methane production and pathway

3.4.1 Methane production

Figure 5 shows the cumulative biogas produced by digesters A, B, C, D, and E respectively for 52 days of fermentation. Digester B (75 wt. % poultry, and 25 wt % pig droppings) was observed to generate the highest quantity of biogas during this period of retention time.

There was no production in all of the five digesters for the first four days of fermentation. This period of no activity can be explained to be due to the metamorphic growth process of the methanogens by consuming methane precursors produced during the early activity as suggested by (Li et al., 2011; Lalitha et al., 1994; Bal and Dhaghat, 2001). The initial stages of the overall biogas production process, acid forming bacteria produce Volatile Fatty Acids (VFA) thereby resulting in pH decline and diminishing growth of methanogenic bacteria and methanogenesis (Vicenta et al., 1984; Cuzin et al., 1992). It can also be explained that the inactivity during this period can be as a result of the inoculum that is in either methanogens or lag phase. Biogas production started on the 8th day in all the digesters except for digester A (100 wt. %, 0 wt. % poultry, and pig droppings respectively) that started production on the 16th day of fermentation. There was a steady increase in biogas production for all the digesters within a retention time of 20 – 40 days. The exponential phase that resulted in increase of biogas was as result of the exponential increase in micro – organisms which enhance an increase in the rate of fermentation which subsequently leads to a corresponding increase in biogas production. The pH of the slurries ranged between 5.7 – 7.1 and was observed among all the digesters. This observed change in pH may be due to the high volatile solids in the elephant grass which were transformed into volatile fatty acids and other acidic metabolites during acidogenesis due to the activities of the aerobes and facultative aerobes that were subsequently metabolized by the methanogenic bacteria to generate biogas (Okewale et al., 2016; Latinwo and Aremu, 2015; Dennis and Burke, 2001; Iyagba et al., 2009). The pH value was observed to increase in all the digesters as the days of fermentation is increased. As retention time increases with the increase in pH the biogas rate of production increases. This biogas yield increase suggests an increase in metabolic activity within the microorganisms present in the digesters.

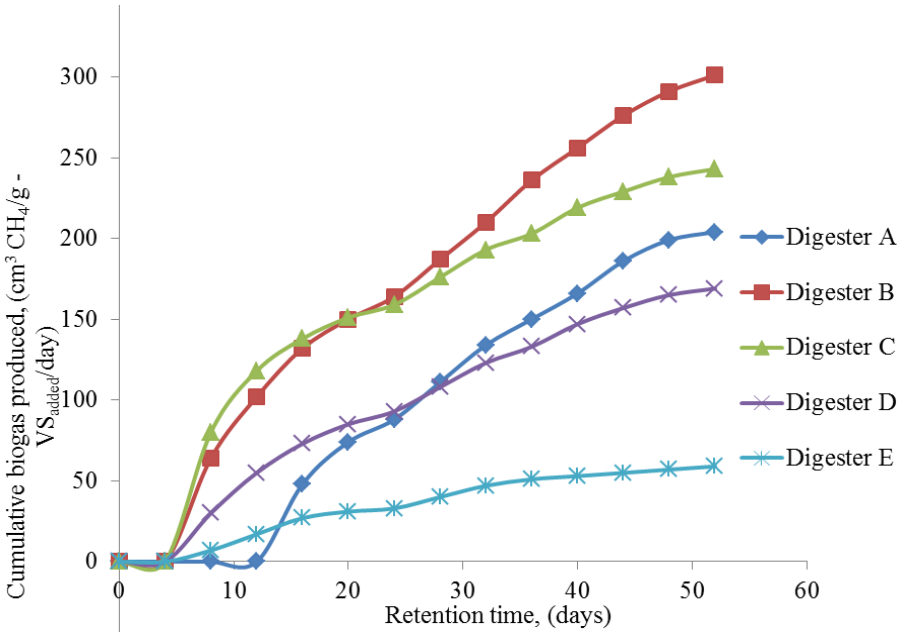

Fig. 5 Cumulative biogas produced with retention time.

3.4.2 Methane pathway

Generally, the methane pathway for biogas production shows that during methane digestion microbiological reduction of sulfates (VI) to sulphides and hydrogen sulphide occurs along with anaerobic ammonification and reduction of nitrates (V) to ammonia. Apart from assimilative reduction of nitrates (V), denitrification may occur (Scherer et al., 2000).

The first stage of methane digestion (hydrolysis) involved biochemical conversions of H_2 and CO_2 to methane and acetate to methane and CO_2 are various enzymes and prosthetic groups which occur only in methanogens. These compounds basic structure comprised of Deazariboflavine derivative F_{420}, methanopterin, methanofurane, nickel – tetrapyrol factor F_{430} and coenzyme M (mercaptan sulfonate) (Zieminski and Frac, 2012). Authotropic binding of CO_2 by methanogeness occurs without a share of

the reaction of ribulose – bisphosphatic cycle. Synthesis of cellular material with CO_2 occurs through the reductive pathway of aceto-CoA with pyruvate (Mashapu, 2005; Saxena et al., 2009). CO_2 is bound by methanofurane (MFR) at the initial stage which is then reduced to methenyl, methylene, methyl and at the final stage – methane, which is bound in turn by coenzymes: tetrahydromethanopterin, 2 – methylthioethanesulfonic acid and 2 – mercaptoethanesulfonic acids (Medigan et al., 2000). The hydrogenase accounts for the assimilation of H_2 this hydrogen activated by hydrogenases reacts with factor F420, which is a reducing force provided. The majority of the methanogenes use hydrogen as an electron source that is connected with hydrogenase occurrences (Zieminski and Frac, 2012). Reduction of CO_2 to methyl groups of pyruvate is accounted for by methanopterin while methyl groups in the carbonylation process are converted into carbonyl groups with a share of CO dehydrogenase enzyme (Saxena et al., 2009; Mashapu 2005). Many coenzymes which do not have any flavinic or quinonic groups are involved in the methanogenesis pathway. Methanogenes C1 participated in the metabolic pathway of methanofurane, methanopterin and coenzyme M, but coenzymes F_{420} and B act as electron donors. These contain monocarbon compounds, such as methane, methanol, dimethyl carbonate and other monocarbons. Methane is produced by methanogenic archaeons using carbon dioxide as an electron acceptor (Mashapu, 2005; Medigan et al., 2000)

3.5 Kinetic model analysis

As seen in figure 6, the rate of biogas production increases as the retention time increases. The coefficient of correlation (R^2) ranges from 0.9434 to 0.9902 for the five digesters. Digester B had the highest value of correlation coefficient 0.9902; this linear plot suggests that it was an ascending limb in digesters B, C, D, and E since the value of b is positive while it is a descending limb in digester A with the negative value of b coefficient. The numerical value of constants a and b in the digesters are; A(0.0482 cm^3CH_4/g - VS_{added}/day and -0.3245cm^3CH_4/g-VS_{added}/day), B(0.0522cm^3CH_4/g - VS_{added}/day and 0.3993 cm^3CH_4/g - VS_{added}/day), C(0.0339cm^3CH_4/g - VS_{added}/day and 0.762 cm^3 CH_4/g - VS_{added}/day), D(0.0302 cm^3CH_4/g - VS_{added}/day and 0.2009cm^3CH_4/g-VS_{added}/day), and E(0.011cm^3CH_4/g-VS_{added}/day and 0.0616 cm^3CH_4/g - VS_{added}/day). It can be established from figure 5 that the rate of biogas production will increase linearly with an increase in time (retention days). The rate of production would decrease after reaching a maximum point linearly to zero as retention time (days) increases.

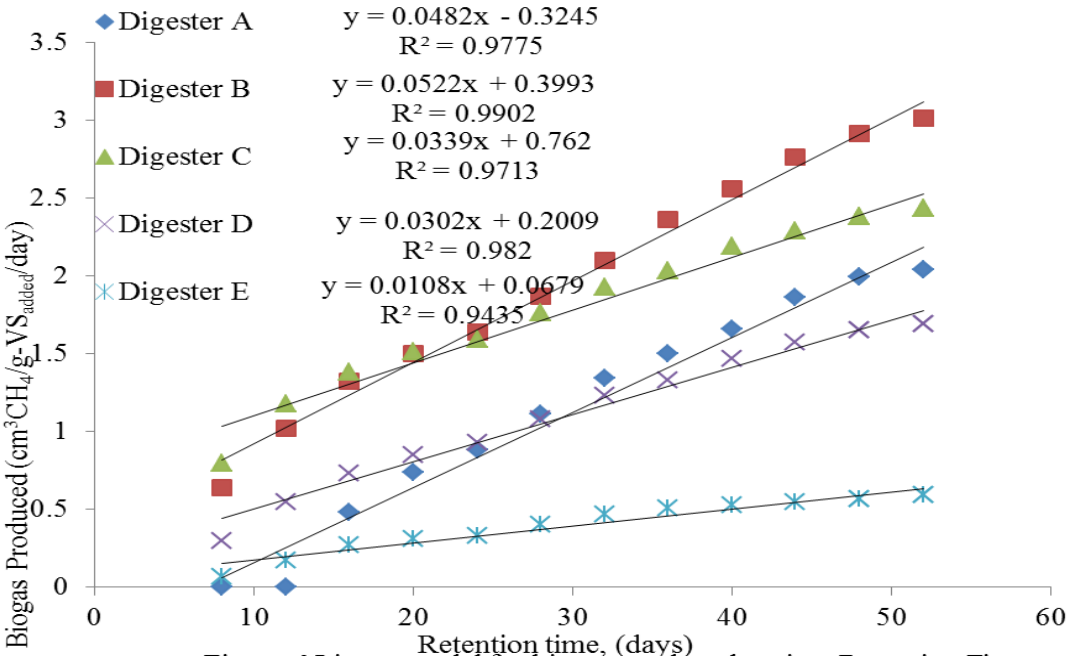

Fig. 6 Linear model for biogas produced against Retention Time.

3.6 Energy production from co – digestion of elephant grass, poultry, and pig droppings

The energy production from anaerobic co – digestion of elephant grass with poultry, and pig droppings for all the digesters are depicted in Figure 7. The energy productions were 8.17, 12.05, 9.73, 6.77, and 2.36kJ/g – VS_{added} for digesters A, B, C, D, and E respectively. These results showed that digester B provided an enhanced benefit in terms of boosting the energy production from these substrates while digester E did not give a better benefit in terms of energy production which might be due to the lignin, hemicellulose present in elephant grass thereby making it difficult for the microorganisms to digest the substrate during the fermentation process. The power generation in (watt) of each digester was calculated by dividing the overall energy production (J) by the overall fermentation time (hr). Therefore, power generation for digesters A, B, C, D, and E in 52 days of fermentation were 6.54, 9.66, 7.79, 5.42, and 1.89 watts, respectively. The amount of elephant grass, poultry, and pig droppings were assessed for establishing a 1MW (1 x 10^6watt) of biomass power plant. For digester A, the elephant grass, and poultry droppings needed are 10,000 kg- VS, and 15,000 kg- VS respectively. For digester B, the elephant grass, poultry, and pig droppings needed are 10,000 11,250, 3,750 kg- VS respectively while 10,000 kg – VS of elephant grass, 7,500 kg – VS poultry dropping, and 7, 500 kg – VS pig dropping were required in digester C. Digester D requires 10, 000 kg – VS elephant grass, 3,500 kg – VS poultry dropping, and 11,250kg – VS pig dropping. 10, 000 kg – VS elephant grass, and 15,000 kg – VS pig dropping are required by digester E.

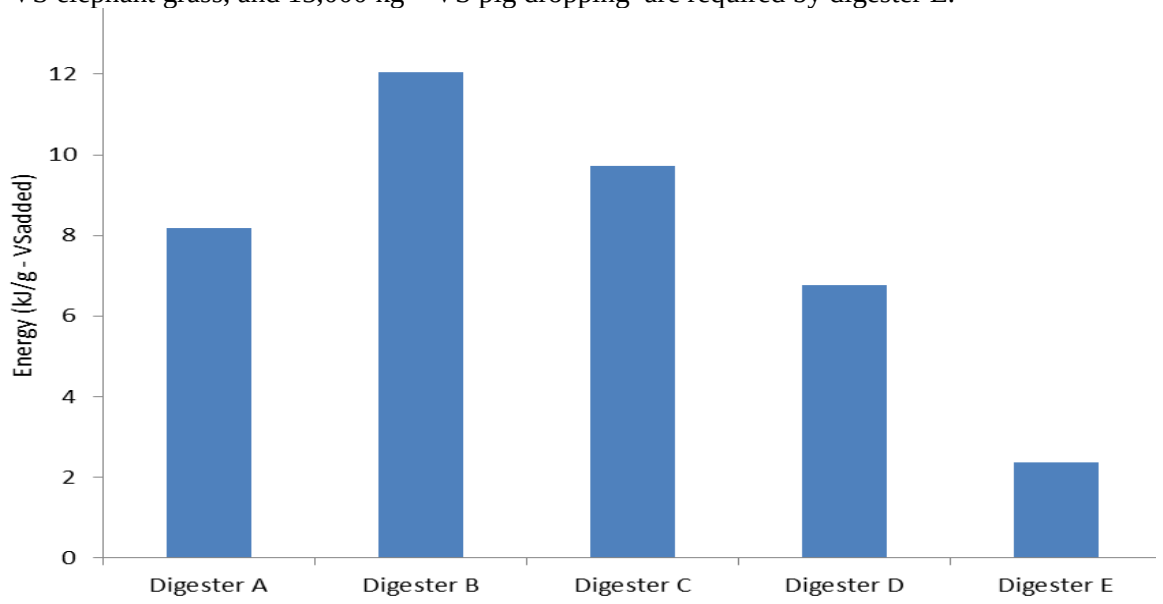

Fig. 7 Energy production for co-digestion of elephant grass, poultry, and pig droppings.

3.7 Gas chromatography analysis of biogas produced

It can be seen in Table 6, that the digester that produced the maximum biogas (digester B) had 69.43 % v/v of methane gas. It is seen in Table 6 that the biogas produced can be used as a source of heat in cooking since it is combustible with the methane (CH_4) concentration above 50 % v/v. This corroborated the reports of (Ezeonu et al., 2005; Igoni et al., 2008; Adeyanju, 2008; Graaf and Fendler, 2010), that the mixture of the gases is combustible if the methane content is more than 50%.

Table 6 Gas Chromatography Analysis of the Biogas Produced in Digester B.

Element	CH_4	CO_2	NH_3	H_2S	O_2	N_2	H_2O
Compositions (% v/v)	69.43	23.22	1.60	2.00	0.4	2.2	1.15

4. Conclusion

Anaerobic co-digestion of poultry droppings, pig droppings, and elephant grass for biogas production was carried out in this work. The co-digestion of poultry droppings (75 wt. %), pig droppings (25 wt. %), and elephant grass gave a higher cumulative biogas yield of 301 cm^3CH_4/g-

VS_{added}. The methane content of 69.43 % v/v was obtained in digester B. The presence of methyl groups, alkanes, and alkenes groups in pig droppings, poultry droppings, and elephant grass makes these materials good substrates for biogas production. The experimental data obtained from the biogas produced fitted well with the linear kinetic model. The net performance for each of the digesters are digester B > digester C > digester A > digester D > digester E. The GC analysis on the biogas produced in digester B had the highest biogas production 69.43 % v/v, 23.22 % v/v, 1.60 % v/v, 2.0 % v/v, 0.4 % v/v, 2.2 % v/v, and 1.15 % v/v for methane (CH_4), carbon dioxide (CO_2), NH_3, H_2S, O_2, N_2, and H_2O respectively. A rich value for fertilizer in initial plant waste is the residue of this anaerobic digestion. Low values of C/N and volatile solids (VS) present in poultry droppings enables them to perform better in biogas production compared to pig droppings. Digester B had the highest energy production value 12.05kJ/g – VS_{added} in 52 days of the fermentation process compared to the other digesters.

References

[1] Adelekan, B. A., & Bamgboye, A. I. (2009). Comparison of biogas productivity of cassava peels mixed in selected mixed in selected ratios with major livestock waste types. *African Journal of Agricultural Research Centre, 12*(1-12), 12 – 17.

[2] Adeyanju, A. A. (2008), Effects of seeding of wood-ash on biogas production using pig waste and cassava peels. *Journal of Engineering Applied Sci, 3*(3), 242–245.

[3] American Society for Testing and Materials, (1991). *Standard test methods for moisture in activated carbon*, Philadelphia, PA: ASTM Committee on Standards.

[4] American Society for testing and materials: Annual Book of ASTM Standard (1996). Volume 15.01, Refractories, *Carbon and Graphic Products*, Activated Carbon, ASTM, Philadelphia, P. A.

[5] APHA, (1995), *Standard Methods for Examination of Water and Wastewater*, 18th ed.; American Public Health Association: Washington, DC, USA.

[6] Bal., A. S., & Dhagat, N. N. (2001). Upflow anaerobic sludge blanket reactor – a review. *Indian J. Environ Health, 43*, 1-82.

[7] Bayer, E. A., Lamed, R., & Himmel, M. E. (2007). The potential of cellulases and cellulosomes for cellulosic waste management. *Curr Opin Biotechnol, 18*, 237 – 245.

[8] Chandra, R., Takeuchi, H., & Hasegawa, T. (2012). Methane production from lignocellulosic agricultural crop wastes: A review in context to second generation of biofuel production. *Renew. Sustainable Energy, 16*, 1462–1476.

[9] Brown, D., & Li, Y. (2013), Solid state anaerobic co-digestion of yard waste and food waste for biogas production Bioresource Technol, *127*, 275–280.

[10] Cirne, D. G., Lehtomaki, A., Bjornsson, L., & Blackhall, L. L. (2007). Hydrolysis and microbial community analysis in two – stage anaerobic digestion of energy crops. *Journal of Applied Microbiology, 103*, 516 – 527.

[11] Coates, J. (2000). Interpretation of Infrared Spectra: *A Practical Approach, in Encyclopedia of Analytical Chemistry*, Meyers, R. A., (Ed.), John Wiley and Sons Ltd., Chichester, 10815 – 10837.

[12] Cuzin, N., Farinet, J. L., Segretain, C., & Labat, M. (1992). Methanogenic fermentation of cassava peel using a pilot plug flow digester. *Bioresource Technol, 41*, 259-264.

[13] Dennis, A., & Burke, P. E. (2001). Dairy waste anaerobic digestion handbook, Environmental Energy Company, 6007, Hill Street, Olympia, W. A., 98516, 20.

[14] Dupont and Accorsi, (2006), Studies on biochemical changes in maize wastes fermented with Aspergillus niger. *Biokemistri, 19*(2), 75-79.

[15] El – Mashad, H. M., & Zhang, R. (2007). *Co-digestion of Food Waste and Dairy Manure for Biogas Production, Transaction, ASABE 50*, 1815 – 1822, 2007.

[16] El – Mashad, H. M., & Zhang, R. (2010). Biogas production from co – digestion of dairy manure and food waste. *Bioresource Technology, 101*, 4021 – 4028.

[17] El-Saeidy, E. A. (2004). Technology Fundamentals of Briquetting Cotton Stalks as Biofuel: an unpublished Ph.D. Thesis, Faculty Agriculture and Horticulture, Humboldt University, Germany.

[18] Ezeonu, S. O., Dioha, I. J., & Eboatu, A. N. (2005). Daily Biogas Production from different wastes and identification of methanogenic bacteria Involved, *Nigerian Journal of Solar Energy, 15,* 80 – 85.

[19] Graaf, D., & Fendler, R. (2010). *Biogas production in Germany, Federal Environment Agency*, Dessau – Rosslau, 29.

[20] Hattori, M., Iwase, N., Furuya, N., Tanaka, Y., Tsukazari, T., Maguire, M. E., Ito, K., Maturana, A., & Nureki, O. (2009). Mg^{2+} - dependent gating of bacterial MgtE channel underlies Mg^{2+} homeostasis, EMBO, 28:3602 – 3612.

[21] Igoni, A., Hikiah, Ayotamuno, M. J, Eze, C. L., Ogaji, S. O. T., & Robert, S. D. (2008). Designs of anaerobic digesters for producing biogas from municipal solid – waste. *Applied Energy, 8,* 430–438.

[22] Iyagba, E. T., Mangibo, I. A., & Mohammad, Y. S. (2009). The Study of Cow Dung as Co-substrate with Rice Husk in Biogas Production. *Scientific Research and Essay, 4*(9), 861 – 866.

[23] Karki, A. B., Shrestha N. J., & Bajgain S. (2005). *Biogas as Renewable Energy Source in Nepal: Theory and Development*, Nepal, BSP., Retrieved on August 17th, 2017, www.snvworld.org.

[24] Kaster, A. K., Goenrich, M., Seedorf, H., Liesegang, H., Wollherr, A., Gottschalk, G., & Thauer, R. K. (2011). More than 200 genes required for methane formation from H_2 and CO_2 and energy conservation are present in *Methanothermobacter marburgensis and Methanothermobacter thermautotrophicus*, Archaea.

[25] Kumar, S., Mondal, A. N., Gaikward, S. A., Devotta, S., & Singh, R. N. (2004). *Qualitative assessment of methane emission inventory from municipal solid waste disposal sites*: a case *study, Atmos. Environ, 38,* 4921–4929.

[26] Lalitha, K., Swaminathan, K. R., & Bai, R. P. (1994). Kinetics of Biomethanation of Solid Tannery Waste and the Concept of Interactive Metabolic Control. *Applied Biochemical Biotechnology, 47,* 73 – 87.

[27] Latinwo, G. K., & Agarry, S. E. (2015). Modelling the kinetics of biogas generation from mesophilic anaerobic co – digestion of sewage sludge with municipal organic waste. *Chemical and process engineering research, 31,* 43 – 53.

[28] Li, Y., Zhang, R. H.; Liu, X. Y., Chen, C., Xiao, X., Feng, L., He, Y. F. & Liu, G. Q. (2013). Evaluating methane production from anaerobic mono- and co-digestion of kitchen waste, corn stover, and chicken manure. *Energy Fuel, 27,* 2085–2091.

[29] Li, J., Jha, A. K., He, J., Ban, Q., Chang, S., & Wang, P. (2011). Assessment of the effects of dry anaerobic co – digestion of cow dung with waste water sludge on biogas yield and biodegradability. *International Journal of the Physical Sciences, 6*(15), 3723 – 3732.

[30] Mata – Alvarez, J., Macé, S., & Llabré, P. (2000). Anaerobic digestion of organic solid wastes. *An overview of research achievements and perspectives, Bioresour Technol, 74,* 3 – 16.

[31] Mashapu, N. (2005). *The microbial composition of natural methanogenic consortium*, A thesis M.Sc., University of Western Cape.

[32] Mathew, A. K., Bhui, I., Banerjee, S. N., Goswami, R., Shome, A., Chakraborty, A. K., Balachandran, S., & Chaudhury, S. (2014). Biogas production from locally available aquatic weeds of Santiniketan through anaerobic digestion, Clean Technology Environ., Policy.

[33] Medigan, M. T., Martinko, J. M., & Parker, J. (2000). Brock biology of microorganisms, Prentice Hall International, New Jersy.

[34] Mshandete, A. M., & Parawira, W. (2009). Biogas Technology Research in Selected Sub-Saharan African Countries A Review. *African Journal of Biotechnology, 8,* 116 – 125.

[35] Mudhoo, A., & Kumar, S. (2013). Effects of heavy metals as stress factors on anaerobic digestion processes and biogas production from biomass. *Int. J. Environ. Sci. Technol, 10,* 1383 – 1398.

[36] Ojolo, S. J., Dinrifo, R. R., & Adesuyi, K. B., (2007). Comparative Study of Biogas Production from Five Substrates. *Advanced Materials Research Journal*, 18-19, 519-525 published by Trans Tech, Retrieved from http://www.scientific.net

[37] Okewale, A. O., Omoruwuo, F., & Ojaigho, R. O. (2016). alternative energy production for environmental sustainability. *British Journal of Renewable Energy, 1*(2), 18 – 22.

[38] Ostream, K. (2004). Greening waste: anaerobic digestion for treating the organic fraction of municipal Solid wastes, Unpublished M.Sc. Thesis in Earth Resources Engineering, Department of Earth and Environmental Engineering Fu Foundation of School of Engineering and Applied Science, Colombia University.

[39] Parawira, W., Murto, M., Zvauya, R., & Mattiasson, B. (2004). Anaerobic digestion of solid potato waste alone and in combination with sugar beet leaves. *Renewable Energy, 29,* 1811 – 1823.

[40] Parkin, G. F., & Owen, W. F. (1986). Fundamentals of anaerobic digestion of wastewater sludges, *J. Environ. Eng, 112,* 867–920.

[41] Qiang, H., Langa, D. L., & Li, Y. Y. (2012). High – solid mesophilic methane fermentation of food waste with an emphasis on iron, cobalt, and nickel requirements, *Bioresource Technology, 103,* 21 – 27.

[42] Reungsang, A., Pattra, S., & Sittijunda, S. (2012). Optimization of Key Factors Affecting Methane Production from Acidic Effluent Coming from the Sugarcane Juice Hydrogen Fermentation Process. *Energies, 5,* 4746–4757.

[43] Ramansu, G., Pritam, C., Arunima, S., Sambhu Nath, B., Amit, K. C., Anil, K. M., & Shibani, C. (2016). An overview of physico – chemical mechanisms of biogas production by microbial communities: a step towards sustainable waste management. Biotech, 6, 1, 72.

[44] Rao, A. G., Prakash, S. S., Joseph, J., Reddy, A. R., & Sarma, P. N. (2011),. Multi stage high rate biomethanation of poultry litter with self-mixed anaerobic digester, *Bioresource Technology, 102,* 729 – 735.

[45] Saxena, R. C., Adhikari, D. K., & Goyal, H. B. (2009). Biomass – based energy fuel through biochemical routes. *A review, Renewable Sustainable Energy Rev, 13,* 167 – 178.

[46] Scherer, P. A., Vollmer, G. R., Fakhouri, T., & Martensen, S. (2000). Development of methanogenic process to degrade exhaustively the organic fraction of municipal grey waste under thermophilic and hyperthermophilic conditions, water Sci. *Technol, 41,* 83 – 91.

[47] Sittijunda, S. (2015). Biogas Production from Hydrolysate Napier Grass by Co-Digestion with Slaughterhouse Wastewater using Anaerobic Mixed Cultures. KKU Res. *J, 20,* 323–336.

[48] Themelis, N. J., & Verma, S, (2004), The Better Option: Anaerobic Digestion of Organic Waste in MSW, *Waste Management World*.

[49] Xie, S., Lawlor, P. G., Frost, J. P., Hu, Z., & Zhan, X. (2011), Effect of pig manure to grass silage ratio on methane production in batch anaerobic co-digestion of concentrated pig manure and grass silage. *Bioresource Technol, 102,* 5728–5733.

[50] Vicenta, M., Pacheco, G., Alamis, M. L. A., Anglo, P. G., Tan, B. V., & Silverio, C. M. (1984). A Study of some Factors Affecting Biogas Production from Pineapple Peelings, In: Bidin., R., Chong, C. N., Wang, C. W., (Eds), *Proceedings of the second ASEAN Workshop on Biogas Production Applied to the Management and Utilization of Food Waste Materials*, Kaula Terengganu, Malaysia, 189 – 202.

[51] Yokoi, H., Saitsu, A., Uchida, H., Hirose, J., Hayashi, S., & Takasaki, Y. (2001). Microbial hydrogen production from sweet potato starch residue, *J. Biosci. Bioeng, 91,* 58–63.

[52] Zhang, Y., Rodionov, D. A., Gelfand, M. S., & Gladyshev, V. N. (2009). Comparative genomic analyses of nickel, cobalt, and vitamin B12 utilization, BMC Genom.

[53] Zhai, N., Zhang, T., Yin, D., Yang, G., Wang, X., Ren, G., & Feng, Y. (2015). Effect of initial pH on anaerobic co-digestion of kitchen waste and cow manure. *Waste Management, 38,* 126–131.

[54] Klomjek, P.K. (2015). Characteristics of Wastewater and Rice Straw in an Anaerobic Co-Digestion System. *Journal of Science and Technology Mahasarakham University, 34*(5), 423-430.

[55] Jirukkakul, P. (2015). Study of Potential of Erianthus sp. for Biogas Production. *Thai Society of Agricultural Engineering Journal, 21*(1), 25-30.

[56] Chanathaworn, J. (2017). Application of Supporting Media for Improvement of Anaerobic Digestion Performance and Biogas Production. *International Journal of Renewable Energy*, 12(2), 65-74.

[57] Ruamtawee, N., Daosud, W., Laoong-u-thai, Y., & Kittisupakorn, P. (2016). Hybrid neural network modeling and optimization of an anaerobic digestion of shrimp culture pond sediments in biogas production process. *KKU Engineering Journal, 43,* 192-195.