ARID ZONE JOURNAL OF ENGINEERING, TECHNOLOGY & ENVIRONMENT
AZOJETE December 2022. Vol. 18(4):587-596
Published by the Faculty of Engineering, University of Maiduguri, Maiduguri, Nigeria.
Print ISSN: 1596-2644, Electronic ISSN: 2545-5818
www.azojete.com.ng

ORIGINAL RESEARCH ARTICLE

INFLUENCES OF pH AND TEMPERATURE ON THE ANAEROBIC FERMENTATION OF GROUNDNUT SHELL AND COW DUNG FOR BIOGAS PRODUCTION

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ARTICLE INFORMATION

Submitted 25 January, 2022
Revised 26 April, 2022
Accepted 30 April, 2022

Keywords:
Biogas production
Substrate pH
Temperature
Groundnut shell
Cow dung

ABSTRACT

The increasing demand, high cost and health implications of using energy derived from hydrocarbon compound have necessitated the continuous search for alternative sources of energy. Groundnut shell (GS) and Cow dung (CD) as renewable source of energy supply have been proven to be very efficient. This study investigated the influences of operating parameters in anaerobic fermentation of GS and CD in the laboratory scale using the simple locally fabricated digesters labeled A-E of 4,000cm^3 working volume. The temperatures were recorded one time daily using a thermometer that was fitted to the digester. The digesters were fed on a batch basis with the slurry of different mix ratio and operated at ambient temperature (28-40°C) for 30days. Digester A being the control containing 100% of CD have pH of 7.00 and 7.20 before and after digestion respectively. This shows that the pH values of cow dung was neutral before digestion and slightly alkaline after digestion while digester E containing 100% of groundnut shell also a control have pH of 7.10 and 7.20 respectively before and after digestion. The temperatures in the five digesters fluctuated optimally between 28°C and 40°C which conforms to the mesophilic range. Most microorganisms grow best under neutral pH conditions, since other pH values may adversely affect metabolism by altering the chemical equilibrium of enzymatic reactions, or by actually destroying the enzymes. Low pH can cause the chain of biological reactions in fermentation to cease.

1.0 Introduction

Production of waste materials is an undeniable part of human society. The wastes are produced by several sectors including industries, forestry, agriculture and municipalities. The accumulation of waste and the "throw-away philosophy" results in several environmental problems, health issues and safety hazards, and prevent sustainable development in terms of resource recovery and recycling of waste materials (Isa *et al.*, 2014). A perspective aimed at promoting greater sustainable development and resource recovery has influenced solid waste management practices, and is gradually becoming implemented through policy guidelines at national levels in a number of industrialized and even developing countries (Isa *et al.*, 2014). Guidelines and directives to reduce waste generation and promote waste recovery are laid down according to the "waste management hierarchy", in which waste retention, reuse, recycling and energy recovery are designed to minimize the amount of waste left for final, safe disposal (Isa *et al.*, 2004). Potentially, all organic waste materials contain some quantities of nutrients essential for the growth and metabolism of anaerobic bacteria in biogas production. Again, every biodegradable material will produce biogas but

the quantity and quality of gas produced will vary depending on the feedstock used (REN21, 2017).

Cattle dung is the most suitable material for biogas plants because of the methane producing bacteria already contained in the stomach of ruminants. The specific gas production, however, is lower and the proportion of methane is around 65% because of pre-fermentation in the stomach (Maramba, 1978). Its homogenous consistency is favourable for use in continuous plants as long as it is mixed with equal quantities of water (Figure 1). Liquid cattle manure, a mixture of dung and urine, requires no extra water. However, the simple animal housing found on most farms in developing countries normally does not allow the collection of all animal excrement. Hence, most of the urine with its valuable plant nutrients is lost. The main advantage of animal manure, with respect to continuous digestion, is that it is easy to collect and easy to mix as slurry and load into digesters.

Figure 1: Cow dung sample

Arachishypogaea(Groundnut) is a native of South America but its cultivation is now widespread globally (Duke, 1981). It was introduced to the African continent during the colonial era (Duke, 1981). Groundnut is produced in Africa majorly by Nigeria, Sudan, Senegal, Chad, Ghana, Congo and Niger (Taphee *et al.*, 2014). Groundnut pyramids were a success story of the Northern Nigeria (Kano State especially) prior to independence while its farming remains a popular practice in Northern Nigerian with the fruit pods being put to no usage (Taphee *et al.*, 2014). Prior to this study, the potentials of groundnut shell (Figure 2) in biogas generation in Nigeria have been reported in few recent studies (Yavini *et al.*, 2014; Ibrahim *et al.*, 2016). The objective of this study is to determine the influence of pH and temperature on anaerobic fermentation of groundnut shell and cow dung for biogas production.

Figure 2: *Arachishypogaea* Groundnut Shell

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2. Materials and Methods

The waste materials that were used in the study are cow dung and groundnut shell. Groundnut shell was collected from a milling station at Pati Shabakolo, a village in Lavun Local Government Area of Niger State, Nigeria, during the 2019/2020 harvest season. The sample was collected in clean bags and transported to the site of experiments, while cow dung was sourced from Federal University of Technology, Minna farm. The waste materials were manually sorted to remove foreign materials and groundnut shell was sun dried for about fourteen (14) days in order to reduce the moisture content and for ease of handling. Groundnut shell was further crushed mechanically using pestle and mortar for size reduction and milled into powdered form and finally sieved with about 1.18µmm sieve tray.

2.1 Anaerobic Digester Set-Up and Operation

A 4,000cm^3 plastic container was obtained from Kure's market in Minna, Niger State, washed and all stains removed. Two holes were drilled; one at the centre with about 1.25cm diameter, and the other drilled at the side of the container with a diameter of 1.25cm. A reinforced flexible hose pipe of 100cm was inserted into the hole that was drilled at the centre of the cover. This pipe served as the gas outlet of the bio digester. It is then tight firmly and glued with epoxy resin steel adhesive in order to prevent any form of leakages and was connected to 2000cm^3 capacity container which served as the water chamber. The 1.25cm diameter side hole was fitted with ⅜ flexible hose pipe, male and female socket and ½inch plug where the sample was taken for *p*H (Figure 3 and 4).

The *p*H was measured daily using a digital *p*H meter. The sample to be analyzed were collected into a dry bottle from the digester and then analyzed. The probe of the *p*H meter was immersed into the samples to be analyzed and the meter was allowed to stabilize before the reading was taken. A hole was drilled at the side of the digester opposite the 1.25cm diameter but of 1.10cm diameter where the thermometer probe was fitted tightly with adhesive gum. The temperature reading was taken between 2pm and 4pm daily throughout the period of the experiment and also the ambient temperature.

Figure 3: The Digester showing the male female socket

Figure 4: The Digester and the Gas Holder

2.2 Biogas Digestion Assay

The slurry combination was formulated to contain about 5% solid content and the bio digester was filled with the slurry to 75% of the digester volume.100% of cow dung and 0% of groundnut shell were mixed with water for Digester A.75% of cow dung and 25% of groundnut shell were mixed with water for Digester B.50% of cow dung and 50% of

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Groundnut shell were mixed with water for Digester C.25% of cow dung and 75% of groundnut shell were mixed with water for Digester D.0% of cow dung and 100% of groundnut shell were mixed with water for Digester E. The slurry was stirred properly to avoid lump, and poured into Bio-digester A, B, C, D and E respectively. The digestion was allowed for a period of 30days under ambient temperature (psychrophilic).The pH of the medium was measured daily in order to ensure that the pH value is within the range at which the biogas can be produce. The temperature of the medium was taken 1time daily

Proximate and ultimate composition of the groundnut shell and cow dung was carried out according to the method of AOAC (2010).

2.3 Proximate Analysis

2.3.1 Determination of Moisture Content (MC)

The hot oven air method of Association of Official Analytical Chemists (AOAC, 2010) would be adopted for this analysis. Porcelain crucibles was washed and dried in an oven at 100°C for 30min. These were allowed to cool in the desiccators. About ten (10g) of the substrates were placed into weighed crucibles and placed in an oven at 105°C for 4h. The samples were removed from the oven after this and were cool and weighed. The drying will be resumed and all the crucibles with the samples will be re-weighed until a constant weight is obtained. The percentage moisture was calculated from the loss of weight of the sample using the following formula;

$$\%MC = \frac{W_1 - W_2}{W_1} \times 100 \qquad (1)$$

Where

W_1 = weight of the original sample

W_2 = weight of final dried sample

2.3.2 Determination of Total Solids (TS)

It is the amount of solid present in the sample after the loss of water molecules present in it. In other words, is refers to as the quantity of the material residue left in the crucible after evaporation of the sample and its subsequent drying in a laboratory oven at 105°C for a period of one hour.

Two crucibles were properly washed and dried in the laboratory oven at a temperature of 105°C for one hour. The crucibles were stored and cooled in a desiccator until needed. The crucibles were weighed (W_2) before use. Laboratory oven was switch on and allowed to reach a temperature of 105°C. This temperature was maintained throughout the experiment. Substrates were added to the crucibles (W_3) and diligently placed in the laboratory oven at a temperature of 105°C. The substrate samples were dried to a constant mass for a period of 1 to 2hours. The crucibles plus substrate residues were allowed to cool in a desiccator to balance temperature. Desiccators are designed to provide an environment of standard dryness. Desiccator was properly lubricated with grease and this is to prevent moisture from entering the desiccator as the test glassware cools. Crucibles plus substrate (material) residue were weighed using electronic precision balance (W_1). Equation (2) was used to determine the percentage of TS.

$$\%TS = \frac{W_1 - W_2}{W_3 - W_2} \times 100 \qquad (2)$$

%TS = Percentage total solid

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W_1 = Weight of dried crucible + dried residue
W_2 = Weight of crucible
W_3 = Weight of wet sample (substrate) + crucible

2.3.3 Determination of Volatile Solids (VS)

The VS is the solid remaining after evaporation or filtration are dried, weighed, and ignited at 600°C. In the determination of VS of the substrates the residues obtained from TS determination were ignited at 600°C for a duration of 30minutes using a muffle furnace. The crucibles and black mass of carbon were allowed to cool partially in air before it was transferred to the desiccator for complete cooling. The samples were weighed once temperature balance is reached (W_4)

The percentage VS were determined using equation (3).

$$\%VS = \frac{W_1 - W_4}{W_1 - W_2} \times 100 \qquad (3)$$

Where,
%VS = Percentage Volatile solid
W_4 = Weight of crucible + weight of residue after ignition

2.4 Ultimate Analysis

The ultimate analysis determines the weight percentage of element present in biomass like carbon, nitrogen, hydrogen, oxygen and sulphur.

2.4.1 Determination of Carbon Content

This was determined using the Walkey and Black method. 10g GS each of the finely ground substrate were weighed into 500ml conical flasks, appendix h. Potassium dichromate 10ml) was poured inside the flasks and the mixture was swirled. H_2SO_4 (20ml) were added and the flasks swirled again for 1min in a fume cupboard. These mixtures were allowed to cool for 30min after which 200ml of distilled water, 1g of NaF and 1ml of phenylalanine indicator were added. The mixture was then shaken and titrated with ferrous ammonium sulphate solution in a burette. The blank was also treated similarly. The percentage carbon content was calculated using equation (4);

$$\%Carbon = \frac{B - T \times 133 \times 0.003 \times 100}{W} \qquad (4)$$

Where;
B = Blank titre value
T = Sample Titre value
C = Concentration of Fe solution
W = Weight of waste sample

2.4.2 Chemical Oxygen Demand (COD)

Chemical Oxygen Demand (COD) is a measurement commonly used to determine substrate quality. The COD values indicate the amount of oxygen (in milligrams per liter of product) needed to oxidize or stabilize these wastes. Biochemical oxygen demand (BOD) and chemical oxygen demand (COD) are two different ways to measure how much oxygen the wastewater from a digester will consume when it enters the environment. Industries normally focus more on COD and municipalities more on BOD removal. Efforts must be

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made to reduce these values to protect the environment (Nwaigwe and Enweremadu, 2015).

2.4.3 Determination of Nitrogen

This was carried out using the micro-Kjeldahl method described by Pearson, 1976. The method involves estimation of the total nitrogen in the sample and subsequent conversion of the nitrogen to protein with the assumption that all the protein in the samples are present as nitrogen. Using a conversion factor of 6.25, the actual percentage of protein in the sample was calculated using equation (5) Micro-Kjedahl digestion/distillation apparatus and 50ml Kjeldahl flasks were utilized in carrying out the analysis.

$$\% \text{ crude protein} = \% \text{ nitrogen} \times F \quad (5)$$

Where:

F = conversion factor (6.25)

2.5 Digestions

Samples (2g) were weighed in Kjeldahl flasks appendix 6h - j. Catalysts such as sodium sulphate and copper sulphate were added in the flasks in the ratio of 3:1. Oxidizing agent (conc. H_2SO_4, 15ml) was then added, glass beads were added to prevent bumping during heating. Heating was carried out cautiously on a digestion rack under fume cupboard until a greenish clear solution appeared. The digest was allowed to clear for about 30min; it was heated for another 30min and allowed to cool. About 10ml of distilled water was added to avoid caking after which the digest was transferred with several washings into a 25ml volumetric flask and made up to the mark with distilled water.

2.5.1 Distillation of the protein

A 50ml receiver flask containing 5ml boric acid (methyl red and blue indicator) will be placed under the condenser of the distillation apparatus so that the tip will be 2cm inside the indicator. A 10ml of 40% NaOH solution was added to the digested sample in the apparatus through the funnel stop cork. Closing the steam by-pass and opening the inlet stop cork on the steam jet arm of the distillation apparatus started off the distillation. The distillate was collected in the conical flask (35ml) with its indicator – methyl red and blue. Titration was then carried out using 0.01M HCl to first pink colouration. The percentage of nitrogen and protein was calculated using equation 6 v;

$$\%Nitrogen\ (N) = \frac{Titre \times 0.0014 \times 250}{Weight\ of\ original\ sample} \times 100 \quad (6)$$

3. Results and Discussion

3.1 Feedstock Characterisation

The results of the physico-chemical analyses of the substrates prior to anaerobic digestion are shown in Tables 1. The result of chemical analyses shows that steam explosion reduces the total solid of GS from 87.90% to 79.42% while the volatile solid was increased from 75.11% to 86.32% as a result of steam explosion pre-treatment. Though the nitrogen content of GS increases after steam explosion the carbon content remain barely constant even after steam explosion. Carbon to nitrogen ratio is one of factor affecting the anaerobic process; it affects methane yield and production rates. It is often suggested that an optimum C/N ratio should be between 20:1 and 30:1 In Table 2 lignocelluloses content of the substrate are: Hemicellulose before and after pre-treatment are 34.11% and 40.20%

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respectively, Cellulose are 30.50% and 28.80% while lignin before and after pre-treatment are 35.39% 31.00% respectively. Hemicellulose consists of several type of sugar unit and sometimes referred to by sugars they contain. Hemicellulose is associated with cellulose and contributes to the structural component of the plant (Rowell, 2005). Cellulose is a main structural component in a plant cell.

Table 1: Characteristics of the Substrates

Properties	Cowdung	Groundnutshell	Pretreated (GS)
Moisture Content (%)	89.50	25..89	81.21
TS (%)	19.60	87.90	79.42
VS (%)	54.01	75.11	86.32
VS/TS ratio	2.76	0.86	1.09
Carbon Content	42.00	62.02	61.90
Nitrogen Content	0.38	0.50	0.70
N/C ratio	0.01	0.01	0.01

Table 2: Lignocellulose Content of Groundnut Shell

Properties	Not treated	Physically pre-treated
Hemicellulose (%)	40.20	34.11
Cellulose (%)	30.50	28.80
Lignin (%)	35.39	31.00

3.2 Operational Parameters

The pH for each of the digesters was measured before and after digestion. The daily ambient and digesters temperatures were adequately monitored.

3.2.1 pH of the Digesters before and After Digestion.

Figure 5 shows the pH values of the media in all the digesters, and were varied almost in the optimal limits of methanogenic bacteria (pH: 6.1 -7.4), all the digesters showed a general increase in pH with minimal fluctuation. This progressive increase in pH before biogas production could account for steady rate of gas production which is in agreement with the studies carried out previously by Ahmadu (2009) and Igboro (2011). Methanogenic bacteria are very sensitive to pH and do not thrive below a value of 6.0 (Karki *et al.*, 2005). Most microorganisms grow best under neutral pH conditions, since other pH values may adversely affect metabolism by altering the chemical equilibrium of enzymatic reactions, or by actually destroying the enzymes. The methanogenic group of organisms is the most pH sensitive. Low pH can cause the chain of biological reactions in digestion to cease.

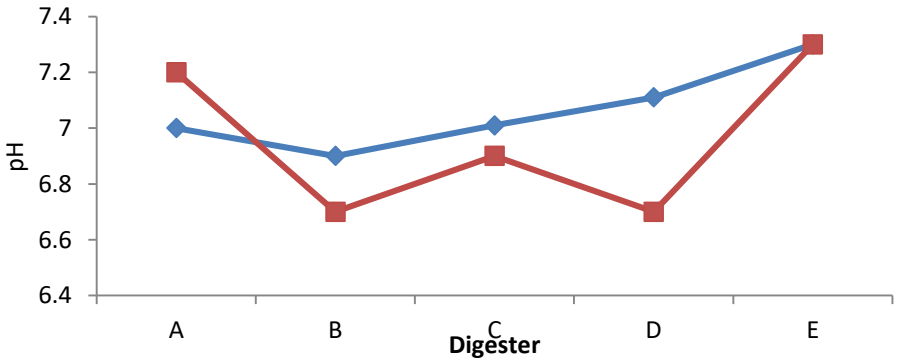

Figure 5: pH of the Slurry before and after Biogas production

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Figure 6 shows that digester A being the control containing 100% of Cow dung have pH of 7.00 and 7.20 before and after digestion respectively. This shows that the pH values of cow dung was neutral before digestion and slightly alkaline after digestion while digester E containing 100% of groundnut shell also a control have pH of 7.10 and 7.20 respectively before and after digestion. The pH of the digester C containing 50% each of cow dung and groundnut shell increases from day one of the digestion to day 7 of the digestion and toward the end of the digestion, the pH shows slightly alkaline solution. Digesters A and E have the highest pH of 7.6 in day five of the digestion and 7.5 in day 30 of the digestion respectively.

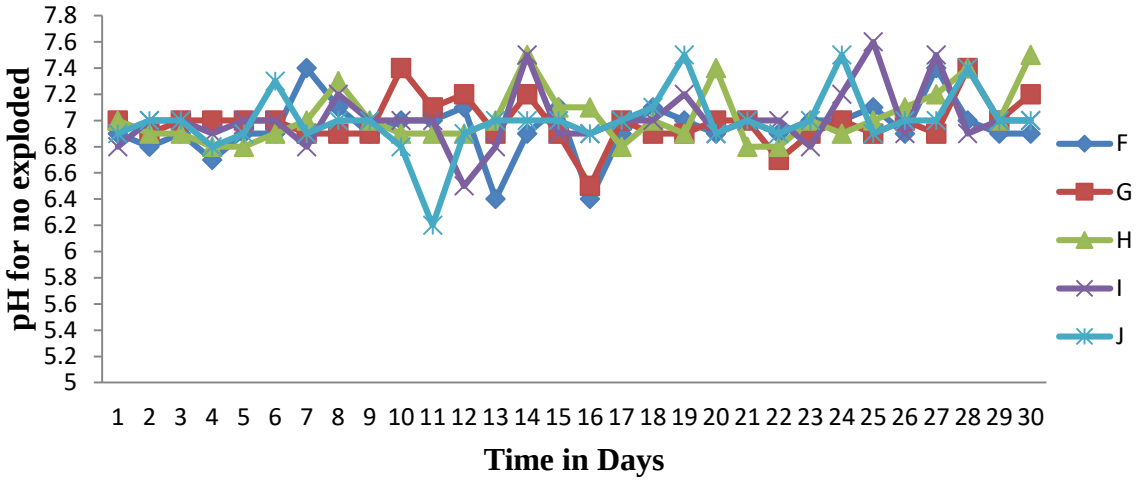

Figure 6: Variation of pH of the Digesters with time

3.3 Digester Temperature during Biogas Production

Figure 7 shows that the temperatures in the Five digesters fluctuated optimally between 28°C and 40°C which conforms to the mesophilic range. The temperatures were recorded once daily using a thermometer that was fitted to the digester. The foregoing show that the digesters operated within the mesophilic temperature range; and that it is possible to install digesters that will operate within this range in Minna and environs as reported elsewhere (Igboro, 2011 and Alfa, 2013). Since all the digesters were operated simultaneously, the temperature across them were the same as shown in Figure 7

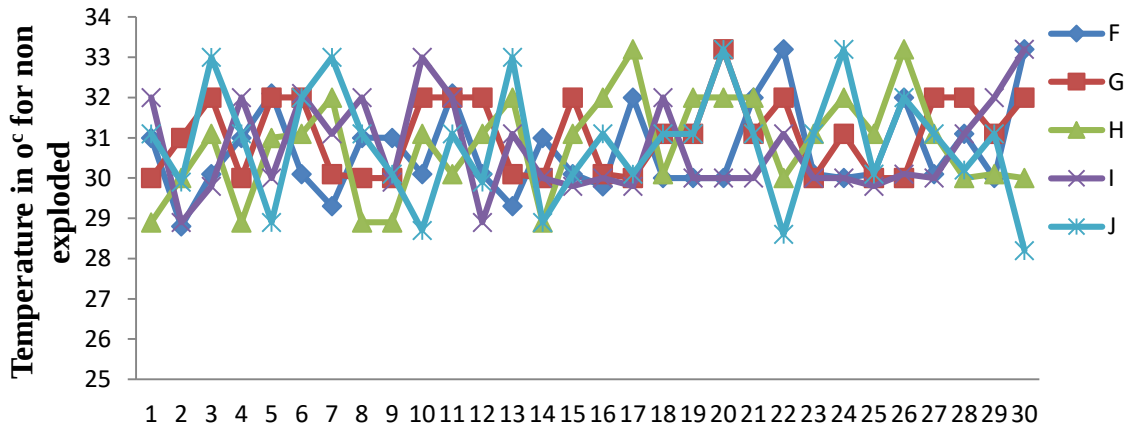

Figure 7: Variation of Digesters Temperature (°C) with time

The study of biogas production from co-digestion of groundnut shell and cow dung was conducted in digesters labeled A-E as shown. Biogas production was monitored and measured until biogas production reduced significantly. Figure 8 shows the fluctuation in the

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quantity of gas produced from each substrate possibly due to variation in the ratio of the substrates. Biogas production was very low in the first week of setup in all the digesters. As the feedstock matures over the days, gas production increases. The co-digestion of 25% CD-75% GS, 50% CD-50% GS, and 75% CD-25% GS have their highest gas production around sixteen and twenty-first day of retention period respectively while their least were recorded toward the end of the retention period. Also, digester A and E containing 100% each of CD and GS being the control for the pre-treated have their highest biogas production around eighteen and twenty first day of retention period respectively and the least are also seen toward the end of the digestion

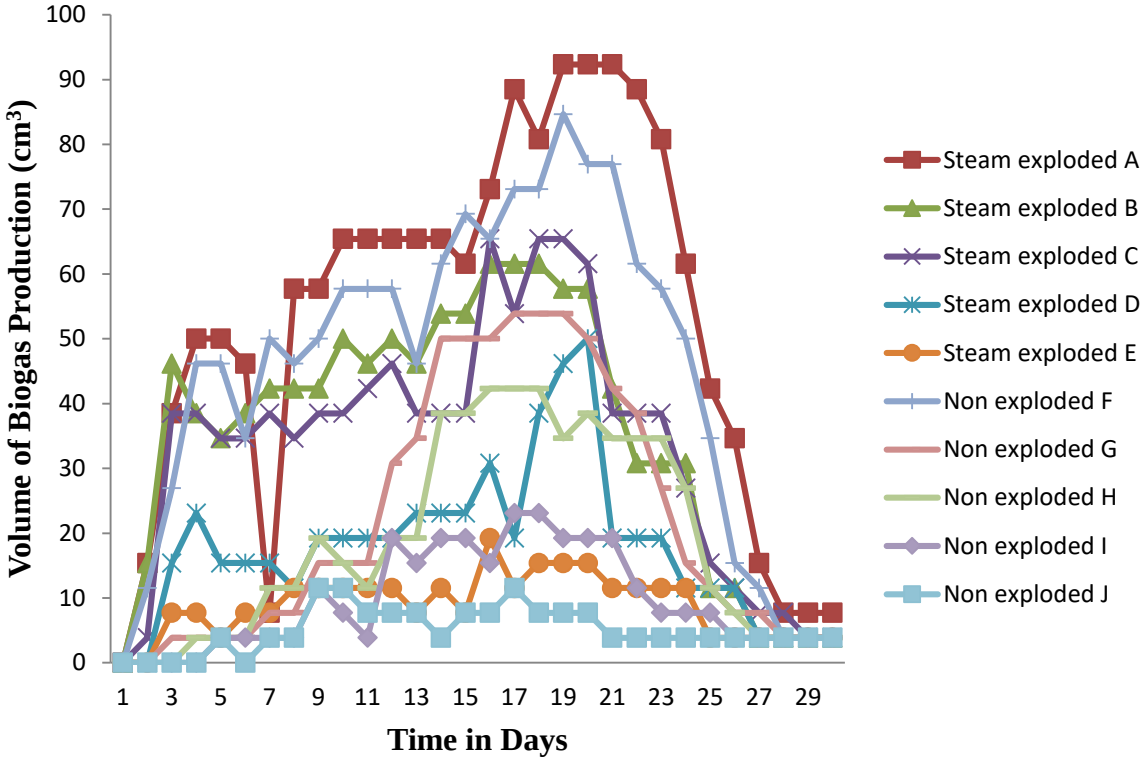

Figure 8: Variation of Volume of Biogas Production (cm^3) with time

4. Conclusion

From the results of the study conducted, a 4,000cm^3 biogas digester fabricated using locally available materials and tested under the existing weather condition in Minna were used for co-digestion of CD and GS. The *p*H values recorded before and after digestion indicates that the digesters operated well. The temperatures inside the digesters were stable fluctuating around 28°C to 40°C which is within the mesophilic range.

Now agricultural waste could be an alternative renewable source of energy in the form of biogas. Anaerobic digestion of GS and CD increased the cumulative biogas yield. The most important conclusion that can be drawn from this research is that neutral pH affects the quantity of biogas production. Also, most microorganisms grow best under neutral pH conditions, since other pH values may adversely affect metabolism by altering the chemical equilibrium of enzymatic reactions, or by actually destroying the enzymes. Low pH can cause the chain of biological reactions in digestion to cease.

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