INVESTIGATION INTO THE MICROBIAL ACTIVITIES IN BIOGAS PRODUCTION USING COW DUNG

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ABSTRACT

Biogas as major sources of energy for most human activities, it plays energetic role in all most operations with energy application. Biogas production field tests were conducted to investigate the microbial activities using cow dung. The experimental field bio-digester volume for the study was 1000 liters of plastic tank. The materials used for the biogas production were cow dung, GEEPEE tank, ball gate valves, filters, pressure meter, male and female adopter, hose, PVC pipe, gas holder, plastic funnel, metal clip and two slots. The parameters such as Total heterotrophic bacteria count (THBC), total heterotrophic fungi count (THF), total coliform counts and Total *Vibrio* counts, Total *Salmonella-Shigella* Counts and Total *Staphylococci species* count and Total *Pseudomonas species* count were determined with their recommended operation procedure. Also, the volume of biogas produced was determined by the aid of the gas holder. The total amount of biogas produced was 0.2012m^3 for the period of 54 day of the experiment. Results reviewed that high level of total heterotrophic bacteria count (THBC) followed by total Staphylococci count and total heterotrophic fungi count (THFC) and final total coliform count. There are variations in percentage of the microorganisms present in the cow dung. Furthermore, the results revealed that the amount of cow dung required generating a specific amount of biogas depends on size of the bio-digester used, gas holder and the microorganisms present. As a result, the amount of cow dung, microbes present and gas holder determine the quantity and quality of biogas produced. Hence, recommends that microbial activities are vital factor during the production of biogas.

Keywords: Biogas Production, Cow Dung, Microorganisms, Microbial Activities, Bio-Digester.

INTRODUCTION

Biogas technology, the generation of combustible gas from anaerobic biomass digestion, is well-known technology. There are already millions of biogas plants in operation throughout the world. Whereas using the gas for direct combustion in household stoves or gas lamps is common, producing electricity from biogas is still relatively rare in most developing countries. In Germany and most industrialized countries, power generation is the main purpose of biogas plants. Conversion of biogas to electricity has become a standard technology, it's unfortunate that in spite of land mark biogas has gain in power generation is not practice in Nigeria (Adelekan, 2002). Biogas is a mixture of methane, carbon dioxide and traces of hydrogen sulphide (Nijaguna, 2002). It is generated from human excreta, animal dung, poultry droppings, and sewage sludge, among others. A biogas plant can convert animal manure, green plants, waste from agro industry and slaughterhouses into combustible gas. Biogas can be used in similar ways; as natural gas in gas stoves, lamps or as fuel for engines

The source of animal waste used in anaerobic digestion is important in ensuring a successful operation of the process because of the lignum components of the animal manure, monogastric animals are known to produce waste that contain more nutrient than ruminants. Ruminant are known to excrete more lignocellulose materials due to extensive enzymic exposure in their four chambers stomach (Werner *et al*., 1989; Willkie, 2005). The high presence of lignum in animal waste can be resist anaerobic degradation even after long retention time (Van, 1993) or may prevent the anaerobic process from commencing Huang and Crookers (2005) thus, a high volatile solid content of substrate may not be necessary translate to high biogas yield due to presence of non-available volatile solid in form lignum. It is important to note that the volatile matter content of any substrate account for the proportion of solid that is transformed into biogas (Willkie, 2005). Other important criteria that were shown to affect biodegradability of substrate in Chad are the carbon to nitrogen ratio in the presence of specific substance like protein, carbohydrate lipids (Jung *et al.,* 2007). Thus, the chemical composition of organic substrates can be said to contribute to the pattern of degradation of such substrate and attempt to quantify this biodegradable substrate fraction were carried out (Chandler *et al.,* 1980; Huang & Crookers, 2005). Hence, for a successful biodegradation to take place, the process of co-digestion of animal waste must provide a balance between the lignum content and the carbon to nitrogen ratio (Huang & Crookers, 2005).

LITERATURE REVIEW

There are several conditions and variables that must applied in order to obtain a proper breakdown of the organic compound. The rate at which the microorganisms grow if paramount importance in the anaerobic digestion process. The operating parameters of anaerobic degradation efficiency of the system are discussed as follows (Verma, 2014; Monnet, 2013).

Mixing anaerobic digester content: mixing within the digester improves the contact between the microorganism and substrate and improves bacterial populations' ability to obtain nutrients. Mixing also prevents the formation of scorn and the development of temperature gradients within the digester. However, excessive mixing can disrupt the microorganism and therefore slow mixing is preferred (Monnet, 2013). Mineral irons, heavy metals and detergents used in livestock husbandry are some of the toxic materials that inhibit the normal growths of pathogens in the digester. Small quantity of mineral irons (e.g. Sodium, potassium, calcium, ammonium and Sulphur) also stimulates the growth of bacterial while very heavy concentration of this irons will have toxic effects (Karki *et al*, 2015)
According to Karki *et al* (2015), the following factors affect the microbial activities in an anaerobic digester.

- Nature of slurry: for proper solubilization of organic material the ratio between solid and water should be 1:1 when the domestic waste are used
- Seeding of bacterial population: actogenic and methanogenic material are naturally present in cow dung, however, there number is quite small. Acid forming bacterial proliferate fast and increase their number, while methanogenic bacterial develop very slowly. Therefore, for the initial reaction, small amount of sludge of another digester is generally used as seeding or inoculums. This sludge contain high concentration of actogenic and methanogenic bacterial which could enhance the process of anaerobic digestion of organic material

Methanogenic bacteria are anaerobic organisms. In anaerobic condition, most of these bacteria are inactive in metabolisms, thus, digesters should be totally air tight to maintain

strictly anaerobic condition. In many places, digesters are buried in the earth to maintain anaerobiosis condition.

Anaerobic digestion mainly takes place at either mesophilic (25^{o}C-40^{o}C) or thermophilic temperature (45^{o}C-60^{o}C) although it can take place at psychrophilic condition (12^{o}C-30^{o}C) too (Usman *et al.,* 2011). The process of anaerobic digestion has the potential of converting biodegradable organs into biogas which comprises methane (55-75%) and carbon dioxide (25-45%) with calorific value of 20mj/m^{3} (Myles, 1985; Steffens *et al*., 2000). Biomass can therefore be a source of decentralized energy source for developing countries especially in this era of insecurity and unpredictability in fossil fuel supply.

According to Van (1993) reported that any technology that tries to harness optimum use of available resources in a given environment while minimizing the negative environmental consequence as appropriate technology. The need for alternative source of energy for both decentralized and centralized power generation has led to the proliferation of research into alternative energy source. Anaerobic digestion (AD) received considerable interest as one of such means of meeting both decentralized and centralized power in recent years (Sixt, 1987). However, there is not yet sufficient information on the investigation into microbial activities in the production of biogas using cow dung. Therefore, there is need to research on the investigation of microbial activities in the production of biogas using cow dung. The aim of this study was to investigate the performance of microorganisms during biogas production using cow dung.

METHODOLOGY

Experimental Site

The experiment was conducted at Plot 142D Trans Amadi Industrial Layout in Oginigba in Obio Akpo Local Government Area, Port Harcourt, Rivers State, Nigeria (4° 49′ 27″ N, and longitude of 7° 2′ 1″ E) The map of the experimental area is shown in figure 1. The experimental design adopted was completely randomized design (CRD). Biogas plant (1000 liters GEEPEE tank) was constructed that suit the use of the cow dung; the fresh cow dung used in this study was collected from an abattoir in Oginigba (Plate 1). The abattoir at Oginigba, Port Harcourt, Nigeria was chosen because of the nearness to bio-digester. Three bio-digesters volume of 1000 liters each was used as replicate to ascertain reliability of the experiment.

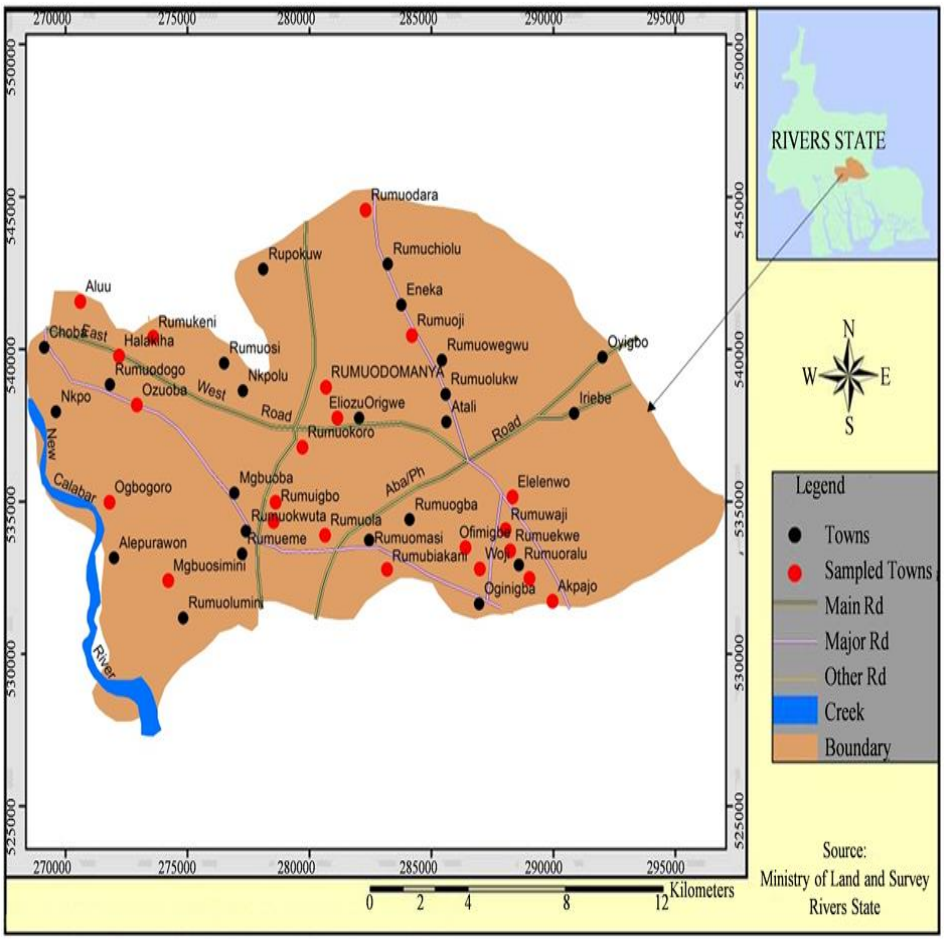

Figure 1: Map of Obio Akpo Local Government Area, Rivers State, Nigeria
Source: Rivers State Ministry of Land Survey

Plate 1: Collection of Cow Dung from an Abattoir in Oginigba

Digester Specification

A Plastic bio-digester made of locally available materials (Nigeria) was used for this study (Plate 2). The bio-digester is made up of 1000 liters tank (Manufacturer: GEEPEE, Country: Nigeria), ball gate valves, filters, pressure meter, male and female adopter, hose, PVC pipe, gas holder, plastic funnel, metal clip and two slots one for the entry of the biogas and another for the exits.

Plate 2: Bio-Digester

Experimental Procedure

Fresh cow dung samples were collected early in the morning from oginigba abattoir in a sterile polythene bag and transported to the laboratory within 6 hours of collection for analysis. The cow dung samples was processed by aseptically transferred ten gram (10g) of the sample into 90ml of sterile normal saline and further tenfold serial dilutions was carried out up to 10^{-6}. The sample of the dung are subjected to analysis which includes heating and incubation of the sample for specific period of time and waited to observed the growth rate of bacteria as adopted by Ogbonna and Inana (2018). Before the introduction Cow dung into the bio-digester and biogas generation started barely four day after, proper selections were performed on the cow dung to remove some non-essential materials (Plate 3). There after the slurry was introduced to the bio-digester (Plate 4)

Total heterotrophic bacteria count (THBC) and total heterotrophic fungi count (THF) were determined using spread plate method as described by Prescott *et al.* (2015), and total coliform counts and Total *Vibrio* counts was determined by spread plate technique adopted by Prescott et al. (2015). Total *Salmonella-Shigella* Counts were determined with the *Salmonella-Shigella* agar using the spread plate method as described by Prescott *et al*. (2015). Total *Staphylococci species* count was determined with the mannitol salt agar using the spread plate technique as described by Prescott *et al.* (2015). Total *Pseudomonas species* count was determined with the centrimide agar using the spread plate technique as described by Prescott *et al.* (2015). Finally, the volume of biogas produced was determined by the aid of the gas holder as described by Kossmann *et al.* (2012) and Ahmadu (2017) that the size of the gas holder i.e. the gas holder volume (Vg) depends on the relating rate of the gas generation and gas consumption.

Plate 3: Sorting of Cow Dung before being Introduced into Biodigester

RESULTS

Table 1 shows the microbial count (cfu/g) and average microbial count in Log_{10}cfu/g of the cow dung.

Table 1: Microbial Count of Cow Dung

Parameter	Counts (cfu/g)	Count (Log_{10}cfu/g)
THBC	$1.5x10^6$	6.17
THFC	$3x10^3$	3.47
Total coliform count	$1.8x10^4$	4.25
Total staphylococci count	$2.8x10^4$	4.45
Total vibrio sp count	0	0.0
Shigella and Salmonella counts	0	0.0
Total pseudomonas count	0	0.0

Key THBC = Total heterotrophic bacterial count, THFC = Total heterotrophic Fungal count
The average microbial count in Log_{10}cfu/g is shown in figure 2 (Table 1). This expresses the population of microbes present.

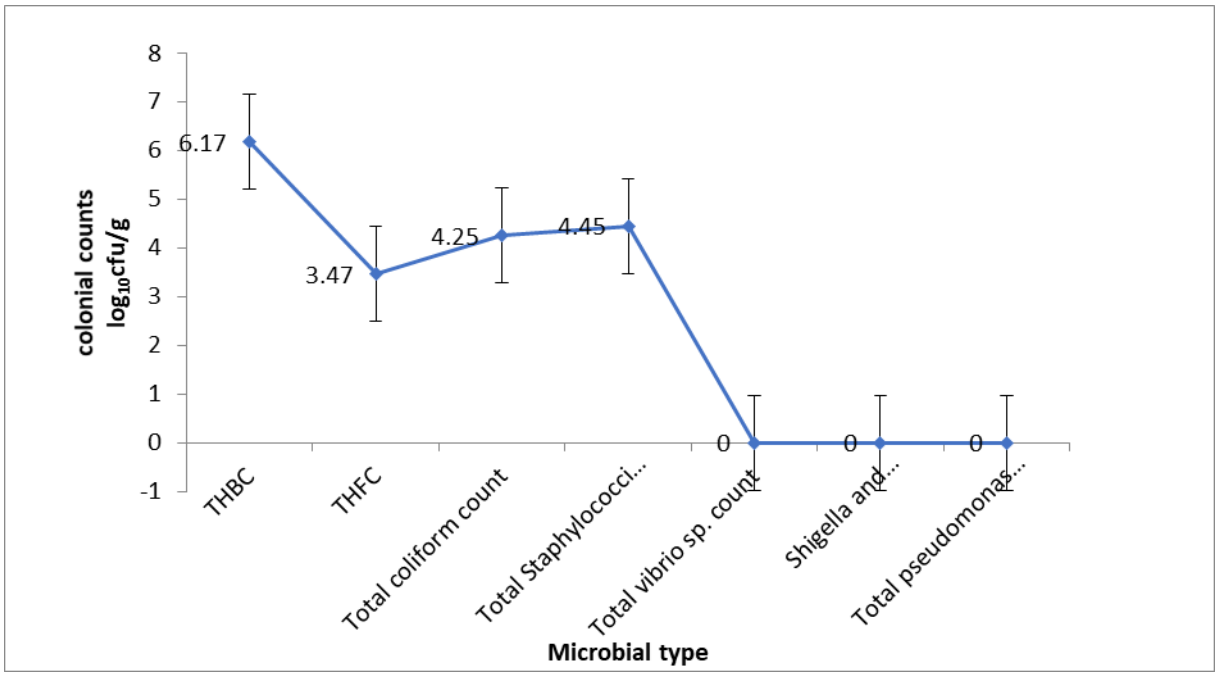

Figure 2: Average Microbial Counts from Cow Dung

The percentage composition of microorganisms in the cow dung is shown in Figure 3. This shows the percentage variation of the microorganisms

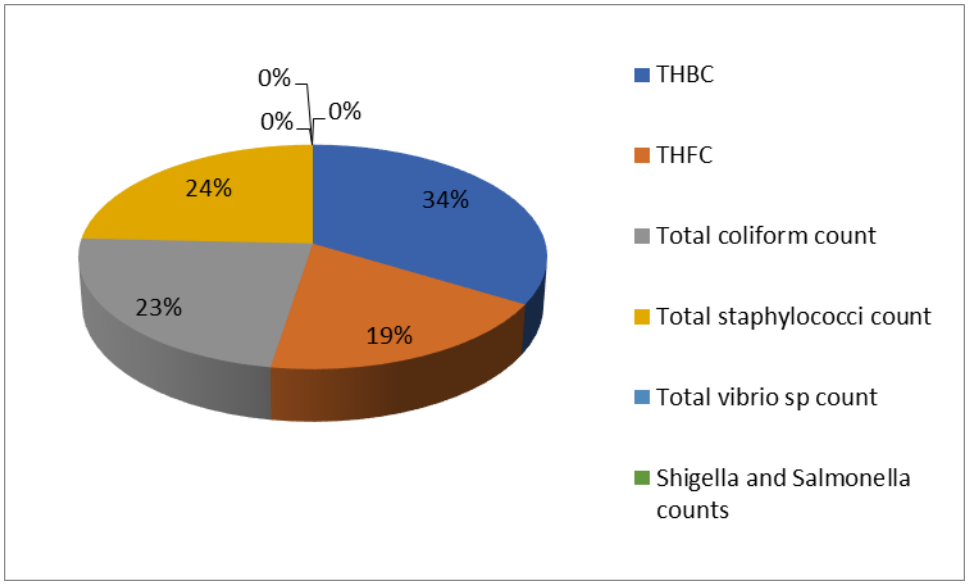

Figure 3: Percentage of Composition of Microorganisms in the Cow Dung

DISCUSSION

Presence of Microorganism in the Cow Dung

From the table 1 there were substantial amount of total coliform count, total staphylococci count, total heterotrophic bacterial count and total heterotrophic fungal count good enough to generate the required biogas needed. The microscopic laboratory analysis result showed that

the total heterotrophic bacteria count was 1.5 x 10^6, total heterotrophic fungi count was 3 x 10^3, total coliform count and total staphylococci count were also reasonably high with 1.8 x 10^4 and 2.8 x 10^4 cfu/g respectively, Also, total vibro specie count, Shigella and Salmonella counts and total pseudomonas counts were significant present in the cow dung. These aforementioned values of microorganism present in the cow dung were good for the purpose of generating enough biogas. A total of 0.2012m^3 of biogas was collected over a period of 54 days (fifty four), with the use of 148 (one hundred forty eight) kg of cow dung. The result has revealed that 1kg of cow dung can generate 0.003m^3 of biogas. For the fact that a total of 10 (Ten) bags of cow dung were collected from Oginigba abattoir which summed up to 148 (One hundred and forty eight) kilograms then were introduced into the bio-digester after thoroughly mixing it with water with the ratio of 1:3 (1kg of dung with equivalent 3kg of water) all the activities took place in a single bio-digester as support. The result showed a high level of THBC, THFC, total coliform and total Staphylococci Count, which were responsible for the generation of the said biogas. This was in agreement with the findings of Ogbonna and Inana (2014).

Average Microbial Count

The figure 2 showed a level of 6.17 colonial count of total heterotrophic bacterial count, 3.47 colonial counts of total heterotrophic fungal count; total coliform count and total Staphylococci count were 4.25 and 4.45 total coliform count Log_{10}cfu/g respectively, and total vibro specie count, Shigella and Salmonella counts and total pseudomonas counts has 0.00 Log_{10}cfu/g for each respectively. This agreed with the findings of Ogbonna and Inana (2014).

Percentage Composition of Microorganisms in the Cow Dung

An exact examination of the figure 3 showed that there were differences in percentage composition of the microbes present in the cow dung. From the results THC of 6.617 log_{10}cfu/g has 34 %, THFC of log_{10}cfu/g has 19 %, total coliform count of 4.25 log_{10}cfu/g has 23 %, and total Staphylococci count of 4.45 log_{10}cfu/g has 24 % respectively; total Vibro specie count, Shigella and Salmonella counts and total pseudomonas counts has 0.00 Log_{10}cfu/g for each has 0 % respectively. The presence of the microbes thrived well the production biogas. They are strong degrading agent that can perform efficiently even in anaerobic condition such as this study. This was in line with the findings of Ogbonna and Inana (2014).

CONCLUSIONS

Investigation into the microbial activities in biogas production using cow dung has been studied. Microbial analysis carried out on cow dung used for the research showed high level of total heterotrophic bacteria count (THBC) followed by total Staphylococci count, and total heterotrophic fungi count (THFC) and final total coliform count. There are variations in percentage of the microorganisms present in the cow dung. These variations showed that there were differences among the population of the microbes that thrive at different rate. These microbes present in the cow dung were sufficient to generate the required biogas according the capacity of the digester. Furthermore, the results revealed that the amount of cow dung required generating a specific amount of biogas depends on size of the bio-digester used and the microorganisms present. Therefore, recommend that amount of cow dung and microbes present determine the quantity and quality of biogas produced.

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