ANAEROBIC DIGESTION OF GROUNDNUT SHELL WITH COW DUNG FOR BIOGAS PRODUCTION

BY

MAMMAN, Paul
PhD/SEET/2017/972

DEPARTMENT OF CIVIL ENGINEERING,
FEDERAL UNIVERSITY OF TECHNOLOGY, MINNA

OCTOBER, 2023

ANAEROBIC DIGESTION OF GROUNDNUT SHELL WITH COW DUNG FOR BIOGAS PRODUCTION

BY

MAMMAN, Paul
PhD/SEET/2017/972

A THESIS SUBMITTED TO THE POSTGRADUATE SCHOOL, FEDERAL UNIVERSITY OF TECHNOLOGY, MINNA NIGERIA IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR THE AWARD OF DEGREE OF DOCTOR OF PHILOSOPHY (PhD) IN CIVIL ENGINEERING (WATER RESOURCES AND ENVIRONMENTAL ENGINEERING)

OCTOBER, 2023

DECLARATION

I hereby declare that this thesis titled: **Anaerobic Digestion of Groundnut Shell with Cow Dung for Biogas Production** is a collection of my original research work and it has not been presented for any other qualification anywhere. Information from other sources (published or unpublished) has been duly acknowledged.

MAMMAN, Paul
PhD/SEET/2017//972
FEDERAL UNIVERSITY OF TECHNOLOGY,
MINNA, NIGERIA.

_____ SIGNATURE/DATE

CERTIFICATION

This thesis titled: **Anaerobic Digestion of Groundnut Shell with Cow Dung for Biogas Production**" by MAMMAN, Paul (PhD/SEET/2017/972) meets the regulations governing the award of the degree of Doctor of Philosophy (PhD) of the Federal University of Technology, Minna and it is approved for its contribution to scientific knowledge and literary presentation.

Engr. Dr. M. SAIDU _____
MAJOR SUPERVISOR Signature and Date

Engr. Dr. A. O. Busari, _____
CO-SUPERVISOR Signature and Date

Engr Prof. S. Sadiku _____
CO-SUPERVISOR Signature and Date

Engr. Dr. M. ALHASSAN _____
HEAD OF DEPARTMENT Signature and Date

Engr. Prof. (Mrs.) Z. D. OSUNDE _____
DEAN, SIPET Signature and Date

Engr. Prof O. K. Abubakre _____
DEAN, POSTGRADUATE SCHOOL Signature and Date

DEDICATION

I dedicate this work to Him that is Omnipotent, Omnipresence and Omniscience for the wisdom He gave me to put this together and to my lovely late Sister: Mrs Victoria Bawusa Nmadu.

ACKNOWLEDGEMENTS

I am grateful to God Almighty who created the universe for the privilege he accorded me. Indeed He is ever faithful. Who is like unto you Lord?

First of all, I would like to thank Engr. Dr. Mohammed Saidu and supervisory team, Engr. Prof. S. Sadiku and Engr. Dr. Busari, Afis Olumide for their invaluable advice and motivation and having faith in me to undertake this PhD thesis under their profound guidance. Their prominence in giving me this opportunity and their constant attention and support throughout my thesis conferred me with a sense of responsibility.

I acquired great deal of knowledge of all the laboratory work from Mr. Yohana of the Animal Production Department of Federal University of Technology, Minna. He was like a second supervisor to me and constantly guided me throughout my thesis work. I thank him for being kind enough to guide me.

My appreciation goes to the following people: All the staff of the Department of Civil Engineering especially Head of Department Engr. Dr. M. Alhassan andFaculty Post Graduate Coordinator, Engr. Dr. Adejumo for their various supports.

I also wish to thank my wife: Mrs. Leah Hassana Mamman. She deserves my accolade because of the role she played during the writting of this report. Also, my beautiful daughters, Abigail Manasoko Mama Mamman and Alice Yebosoko Mamman for their understanding. My appreciation goes to my parents and to my siblings for their support, prayers and encouragement in the course of putting together this report. I dare not forget my relations and friends that stood by me. They indeed helped in so many ways.

I will never forget the staff of Niger State Environmental Protection Agency especially Mallam Isah, Head of Waste Management Department for helping me with the relevant information and materials to work with. Finally, my gratitude goes to my colleagues: Engr. Abraham Obakueki, Engr. Dominic Akuboh, Godwin Adama, Engr Felix, my M.D. Engr Abubakar Sale, and the staff of Niger State Roads Maintenance Agency (NIGROMA) for the advice and support they rendered.

Abstract

The demand, high costs and health implications of using energy derived from hydrocarbon compounds have necessitated the continuous search for alternative source of energy. This study investigated the production of biogas renewable energy using anaerobic digestion of groundnut shell and cow dung. Ten (10) plastic digester of size 4,000cm^3 were constructedand labeled A to J. The result of chemical analyses showed that steam explosion reduced the total solid of groundnut shell from 87.90% to 79.42% while the volatile solid was increased from75.11% to 86.32% as a result of steam explosion pre-treatment. Though the nitrogen content of GS increased after steam explosion the carbon content remained barely constant even after steam explosion. Lignocelluloses content of the substrate are: Hemicellulose before and after pre-treatment are 34.11% and 40.20% 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. Digester A being the control which contained 100% of Cow dung had 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 that contained 100% of groundnut shell also a control have pH of 7.10 and 7.20 respectively before and after digestion. There was fluctuation in the quantity of gas produced from each substrate possibly due to variation in the ratio of the substrates. The co-digestion of 25%CD-75%GS, 50%CD-50%GS, and 75%CD-25%GS had 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 had their highest biogas production around eighteen and twenty first day of retention period respectively and the least is also seen in toward the end of the digestion. Also, from the results it was observed that digester A and F that contained 100%CD each produced the highest biogas and this could be attributed to multiplication of microbial organisms within the methanogenesis stage, the digester E and J containing 100% digested GS alone produce the least biogas.The modified Gompertz equation also revealed that digesters A and F have the highest biogas production potential of 58cm^3 and 30cm^3 at a biogas production rate of 92.35cm^3 and 84.66cm^3 with a lag phase of 21days and 19days respectively. Digesters A and F contains 100% of CD which is an indication that they are a good source of catalyst to increase the volume of biogas production. So, pre-treatment of groundnut shell before digestion enhance gas production. Thus biogas production from cow dung is a good and cheap alternative source of energy. Groundnut shell (GS) and Cow dung (CD) as renewable source of energy supply have been proven to be very efficient.

TABLE OF CONTENTS

CHAPTER TWO

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

Abbreviation and Glossary

AD	Anaerobic Digestion
AOAC	Association of official Analytical chemicals
CD	Cow Dung
CDM	Clean Development Mechanism
DP	Degree of Polymerization
ECN	Energy Commision of Nigeria
GDP	Gross Domestic Product
GS	Groundnut Shell
GHE	Green House Emission
HMF	Heterocyclic Compounds Furfural
HRT	Hydraulic Retention Time
MSWM	Municipal Solid Waste Management
MSW	Municipal Solid Waste
NAPRI	National Animal Production Research Institutes
NISEPA	Niger State Environmental Protection Agency
NPC	National Population Commission
OECD	Organisation for Economic Cooperation and Development
OFMSW	Organic Fraction of Municipal Solid Waste
SERC	Sokoto Energy Research Centre
SF	Steam Explosion
SRT	Solid Retention Time
TSS	Total Suspended Solid

UNDP	United Nation Development Programme
USEPA	United State Environmental Protection Agency
UN	United Nation
USW	Urban Solid Waste
WAMASON	Waste Management Society of Nigeria
WHO	World Health Organization

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background to the Study

World population is growing rapidly, and this explosion has led to rapid consumption of oil resources and a tremendous increase in the volume of wastes generated. Globally, about 17 billion tonnes of total solid wastes are generated per year and the amount is estimated to reach 27 billion tonnes in 205045 (Karak *et al.*, 2012). Continuous emissions of carbon dioxide, methane, and other greenhouse gases from these waste streams and the burning of fossil fuels has led to a global environmental crisis. About 16% of the global population does not have access to electricity and about 38% of the population uses solid wastes (forest residue, animal manure, crop and other wastes residues) for residential heating and cooking in poorly ventilated areas, which results in environmental and health hazards, Renewable Energy Policy Network for the 21st Century (REN21), 2017. Concerns about these environmental pressures and energy insecurity have increased the need for research on energy generation from renewable sources. The undervalued and abundant solid wastes that are generated have great potential as sources of biomass for energy production if properly harnessed and could lead to reduced environmental pollution and increased renewable energy production.

Although Nigeria's natural resource (including renewable energy potentials) has been well documented and acknowledged, the contribution of renewable energy sources to

the total national energy supply and demand is currently very low or negligible Nigerian National Petroleum Corporation (NNPC), 2005. The use of fossil fuels by a large proportion of the population for public automobile transport, domestic cooking, and lighting and so on, also aggravates the existing ecological degradation. The rural populace relies heavily on biomass as a source of energy (REN21, 2017).

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). Some feedstock have been identified as very good biogas producers including most animal wastes while most plant wastes have been noted as poor biogas producers because of the presence of lignin, plant wax, cellulose and hemicelluloses in the plant structures. Therefore when pre-treated or subjected to either co-digestion with the better producing wastes or chemically treated, these plant wastes have been observed to increase their biogas production (Adeyanju, 2008). These have been attributed to the synergy in operation between the wastes being co-digested.

The use of biogas is capable of providing a special impetus in both rural and urban areas. Biogas plant can be built by using materials which are locally available in most developing countries like Nigeria (Esan, 2008). Biogas is a renewable, alternative and sustainable form of energy (Godi *et al.*, 2013). Not only does biogas technology help to produce an alternative energy source, but it also helps in maintaining the

environment and improving health conditions. Ignorance about this technology has made majority of people in the developing countries mainly depend on solid fuel like wood, to meet their cooking and light needs (Babatola, 2008). The energy in plant vegetation, animals, industrial and domestic waste matter can be released in terms of a useful gas when fermented anaerobically.

1.2 Statement of the Research Problem

Several thousand tons of agricultural wastes such as groundnut shell or peanut are generated in Nigeria annually most of which end up as pollutants in the environment without being put to any meaningful usage. Despite the huge availability of this biomass in their various locations of production, they mostly end up as solid wastes in the environment as little or no usage has been sought for them over the years. Even when some of the biomass has been experimented on for biofuel production, the various arrays of microorganisms involved in their biodegradation are yet to be documented in biofuel literature.

Also, there is an urgent need for alternative energy sources as a result of the dwindling energy resources which has become a global concern. This has made it imperative to search for new sources of domestic energy. The quest for wood as a source of domestic energy has led to deforestation and erosion in the southern parts and near desertification in the northern parts of the country (Ilochi and Nwachukwu, 1989). Raw materials for biogas production cover a wide range of feedstock including animal wastes, household wastes, crop residues, sewage sludge, food waste, and

wastewater. Manure component (carbohydrates, proteins, and lipids) carbon is ultimately transformed into methane (CH_4) and carbon dioxide (CO_2) (Masse *et al.*, 2011), and its emission contribute to Green House Emission (GHE).

It is widely accepted that the breaking of the lignocellulosic bonds in biomass will enhance the digestibility and accessibility of the energy potentials (cellulose and hemicellulose). Earlier work has been conducted using different pretreatment methods on biomass. However, perusal of literature shows that little or no work has been done using the Auto clave technique on the pretreatment of lignocellulosic biomass. Auto clave has inherent advantages of uniform volumetric heat transfer, deep heat penetration within the samples, and can easily scale up. Therefore, the present study will be conducted with the intention of exploring the applicability of Auto clave heating for the pretreatment technique in the deconstruction and disruption of lignocellulosic biomass, and to alter its physical and chemical structures.

Renewable accounted for about 62% of the net additions to global power generating capacity in 2016, and the vast majority of renewable energy for heating was supplied by biomass, with smaller contributions from solar thermal and geothermal energy (REN21, 2017). Biogas production is an appropriate technology needed in Nigeria to ease the nation's energy and environmental challenges. To this development, the conversion of waste to energy through anaerobic digestion is a promising option.

1.3 Aim and Objective of the Study

The aim of the study is to produce biogas through anaerobic digestion of groundnut shell with cow dung. The objectives are:

i. To determine the influences of operating parameters in anaerobic production.

ii. To determine the effect of lignocellulose reduction using steam explosion and biological pretreatment method. .

iii To compare the laboratory biogas rate and the kinetic of the biogas production using modified Gompertz equation.

1.4 Justification for the Study

Kerosene and other oil based sources of fuel are scarce and costly for common house hold users in Nigeria. Furthermore, frequent alarming hike in prices of imported oil and chemical fertilizer have serious economic threat to the rural poor. In this context, to reach the self-sufficiency in energy and fertilizer and to minimize the pressure on traditional biomass fuel, biogas technology has been the best alternative energy solution, which could be achieved through the active mobilization and economic utilization of local indigenous resources available in the country.

A study that assessed Nigeria's biogas potentials (minimum value) from solid waste and livestock excreta revealed that in 1999, Nigeria's biogas potential represents a total of 1.382×10^9 m^3 of biogas/year or an annual equivalent of 4.81 million barrels of crude oil (Ojolo *et al.,* 2007). In addition, 20kg of municipal solid waste (MSW) per

capita has been estimated to be generated in the country annually (Mshandete and Parawira, 2009). By the 2005 census figure of about 140.4 million inhabitants, the total generated MSW will be at least 2.81 million tonnes every year. With increasing urbanization and industrialization, the annual MSW generated will continue to increase. Biogas production therefore may be a profitable means of reducing or even eliminating the menace and nuisance of urban wastes in many cities in Nigeria.

With the rapidly increasing waste generation threatening to prevent humans from carrying out their activities for lack of space, the society is therefore faced with the choice to either allow this biomass waste to continue polluting the environment; methane and carbon dioxide production, to continue to increase global warming or boldly take the initiative of converting the biomass into alternative energy (Igboro, 2011). The study therefore explores means of converting these and other organic wastes to energy. While converting wastes into energy is especially appealing conceptually, there is still much to be done before the technology becomes commonplace. Also, the research is necessary in order to optimize the technologies, build confidence in their effectiveness, and prepare them for the market (Igboro, 2011). This will require significant efforts in outreach. Policy makers, farmers, engineers, the business community and the rural community need to know about issues of sustainable development which confront us and biogas utilization would just be the panacea for our energy problem.

1.5 Scope of the Study

The scope in this research is limited to the investigation of the biogas production using groundnut shell with cow manure. The digestion was carried out anaerobically using digesters that were fabricated locally. The experimental aspect of the research was carried out in Animal Production Department and Tetfund Laboratory of Federal University of Technology, Minna.

1.6 Thesis Structure

This Thesis consists of five (5) chapters as described in the paragraphs below.

Chapter one is the introduction. It covers general introduction to the subject of the research and includes general background of study, statement of the research problem, aim and objectives of the study, justification of the study, scope of the study and thesis structure.

Chapter two is the literature review. It covers the historical perspectives of waste generation, world population, management of waste and the issues arising from improper management. A review of anaerobic digestion, co-digestion of different substrates, biogas production in Nigeria and challenges, characteristics of biogas feedstock and different forms of pretreatment of lignocellulose biomass.

Chapter three is the methodology for the research. It clarifies the process and methods which were used during the fabrication of the digesters, pretreatment process, lignocellulose content determination, and preparation of the substrate for oyster

mushroom cultivation, anaerobic digester set-up, fermentation procedures for the biological and the physical pre-treatment, sample and sampling technique, data collection, materials, instrumentation and techniques used to achieving the objectives of the research.

Chapter four is results, presentation of data and discussion. It presents the results obtained from the laboratory investigation of the substrates characterisation, calculation, comparing of results, general analysis, discussion of the results obtained and findings made based on observation and laboratory investigation of the substrates.

Chapter five is conclusion and recommendation. It states the conclusions and recommendations arising from the research.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Waste Materials and Sustainability

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" result 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. 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.*, 2014).

2.2 Potential of Solid Waste

Solid wastes are potential biomass sources because they are readily available and their conversion to energy through biological processes is feasible with low capital investment. Biomass comes from a range of sources, as shown in Plate I which can be classified according to the activities generating these wastes or the locations where these wastes are generated (Kofoworola, 2007). Agricultural wastes account for the

largest potential feedstock and wide varieties of these wastes can be used as sources of biomass energy. The most common sources (animal manure, forest and crop residues) are discussed in this chapter.

Plate I: Solid wastes; potential sources of biomass (Uwaegbulam, 2017)

2.3 Municipal Waste Generation and the World Population

The United Nations (UN) in 2007 projected that the world population will increase by 2.5billion between 2007 and 2050, that is, from 6.7 billion in 2007 to 9.2 billion in 2050 (UN, 2007). This increase is equivalent to the population of the world in 1950 and will be absorbed by the less developed parts of the world, whose population is likely to rise from 5.4 billion in 2007 to 7.9 billion in 2050, whilst the developed nations are expected to remain marginally constant at 1.2 billion people (UN, 2007).

As a result of the continuous growth of the world population as well as improvement in the standard of living of people and industrial advancement, the amount of municipal solid waste (MSW) generated is on a constant rise. In addition, it is reported that in 2012 each person within the 28 EU member states generated an average of 492 kg of MSW (Eurostat, 2014). In the same report, each person in the UK generated an average of 472kg of MSW (Eurostat, 2014). Similarly, Department of Environment, Food and Rural Affairs (DEFRA) in 2013 reported that the UK generated 31.1 million tonnes of MSW in 2012 (Themelis and Verma, 2013). It is established that MSW is a global problem, but the improper waste management in developing countries increases the susceptibility to environmental and health hazards.

2.4 Different Types of Waste Management Concepts

Before the advent of industrial revolution, the nature of waste produced was mainly biodegradable, such as vegetable, human waste and ashes from the incineration of other waste materials (Isa *et al.*, 2014). The management of waste was not complex due to the utilisation as fertiliser or soil conditioner on farmland (Sangodoyin, 2017). The industrial revolution in the 19th century created a significant increase in the global economic activities, resulting in mass migration of people to the industrial cities from the rural areas. The increase in industrial activities and the constant growth in human population, especially in the cities led to a significant increase in the quantity of solid waste generation (Isa *et al.*, 2014). As a result, waste management becomes a critical issue to humanity. The different techniques used to minimise the

challenges of waste management include landfill, incineration, recycling and biological reprocessing.

2.4.1 Sanitary landfill

Historically landfills have been the most common method of organized waste disposal and remain so in many places around the world. Sanitary landfills involve burying and managing wastes within a controlled environment. Ogwueleka (2009) reported that a sanitary landfill has controls in place to collect gases generated, leachate management systems and other mitigations in place to control the impact on the environment and society. Sanitary landfills are an environmentally accepted method of waste disposal but are capital intensive at roughly 3-8 times more expensive than open dumping (Ogwueleka, 2009; Sridhar and Hammed, 2014).

Sanitary landfills were introduced in Lagos and Onitsha two decades ago, but currently the landfills are not operating (Ogwueleka, 2009; Nwosu *et al*., 2016).They require much greater initial investment and hence higher operating costs than uncontrolled or open dumps.

2.4.2 Incineration

Incineration is the combustion of wastes at high temperatures which converts waste into ash, flue gas, and heat (Ogwueleka, 2009). Knox (2005) emphasized that flue gases must be cleaned of gaseous and particulate pollutants before they are dispersed into the atmosphere. Two of the primary advantages of incineration are that waste

volumes are reduced by an estimated 80-95% and the need for landfill space is greatly reduced (Greentumble, 2015). For urban areas, this can be especially important, as urban land is often at a premium. In Nigeria incineration is not widely practiced (Obasioha, 2015; Ogwueleka, 2009) except in hospitals where medical waste is sometimes incinerated at a small scale but without energy recovery. Phillips and Williams(2022) reported that 3 modern incinerators were built in Lagos with a European Economic grant at a cost of $30 million (£23 million) but they were never used.

2.4.3 Recycling

Although recycling exists in Nigeria (Kofoworola, 2007), it has not received the attention of government and the waste management authorities, either in the past or at present. Therefore whilst recycling is common in most Nigerian cities (Otitoju, 2014) it is normally implemented by the informal sector (uncontrolled recycling) rather than government agencies (controlled recycling). Recycling can bring a range of benefits including economic growth, litter control, prolonging the lifespan of landfill, and conserving resources and energy (Ezeah *et al*., 2013; Konya *et al*., 2013; Oumarou *et al*., 2012).

In Nigeria whilst there is an emerging awareness of the need to recycle, as mentioned above the activity is driven by entrepreneurs who seem to be light-years ahead of government (Umaru, 2010). Recycling of solid waste in Nigeria is mainly uncontrolled and revolves around the activities of informal workers, while controlled

recycling is rarely practiced since government is not involved, and there are no formal recycling collection schemes. In local parlance (particularly in northern and central Nigeria) they are referred to as *Yan Bola* (Guardians of the garbage), *Yan Panteka* (Motor scrap cannibals), *Yan Gwangwani* (Metal scrap collectors), *Yan Makera* (Metal fabricators/smiths) or *Yan Tinka* (Tin boys). Informal workers recover items of value from household garbage bins, construction sites, garages, markets and factories (Kofoworola, 2007; Umaru, 2010). In addition many people survive in Nigeria by scavenging open dumpsites for materials that could be sold (Ogwueleka, 2009).

2.4.4 Anaerobic digestion

Anaerobic digestion of organic waste is an efficient process to produce biogas with high energy value. In recent times, this technology has attracted so much attention due to the added advantage of minimizing greenhouse gas emissions. The produced biogas serving as an alternative or supplement for fossil fuels products results in reduced emissions (Iweka*et al.*, 2021). In addition, anaerobic digestion is considered an economical and effective technique due to the use of waste as a substrate. Organic waste such as animal waste, sewage sludge, industrial organic residue, and agricultural waste are usually employed with animal waste topping the list. Among animal waste, cow dung is mostly used as a substrate due to its universal abundance and availability. Not only does cow dung serve as a substrate, but it is also used for thermal insulation and as a fertilizer for soil conditioning.

In terms of biogas production, previous studies have shown its effectiveness as a substrate. For instance, the effectiveness of cow dung for biogas production was carried out by (Mukumba and Makaka, 2015). The study revealed that cow dung produced a biogas yield with a 50% average methane composition. Interestingly, the study noted that the use of cow dung resulted in an early retention time with a high biogas yield. On a similar note, (Obileke *et al.*, 2019) compared the performance of an aboveground and underground fixed dome digester using cow dung as substrate installed in the Eastern Cape Province. The findings showed that the optimum methane yield was 50% and 60% for aboveground and underground digester systems respectively. This is a further indication that the use of cow dung as substrate results in higher methane yield. Although studies have noted that the type of organic substrate used, play a significant role in the composition of biogas produced.

In spite of its early start in Africa, biogas technology on the continent is still at an embryonic stage. Specifically in Nigeria, the status of biogas technology remains very poor, with no record of any existing commercial size plants that could contribute electricity to the national grid. The earliest record of biogas technology in Nigeria was in the 1980s, when a simple biogas plant that could produce 425 litres of biogas per day was built at Usman Danfodiyo University, Sokoto (Akinbomi *et al*., 2000; Sambo, 2010). Since then about 21 small scale pilot digesters with a capacity of between 10 m^3 to 20 m^3 have been set up in different parts of the country (Chima *et al*., 2013) reports that the national government through Universities and research centres are carrying out more research on anaerobic digestion, with a view to fully

embrace and establish this technology. However, to date, biogas technology in Nigeria has stagnated at the institutional research and pilot stages rather than being rolled out commercially. Okoro-Shekwaga and Horan (2015) cited a range of barriers including ignorance, lack of a coordinating framework, and lack of political will from government. Moreover research at universities is frequently considered as being too academic and as such is rarely implemented in real life. On the other hand biogas technology is spreading across other African countries with Kenya taking the lead and further examples in Ethiopia, Tanzania, Uganda, Rwanda and Burkina Faso (Stichting Nederlandse Vrijwilligers, (SNV) 2017).

2.5 Motivation of Anaerobic Digestion for Organic Waste Management

Appropriate waste management practice is crucial for any sustainable society. It prevents air, soil and water pollution as well as improves public health, decreases greenhouse gas emission and preserves natural resources. The AD system, which is a type of biological reprocessing, is the ideal waste management technique (Chima *et al.* (2013). AD is the process by which bacteria breakdown organic material to produce biogas (renewable energy source) and digestate (biofertiliser) in the absence of oxygen. It involves not only the collection and safe disposal of organic waste but also sustainable management of organic material to create a source of renewable energy as well as provide economic, health and environmental benefits. According to Chima *et al.*, 2013, AD reduces greenhouse gas emission more than any other waste management systems. It is an effective method of recovering energy and nutrients

from organic material. The following are some of the benefits of AD system (DEFRA, 2013).

i. Contribution towards mitigation of climate change and other environmental targets.
ii. Treatment of biodegradable wastes to generate biogas, a renewable energy that can be utilised to produce electricity and heat from combined heat and power (CHP) or for vehicle fuel.
iii. Diversion of organic wastes from landfills and capturing of methane emission from organic wastes.
iv. Provision of organic fertiliser and soil conditioner for agriculture and land use; and
v. A source of revenue generation to farmers and other practitioners, as excess energy and digestate are sold, which adds to the nation's gross domestic product (GDP).

2.6 Anaerobic Digestion

Anaerobic digestion (AD) occurs when organic matter decays in an oxygen-free or low oxygen environment. Anaerobic methane recovery occurs in bio-digesters, where organic matter is digested, and produces a fuel called biogas. (Monnet, 2003) This process conserves nutrients and reduces pathogens in organic matter.

There are four key biological and chemical stages in anaerobic digestion these include the following:

a. Hydrolysis

In most cases, biomass is made up of large organic polymers. For the bacteria in anaerobic digesters to access the energy potential of the material, these chains must first be broken down into smaller constituent parts. These constituent parts or monomers such as sugars are readily available to other bacteria. The process of breaking these chains and dissolving the smaller molecules into solutions is called hydrolysis. Therefore hydrolysis of these high molecular polymeric components is the necessary first step in AD (Sleat and Mah, 2006). Through hydrolysis, the complex organic molecules are broken down into simple sugars, amino acids and fatty acids.

$$C_6H_{10}O_4 + 2H_2O \longrightarrow C_6H_{12}O_6 + 2H_2 \qquad (2.1)$$

b. Acidogenesis

Acetate and hydrogen produced in the first stages can be used directly by methanogens. Other molecules such as Volatile Fatty Acids (VFA's) with chain length greater than that of acetate must first be catabolised into compounds that can be directly used by methanogens (Boon and Mah, 2006). The biological process of acidogenesis results in further breakdown of the remaining components by acidogenic (fermentative) bacteria. Here, VFA's are created along with ammonia, carbon dioxide and hydrogen sulfide as well as other by-products. The process of acidogenesis is similar to the souring of milk.

$$C_6H_{12}O_6 + 2H_2 \longrightarrow 2CH_3CH_2COOH + 2\,H_2O \qquad (2.2)$$

c. Acetogenesis

The third stage of AD is acetogenesis. Here, simple molecules created through the acidogenesis phase are further digested by acetogens to produce largely acetic acid as well as carbon dioxide and hydrogen.

$$C_6H_{12}O_6 + 2H_2O \leftrightarrow 2CH_3COOH + 2CO_2 + 4H_2 \quad (2.3)$$

d. Methanogenesis

The terminal stage of anaerobic digestion is the biological process of methanogenesis. Here, methanogens use the intermediate products of the proceeding stages and convert them into methane, carbon dioxide and water. These components make up the majority of the biogas emitted from the system as shown in Figure 2.1. The remaining indigestible material which the microbes cannot use and any dead bacterial remains constitute the digestate. A simplified generic equation for the overall processes outlined above is as follows:

$$C_6H_{12}O_6 \longrightarrow 3CO_2 + 3CH_4 \quad (2.4)$$

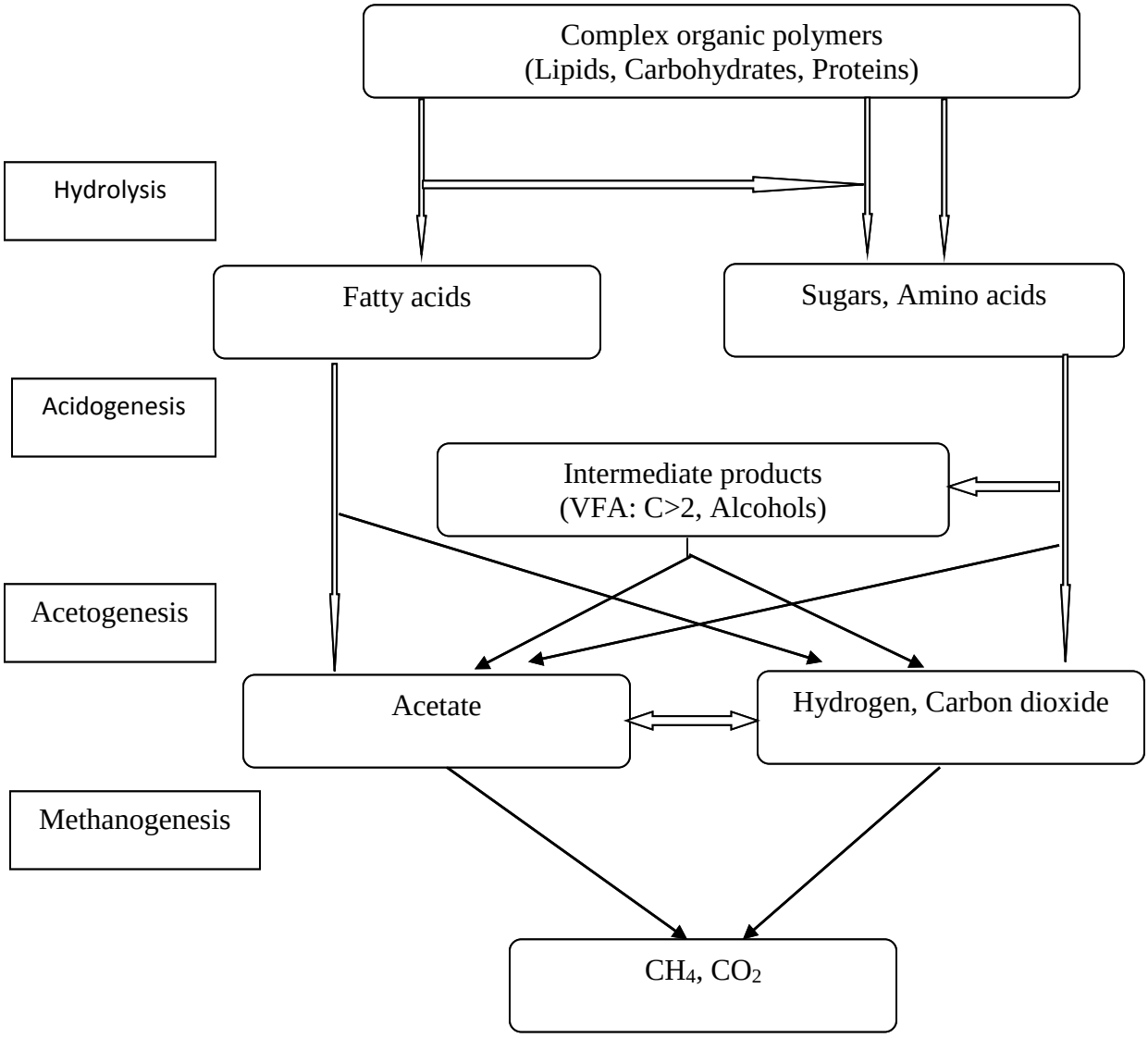


Figure 2.1: Simplification of anaerobic digestion process (adapted from Gujer and Zehnder, 1983)

2.7 Co-digestion of Substrates

Co-digestion is a process in AD by which two or more substrates are homogenously blended and simultaneously digested. The AD was initially applied to a single substrate, single purpose treatment plant. However, studies have shown that AD process is more stable and achieves more potential benefits as a result of co-digestion

(Mata-Alvarez *et al*., 2003). Co-digestion enhances biogas production due to the supply of the required nutrients by the co-substrates involved (Mata-Alvarez *et al*., 2003). This achievement in nutrient balance leads to better digester performance and improved biogas production. Mata-Alvarez *et al* (2003) reported that co-digestion of solid slaughterhouse waste, fruit, vegetable and manure improved the buffering capacity of digester – the ability of digester to react to changes in pH. In other words, the buffering capacity of the digester is the measure of the amount of alkalinity present in the digester.

Different types of manure present variation in organic composition and dry matter content (1.5–30.0 %), which affects the biogas produced (Mano Esteves *et al.*, 2019). Co-digestion is often used for the very reason that the optimal carbon-nitrogen ratio on biogas production is in the rage of 20:1 to 30:1, but in general, manure has very low carbon ratio and it is important to mix it with other substrates that are carbon-rich to increase the biogas yield (Conti *et al.*, 2019).

In addition, co-digestion is found to maintain the appropriate balance of C/N ratio in AD and to reduce the concentration of nitrogen. For instance, the mixture of substrate with low nitrogen content and lipid is found to enhance biogas production as well as reduce the risk of acid accumulation and high concentration of ammonia that cause digester failure (Khalid *et al*., 2011). In another study, co-digestion of the substrate that contains high moisture (liquid manure or sewage sludge) and substrate of poor moisture content is found to enhance the total solids (TS) content of digester.

Similarly, co-digestion involving substrate with high moisture content is found to dilute toxic compounds, thereby improving digester performance and biogas production (Khalid *et al*., 2011; Braun, 2007).

Other benefits derived from co-digestion include increase in biodegradation of organic materials, stability and digestion rate improvement (Braun, 2007) as well as higher mass conversion, resulting in lower weight and volume of digestate (effluent). However, co-digestion has some disadvantages such as, an increase in digester effluent COD (chemical oxygen demand), need for additional pre-treatment, higher mixing requirement and higher energy requirement (Braun, 2007). Co digestion is a well-established practice in Europe, especially in Germany and Stichting Nederlandse Vrijwilligers (SVN) (2010). The first co-digestion plant built in the UK is for the digestion of animal manure and food waste.

2.8 Biogas

Biogas is an environment friendly, clean, cheap and versatile gaseous fuel. It refers to a gas produced by the biological breakdown of organic matter in the absence of oxygen (anaerobic digestion). The organic waste materials include animal wastes, agricultural wastes, municipal wastes, industrial wastes, domestic wastes, human wastes, solid organic wastes (Abubakar, 1990). The gas is composed of mainly methane (50 – 70%), carbondioxide (20 – 40%) and traces of other gases such as nitrogen, hydrogen, ammonia, hydrogen sulphide, and water vapour (Edelmann *et al.*, 1999). The gas is odourless and flammable and yields about 1,000 British Thermal

Units (BTU) (252 kilocalories) of heat energy per cubic foot (0.028 cubic meters) when burned (De Bruyn and Hilborn, 2007). The other type of biogas is wood gas which is created by gasification of wood or other biomass. This type of biogas is comprised primarily of nitrogen, hydrogen and carbon monoxide with traces of methane (Anonymous, 2011).

The gases methane, hydrogen and carbon monoxide can be combusted or oxidized with oxygen. This energy release allows biogas to be used as a fuel. Biogas can be used as a fuel for heating and cooking purposes. It can also be used in modern waste management facilities where it can be used to run any type of heat engine to generate either mechanical or electrical power. Biogas is a renewable fuel, so it qualifies for renewable energy subsidies in some parts of the world (Anonymous, 2011). Table 2.1 shows a typical composition of biogas.

Table 2.1: Average Composition of biogas

Chemical	Compound	Percentage (%)
Methane	CH_4	50 – 75
Carbon dioxide	CO_2	25 – 50
Nitrogen	N_2	0 – 10
Hydrogen	H_2	0 – 1
Hydrogen Sulphide	H_2S	0 – 3
Oxygen	O_2	0 – 0

Source: Anonymous (Anonymous, 2011).

Biogas is somewhat lighter than air and has an ignition temperature of approximately 700°C (diesel oil 350°C; petrol and propane about 500°C). The temperature of the flame is 870°C. However the main constituents of biogas are CH_4 and CO_2 gases.

Biogas burns very well when the methane content is more than 50%. If the methane content is considerably below 50%, biogas is no longer combustible. Therefore, biogas can be used as a substitute for kerosene, charcoal, and firewood for cooking and lighting. This saves time and money and above all it conserves the natural resources such as cutting trees to get firewood (Igboro, 2011).

2.9 Brief History of Anaerobic Digestion

Scientific interest in the manufacturing of gas produced by the natural decomposition of organic matter was first reported in the 17th Century by Robert Boyle and Stephen Hale, who noted that flammable gas was released by disturbing the sediment of streams and lakes (Fergusen *et al.*, 2014). In 1808, Sir Humphrey Davy determined that methane was present in the gases produced by cattle manure. The first anaerobic digester was built by a leper colony in Bombay, India, in 1895. In 1895, the technology was developed in Exeter, England, where a septic tank was used to generate gas for the sewer gas destructor lamp, a type of gas lighting.

Also in England, in 1904, the first dual purpose tank for both sedimentation and sludge treatment was installed in Hampton. In 1907, in Germany, a patent was issued for the Inhof tank, an early form of digester. Through scientific research, AD gained academic recognition in the 1930's. This research led to the discovery of anaerobic bacteria, the microorganisms that facilitate the process. Further research was carried out to investigate the conditions under which methanogenic bacteria were able to grow and reproduce (Humenik and Hanna, 2007). This work was developed during

World War II during which, in both Germany and France, there was an increase in the application of AD.

2.10 Biogas in Nigeria

Nigeria, with a growing population of about 186million, has the largest economy among the nations in Africa with a gross domestic product (GDP) of about $405 billion, according to World Bank 2016, yet Nigeria continues to face energy and environmental challenges. About 96% of the population of Nigeria is connected to the national grid, but only 18% of the functioning connections have a reliable supply of electricity, which is a major challenge that depletes the energy and enterprises necessary for the country's development. Also, continuous burning of fossil fuels and solid wastes has resulted in 94% of the population of Nigeria being exposed to air pollution levels (measured in PM2.5) that exceed the WHO guidelines and air pollution damage costs of about 1% post of gross national income (World Bank, 2015). Water and soil pollution is also a challenge due to improper management of human sewage and the tremendous amounts of solid wastes generated. As water, soil, and air pollution as well as energy poverty pose great challenges to human health and the environmental and economic development of the country, the need for renewable energy cannot be overemphasised.

Anaerobic digestion of waste is contributing significantly to solving energy, environmental and agricultural-related problems. This has encouraged the development of biogas technology globally as well as the need to study its economic

viability (Kozlowski *et al.*, 2019). Bhatt and Tao, (2020) mentioned that current and future research in renewable energy has contributed to the rapid increase in investment and implementation of clean energy technologies around the world. According to Ngumah (2013), Nigeria generates about 542.5 million tonnes of total wastes per annum (livestock wastes, human excreta, crop residues, and municipal solid wastes). This tremendous amount of wastes has the potential to produce an estimated 25.53 billion m^3 of biogas and 88.19 million tonnes of bio-fertilisers annually. The biogas produced can be used for heating, cooking, transportation, and generation of electricity if properly harnessed, and the residue remaining after biogas production is suitable for improving agricultural development in the country. Biogas can augment the conventional energy sources in the country, thereby improving the quantity and quality of the energy supply while also reducing environmental pollution.

2.11 Biogas Challenges and Pioneered Projects in Nigeria

The fixed-dome reactor is one of the commonly used biogas reactors in Nigeria because of its long lifespan, but the technology is expensive, labour intensive, and requires skilled supervision. The lack of government commitment and poor continuity of previous biogas programme initiatives through successive governments is a major factor limiting the advancement of this technology (Akinbomi *et al.*, 2000). An additional challenge is corruption, which increases the investment costs for biogas implementation and thereby reduces the rate of return for the investment (Taherzadeh and Rajendran 2014).

In spite of these challenges, there are some existing biogas plants in Nigeria: less than 20 pilot projects, including the United Nations Development Programme (UNDP) project in Kano state, have been established (ECN and UNDP, 2005). The Cows-to-Kilowatts Project, located in Ibadan, the capital of Oyo State, was begun in May 2008 (Dahlquist, 2013) with collaboration among the Global Network for Environment and Economic Development Research, Nigeria (NGO), the Biogas Technology Research Centre, KMUTT, Thonburi, Thailand (research institute), the Centre for Youth, Family and the Law, Nigeria (community-based organisation), and the Sustainable Ibadan Project, Nigeria (UN-HABITAT Programme). The biogas plant, shown in Figure 2.2, employs an anaerobic fixed-film reactor with a volume of 3000m^3 for treatment of abattoir waste to produce biogas and organic fertiliser. The Bodija market in Ibadan slaughters about 1000 cows per day, and the plant is capable of producing about 1500 m^3 of biogas (900 m^3 of methane) Cows to Kilowatts, 2005). The plant is also capable of producing about 1500 litres of bio-fertiliser per day for farmers.

Figure 2.2: Biogas plant in Nigeria for the treatment of abattoir wastes from Bodija market, Ibadan. Source (Cows to Kilowatts, 2005)

Another fixed-dome bio-digester of 20 m^3 which was built by the Energy Commission of Nigeria (ECN) in 1998 is fed with cow dung. The reactor is located at the Mayflower Secondary School, Ikenne Ogun state, Nigeria (Dahlquist, 2013). The plant produces gas for cooking and bio-fertiliser for farmers. Additionally, the National Centre for Energy Research and Development, University of Nigeria Nsukka (NCERD/UNN) built a biogas plant of 10 m^3 for Women at Achara, Nsukka, Enugu state. The biogas plant is fed with domestic animal wastes, cassava peels and wastes from the milling of cowpea, and bambara nuts from a food processing plant (Dioha and Nfor, 2017). The Sokoto Energy Research Centre (SERC) also constructed a 30 m^3 biogas reactor at the National Animal Production Research Institute (NAPRI) in Zaria. The reactor is fed with human excreta, and the biogas produced is used for cooking at the Zaria prison.

2.12 Benefits of Biogas Utilization

When biogas is utilized, many advantages arise. In North America for example, utilization of biogas would generate enough electricity to meet up to three percent of the continent's electricity expenditure. In addition, biogas could potentially help reduce global climate change. Normally, manure that is left to decompose releases two main gases that causes global climate change; nitrous oxide (NO_2) and methane (CH_4). NO_2 warms the atmosphere 310times more than carbon dioxide and methane 21times more than carbon dioxide. By converting cow manure into methane biogas via AD, the millions of cows in Nigeria would be able to produce one hundred billion kilowatt hours of electricity enough to power millions of homes across the country. In

fact, one cow can produce enough manure in one day to generate three kilowatt hours of electricity; only 2.4 kilowatt hours of electricity are needed to power a single one hundred watt light bulb for one day (SECOT, 2009). The 30million rural households in China that have biogas digesters enjoy the benefits of; saving fossil fuels, saving time collecting firewood, protecting forests, using crop residues for animal fodder instead of fuel, saving money, saving cooking time, improving hygienic conditions, producing high quality fertilizer, enabling local mechanization and electricity production, improving the rural standard of living and reducing air and water pollution.

Biogas produced from anaerobic digestion often has high amounts of sulfur, which is what causes an uncomfortable smell. This is only very problematic if the intent is to use the biogas in a fuel cell, because the sulfur will poison the fuel cell. There are sulfur scrubbers available to remove the sulfur if the intent is to use the biogas in a fuel cell, but this adds significantly to cost. If the gas is just to be burned as cooking fuel or in a generator, then sulfur production is not necessarily a problem.

2.13 The Environmental Impact of Biogas

Biogas is environmentally friendly (especially when CO_2 is removed from its composition) tool for reducing greenhouse gas emissions. It is a very effective means of addressing issues like indoor air pollution, deforestation and reducing greenhouse gas emission through manure and solid waste as feedstock for biogas production (Arthur *et al.*, 2013). Indoor air pollution and deforestation mainly related to

developing countries, where biomass resources such as firewood are used for cooking and lightning. The raw biogas which is normally used in Europe for heating, cooling and electricity generation comprises 60% methane and 40% CO_2. H_2S and NH_3 are also available in the tanks but their uses are minimal (Berglund and Börjesson, 2006).

2.14 Economic benefits of Biogas

The economic feasibility of biogas plants can be investigated with different factors, such as output (biogas) substitution of fuel, slurry (the use of residues and nutrients ratio), and health benefit and pollution abatement clean development mechanism (CDM). There are several studies which explain the economic benefit of biogas with fossil fuel, digestate as organic fertilizer and to achieve carbon credits under CDM through biogas plants. Biogas is a clean renewable resource for energy production. Renewable energy ensures environmental sustainability, economic profitability through a cheap source of energy and the creation of job opportunities for people all over the world (Isci and Demirer, 2007). That is the reason why renewable energy has become popular in recent times. As reported, the use of biogas for vehicles has gained in popularity not only because of the high price of alternative fuels but also due to the great concern for the impact on global warming as a result of burning fossil fuels (Richards *et al.*, 2010).

Assessing economic impact of biogas system is very complex in developing countries where the biomass fuels are not well marketed. Nevertheless, there is one main driver that reduces the pressure on woodland through which we can make economic

assessment (Bond and Templeton, 2011). In conclusion, biogas is an economical source of energy for countries such as Nigeria where power outages present a big challenge for government. Despite the availability of enormous biomass resources, the absence of good understanding and the use of key concepts of cost estimation may affect project profitability and technical solutions for the commercialization of biogas plants in Nigeria.

2.15 Digestate

Anaerobic digestion can be seen as a method to treat the organic wastes but, in order to extract the maximum recovery value from these wastes, the digester should have a useful purpose and benefit should be derived from its production (Monnet, 2003). Anaerobic digestion draws up carbon, hydrogen and oxygen from the feedstock. Meanwhile, essential plant nutrients (nitrogen (N), phosphorus (P) and potassium (K)) remain largely in the digestate. Its main advantage is that it has a high nutrient content. The availability of nutrients is higher in digestate than in untreated organic waste. For instance, digestate has 25% more accessible NH_4-N (inorganic nitrogen) and a higher pH value than untreated liquid manure (Monnet, 2003).

More so, it reduces the odour nuisance by about 80%. The digestate leaving the chamber is a thick sludge with a moisture content of about 80%, close to the consistency of a milk shake. It is obvious that transporting this would be uneconomic. Therefore, digestate is normally dewatered. The solid is reduced to a liquid content of about 50% - 70% and the remaining water can be collected (Igboro, 2011). Mata-Alvarez *et al.* (2003) noted that the quality and composition of the dewatered solid

depend on the feedstock and the digestion process. Additionally, even if digestion were allowed to proceed for long time periods, a maximum of only about 70% of the total organics are available for degradation.

2.16 Substrates

The amount and characteristics of organic materials available for digestion vary widely. In rural areas, the digestible material will depend upon the climate, the type of agriculture practiced, the animals used and their degree of confinement and the methods of collecting wastes (Braun, 2007).

2.16.1 Cow dung

Cow dung (Plate II) 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. Fresh cow dung is usually collected and carried to the system in buckets or baskets. Upon arrival it is hand-mixed with about an equal amount of water before being fed into the digester (Maramba, 1978). Liquid cow 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 (Ibrahim and Imrana, G. 2016). The main advantage to animal manure, with respect to continuous digesters, is that it is easy to collect and easy to mix as slurry and load into digesters.

Plate II: Cow Dung

2.16.2 Groundnut shell

Groundnut (*Arachis hypogaea*) is a native of South America but its cultivation is now widespread globally. It was introduced to the African continent during the colonial era (Duke, 1981). It entered Africa during the Portuguese exploration. World total production as at 2007 was 34.9million metric tons (Food and Agricultural Organization, 2007). Groundnut is produced in Africa majorly by Nigeria, Sudan, Senegal, Chad, Ghana, Congo and Niger. 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 and Jongur, 2014). Groundnut shell (Plate III) is being dumped indiscriminately in Nigeria.

Plate III: *Arachis hypogaea (*Groundnut) Shell

2.17 Factors that affect Biogas Production

2.17.1 Retention time

The incubation period for the Bio Methane Potential (BMP) test is typically 30 days, which ensures virtually complete decomposition of biodegradable organics in most cases. Some organics may require a longer period for acclimation. As such, it is necessary to select the period 51 depending on the specific waste and operational conditions. It is important to mention that many researchers have reported an ultimate methane production peak before or around the 10^{th} day. The number of days the organic material stays in the digester is called the retention time. There are two significant retention times in an anaerobic digester. Solids Retention Time (SRT) and Hydraulic Retention Time (HRT). The SRT is the average time the bacteria (solids) are in the anaerobic digester. The HRT is the time the liquid is in the anaerobic digester. SRT is the most important retention time, and should be determined correctly because it indicates the potential of bacteria wash out. If a significant wash out of bacteria occurs, the digester can fail. The solid retention time is a fundamental design parameter used in process control of AD (Table 2.2). In tropical countries like

India, HRT varies from 30–50 days while in countries with colder climate it may go up to 100 days. Shorter retention time is likely to face the risk of washout of active bacterial population while longer retention time requires a large volume of the digester and hence more capital cost. Hence there is a need to reduce HRT for domestic biogas plants based on solid substrates (Gashaw, 2014). SRT is the theoretical time that microbial cell are retained in a biological system. It is determined as the ratio of mass of biomass in the system to the amount of biomass leaving this system per given time.

$$SRT = \frac{\text{Mass of microorganisms in the system (mg)}}{\text{Mass of microorganism leaving the system per time (mg/day)}} \quad (2.5)$$

Table 2.2: Retention Time

Biomass	Operating Temp. (^{o}C)	Optimum Retention Time (days)	Gas Production litre/day	Volatile Solid Destroyed (%)
Cattle manure	15	60	0.25	40.00
	25	35	0.48	60.00
	35	30	0.66	65.00
Groundnut shell	15	60	0.15	19.00
	25	35	0.24	28.00
	35	30	0.32	29.00

Source: (Hawkes, 1979)

2.17.2 Temperature

Temperature affects bacterial activity. Another way of classifying bacteria is according to their preferred temperature of operation. Psychrophilic bacteria work

best at between 10^{o}C and 20^{o}C (most often referred to as ambient conditions (25^{o}C)). The other two conventional operational temperature levels for anaerobic digesters are determined by the species of methanogens in the digester and they are (i) mesophilic temperature range which operates between 25^{o}C and 40^{o}C. Mesophiles are the primary micro-organisms present (ii) thermophilic temperature range which takes place optimally around 45^{o}C and 60^{o}C or at elevated temperatures up to 70^{o}C. Here thermophiles are the primary micro-organisms present (Song *et al.*, 2004). While anaerobic digestion is very efficient in the thermophilic range, rural users of digesters use mesophilic bacteria because higher temperatures are difficult to maintain (Fulford, 1998). The gas production rate roughly doubles for every 10^{o}C rise in temperature between 15^{o}C and 35^{o}C. Methanogenic bacteria are sensitive to temperature changes. A sudden change of more than 5^{o}C in a day can cause them to stop working temporarily, resulting in a build-up of undigested volatile acids, the digester plant goes "sour". This is less of a problem in large-volume digesters where the high heat capacity of the slurry ensures that its temperature changes slowly.

2.17.3 pH

pH value indicates the degree of acidity or alkalinity of a solution. The pH value is represented as the logarithm of the reciprocal of the hydrogen ion concentration in gm equivalent per litre of solution. pH value in the range 0-7 and 7-14 indicates an acidic or alkaline solution respectively. The micro-organisms require a neutral or mildly alkaline environment – too acidic or too alkaline environment will be detrimental. Ideal pH value is between 7.0 – 8.0 but can go up or down by a further 0.5. In the

initial stages of acid forming stage of digestion, the pH value may be around 6.0 or less, however during methane formation, the pH value of 7.0 is maintained since methane formers are sensitive to acidity. The pH value depends on the ratio of acidity and alkalinity and the carbon dioxide content in the digester, the determining factor being the density of the acids. For the normal process of fermentation, the concentration of volatile acid measured by acetic acid should be below 2000 parts per million too high a concentration will greatly inhibit the action of the methanogenic microorganisms. The survival of methanogenic bacteria also depends on the acidity of the environment that they are in: methanogensis requires a near-neutral pH (between 6.5 and 7.5). A decrease in pH can inhibit gas production and can lead to further accumulation of acids.

2.17.4 Nature of raw materials

Any material containing food substances such as fats, carbohydrates or proteins can be digested in a biogas plant. Feedstocks can include biodegradable waste materials such as waste paper, grass clippings, leftover food, sewage and animal wastes. However, the rate and efficiency of digestion of the feedstock depends on the physical and chemical form. Raw plant material is bound up in plant cells, usually strengthened with cellulose and lignin which are difficult to digest. In order to let the bacteria reach the more digestible foods, the plant material must be broken down. Cattle dung is the easiest feedstock to use for a biogas plant; it already contains the right bacteria and has been ground up by the animal's teeth and subsequently broken down chemically by acids and enzymes in the animals gut. Human, pig and chicken

manure are also good but need a “starter” (seeding), such as slurry from a working plant. Some animals such as horses and elephants are less good at breaking down fibrous materials, so their dung contains more indigestible matter. This can be screened out or chopped mechanically. Goat and sheep dung are rich in nutrients (Yavini *et al*., 2014) but they are in the form of pellets that must be broken up mechanically. Raw vegetable plants usually need to be treated before it can be used. It can be physically chopped up or minced, or it can be treated chemically (Ofoefule *et al.*, 2008).

Murphy *et al.* (2011) indicated that the composition of crops and thus their suitability as AD feedstock varies with the stage of maturity. In general, cellulosic content increases with maturity, negatively affecting the digestibility and the methane yield of the crop. Less mature crops, however, have higher moisture content, making storage difficult.

2.17.5 Agitation/mixing

Mixing is required to maintain fluid homogeneity, hence process stability, temperature distribution, within a digester. The objectives of mixing are to combine the incoming substrate with the bacteria, to reduce the formation of scum, and to avoid pronounced temperature gradients within the digester. Very rapid mixing can disrupt the microbial balance while too slow stirring can cause short-circuiting and inadequate mixing (Abbasi*et al.*, 2011).

2.17.6 Solid content

In a typical scenario, three different operational parameters are associated with the solids content of the feedstock to the digesters. They are (i) high solids (dry – stackable substrate), (ii) high solids (wet-pumpable substrate), (iii) low solids (wet-pumpable substrate). High solids (dry) digesters are designed to process materials with solids content between 25 and 40%. Unlike wet digesters that process pumpable slurries, high solids (dry – stackable substrate) digesters are designed to process solid substrates without the addition of water. The primary styles of dry digesters are continuous vertical plug flow and batch tunnel horizontal digesters. Continuous vertical plug flow digesters are upright, cylindrical tanks where feedstock is continuously fed into the top of the digester and flow down ward by gravity during digestion. In batch tunnel digesters, the feedstock is deposited in tunnel – like chambers with a gas-tight door. The amount of pretreatment such as contaminant removal depends both upon the nature of the waste streams being processed and the desired quality of the digestate. Wet digesters can be designed to operate in either high solids content with a Total Suspended Solids (TSS) concentration greater than ~ 20% or a low-solids concentration less than ~ 15% (Jewell *et al.*, 1993).

High solids (wet) digesters process thick slurry that requires more energy input to move and process the feedstock. The thickness of the material may also lead to associated problems with abrasion. High solids digesters will typically have a lower land requirement due to the lower volumes associated with the moisture. High solids digesters also require correction of conventional performance calculations (such as

gas production, retention time, kinetics) originally based on very dilute sewage digestion concepts, since larger fractions of the feedstock mass are potentially convertible to biogas (Richards *et al.*, 2010). Low solids (wet) digesters can transport material through the system using standard pumps that require significantly lower energy input. Low solids digesters require a large amount of land than high solids due to the increased volumes associated with the increased liquid-to-feedstock ratio of the digesters. There are benefits associated with operation in a liquid environment as it enables more thorough circulation of materials and contact between the bacteria and their food. This enables the bacteria to more readily access the substances on which they are feeding and increases the rate of gas production.

2.17.7 Toxicity

Mineral ions, heavy metals and the detergents are some of the toxic materials that inhibit the normal growth of pathogens in the digester. Small quantity of mineral ions (e.g. Na, K, Ca, Mg, NH_4 and S) stimulates the growth of bacteria while very heavy concentration of these ions will have toxic effect. Similarly heavy metals such as copper, nickel, chromium, zinc and lead in small quantities are essential for the growth of bacteria but their high concentrations have toxic effects. Likewise, detergents, antibiotics, organic solvents, inhibit the activities of methane-producing bacteria and addition of these substances in the digester should be avoided (Anonymous, 2011). Chlorinated hydrocarbons such as chloroform and other organic solvents are particularly toxic to biogas digestion. Care must be taken that the feedstock used in a biogas plant has not been affected by these chemicals.

2.17.8 Dilution

Water should be added, if necessary, to the substrate to generate slurry which is neither too thick nor too thin. If slurry too diluted, the solid particles may settle down in the digester and may not get degraded properly. If the slurry is too thick, it may be difficult to stir and may impede the flow of gas to the upper part of the digester. Different systems can handle different levels of slurry density, generally in the range of 10-25% of solids (Abbasi *et al.*, 2011).

2.18 Characteristics of Biogas Feedstock

2.18.1 Suitability and availability

The substrates used in practice for biogas production are selected based on their suitability and availability. Suitability in this case is defined by a number of characteristics and parameters such as the content of easily digestible organic matter, methane potential, particle size, dry matter content, C and N ratio, the content of macro and microelement. Availability means that the feedstock is easily accessible for biogas plant operators and can be supplied in sufficient amounts on a renewable basis (Arthur *et al.*, 2013). The biomass resources suitable as feedstocks for biogas production vary significantly in term of composition, digestibility, methane potential, dry matter content, content of nutrient and other characteristics. The importance of these characteristics is that they can be used to optimize the AD process and methane production.

2.18.2 Digestibility

Digestibility is the main AD feedstock parameter, with direct influence on methane production, and refers to the ability of the substrate to be decomposed through AD. The digestibility of a certain material depends on its content of easily digestible compounds such as simple sugars. However, biogas feedstock can also contain various amounts of low digestible compounds, known as recalcitrant matter, such as lingocelluloses. (Steffen *et al.*, 1998) noted that the anaerobic degradation rate varies significantly with feedstock composition. Feedstock composition also determines the amount of time necessary to decompose a specific feedstock and thus the necessary retention time of the feedstock inside the digester. Low molecular weight carbohydrates, volatile fatty acids and alcohols are digested in hours; proteins, hemicelluloses and lipids in days while cellulose needs several weeks to be decomposed in anaerobic conditions. Feedstock substrates consisting of fats and oils, known for their very high methane yields, require longer retention times and larger digester volumes compared with substrates rich in carbohydrates and protein. In practice, for economic reasons, digesters are operated with the shortest retention times and the highest methane yields possible.

2.18.3 Presence of impurities

Together with the supplied feedstock, various unwanted components can be accidentally supplied to the biogas plant. Once they enter the digester, their presence can cause perturbations of the normal operation. Common problems are reduction of the active volume of the digester (caused by sedimentation of sand on the bottom of

the digester), process failure through foaming, phase separation and floating layers, or even damage to machinery such as pumps, caused by metallic impurities or other disturbing components (Arthur *et al.,* 2013). The most common disturbing materials is sand, often supplied with animal manure. Light materials such as straw and wood particles may cause floating layers and perturbations of the fluid dynamics. The presence of straw can also have disturbing effects, although this depends on particle size: small-particle straw does not disturb the process and can improve the methane yield considerably (Steffen *et al.,* 1998). Inorganic materials such as glass and metals scrap, polymeric compounds like plastics (often supplied with biogenic wastes) and salts and fatty compounds present in some industrial wastes are also considered disturbing components. Once they occur, disturbing effects are difficult to control. For this reason, all feedstock types must be carefully selected and those containing disturbing components must be avoided or properly pre-sorted before being fed to the digester. The classic example is organic household waste, which is best separately collected (source separation) in order to obtain the required purity and guarantee trouble-free AD and high-quality end products.

2.18.4 Inhibitors

Some compounds in the feedstock (and thus supplied to the digester with the feedstock) can have a negative effect on the microbiology inside the digester, causing imbalance or complete cessation of microbiological activity in the worst case. These are named inhibitors and their inhibitors effects depend to a large extend on their concentration in the feedstock mixture, but also on other local conditions inside the

digester (Steffen *et al.,* 1998). For example, an increased amount of volatile fatty acid (VFA) can cause process imbalance if their concentration inside the digester exceeds the pH buffer capacity of the AD process, reaching so-called shock-levels (Steffen *et al,* 1998). Increased levels of VFA can occur as a consequence of rapid degradation of large amounts of organic macromolecular matter (lipids, carbohydrates or protein). In a 'healthy digesters', microbial adaptation to increased concentrations of VFA occurs eventually. High concentrations of end products such as free ammonia can also have inhibitory effects through accumulation inside the digester.

2.19 Biogas Plants

The biogas plant is a device that converts organic wastes into flammable gas called biogas and into good quality organic manure under anaerobic conditions. Generally a biogas plant is made up of a digester where organic matter ferments and a gas holder where the gas produced is stored for use in cooking, lighting and the generation of electrical or mechanical power as may be required (Maishanu *et al.*, 1990). The success of any biogas plant lies in its construction, operation and maintenance. Anaerobic digester for biogas production also poses some challenges to the effectiveness of the biogas process depending on the process configurations and operating conditions of the reactors. A digester may be suitable and economical for a particular type of feedstock or co-substrate but may not be suitable for another. Therefore, for overall effectiveness of the biogas production process, reactors must be selected with consideration of the feedstock composition, amount of feedstock to be treated, desired product, and process economy (Patinvoh, 2017).

2.20 Types of Anaerobic Digester

The anaerobic digester is the main component of AD. It is an airtight tank that can be of any shape (cylindrical, rectangular, square and egg-shaped), where biodegradation of substrate and biogas production take place (Ward *et al*., 2008). The design of anaerobic digester is required to contain the following basic objectives; to make a sustainable and continuous high organic load rate possible, to achieve a short HRT as possible and to optimise the production of methane (Khalid *et al*., 2011; Ward *et al*., 2008). The anaerobic digester can be broadly categorised according to the feeding mode (batch and continuous) and by the solid content of the substrate (dry and wet digestion) (Ward *et al*., 2008).

2.21 Feeding Mode (batch and continuous)

The batch digester is considered the simplest to design, construct and operate (Khalid *et al*., 2011). The digester is fed with substrate mixed with water to form slurry, then sealed and allowed for a length of time. During the HRT, biogas production progresses to a maximum and then decreases slowly as bacterial effectiveness decreases. Not only that the batch digester is simple to construct and operate, it is also not expensive. It can perform rapid digestion process and be utilised easily to measure the rate of digestion (Khalid *et al*., 2011; Koppar and Pullammanappallil, 2008). However, batch digester has some limitations, including volume restriction, the inconsistent rate of biogas production and varying the proportion of methane content in biogas (Linke *et al*., 2006).

Unlike batch system, continuous fed digester requires daily loading of an organic substrate (Ward *et al*., 2008). The volume is designed large enough to contain the daily substrate feeding throughout the HRT (Ward *et al*., 2008). Continuous feed system can be divided into two types, namely, single-stage and two-stage or multistage digesters (Vandevivere *et al*., 2002). In one-stage continuous feed system, all the four steps of AD process take place simultaneously in one digester. One major disadvantage of this system is that the entire biochemical processes are kept under the same operating parameters, despite the fact that the growth rate of the various bacterial groups involved and their optimal pH ranges are different (Vandevivere *et al*., 2002).

2.22 Digester Design Parameters

When biogas digester is to be designed, the main variable to be defined is its internal volume. The amount of gas produced depends on the volume of slurry in the pit. The digester volume is related to two other parameters, the retention time (R) measured in days) and the feed rate. For a batch digester, the retention time is simply the time the slurry has been left in the pit. For a continuous digester, it is given by the volume of the digester pit (V, m^3), divided by the volume of the daily feed (v, m^3/day).

$$R = \frac{V}{v}\ days \qquad (2.6)$$

The volume feed rate (v) is given by the mass of total solids (m, Kg) fed daily divided by the proportion of Total Solids (TS) in the mixed slurry (assuming the density of feed is 1000 kg/m^3).

$$v = \frac{m}{TSX \times 1000} \; or \; v = \frac{m}{TS\% \times 10} m^3/day \qquad (2.7)$$

The retention time is always a compromise between gas production rate and efficiency. If the supply of feed is limited and the temperature is low (less than 20ºC), the retention time should be as long as possible (up to 100 days) to get maximum gas from the feed. Long retention times also allow less digestible materials in the feed to be broken down. The volume of the plant will be large, though making the cost high. If the feed is in plenty supply and the temperature can be kept high (30ºC), a retention time of 10days is possible, given a high rate of gas production. Special high rate thermophilic reactors can have retention times down to one or two days but these are very expensive to build and operate. At low temperatures, it is important to keep the retention time long, as the bacteria grow more slowly. If the bacteria are removed with the spent slurry faster than they can replace themselves in the digester pit, "washout" occurs and the plant will fail. As the methanogen's multiply more slowly than acid-forming bacteria, the main symptom of wash out is the plant becoming sour. The plant may recover if feeding is stopped for a time. The loading rate (r, kg. VS/m^3/day) of digester is defined as the mass of volatile solids added each day per unit volume of digester. It is related to the mass of feed rate.

$$r = \frac{m \times VS}{v} \quad or \quad r = \frac{\times VS\%}{v \times 100} K_g \; VS/m^3 day \qquad (2.8)$$

Typical values for the loading rate are between 0.2kg.VS/m^3/day and 2.0kg.VS/m^3/day (Fulford, 1998).

2.23. Basic Considerations for Digester Construction

Among other site specific factors, the criteria for the selection of an ideal design should be based on the following considerations:

i It should be simple in terms of construction and operation,

ii It should be cost effective and durable so that the general population is able to embrace this technology,

iii It should be efficient, i.e., the gas production should be optimum per unit volume of a biogas plant for given type and quantity of input,

iv It should be constructed using local materials as much as possible; and

v Repair and maintenance requirement should be minimal.

The biogas plant is also commonly known as a bio-digester, bioreactor or anaerobic reactor (Karki *et al.*, 2005).

2.23.1 Site selection

In order to ensure the sustainability of a biogas installation, a careful selection of the best site for the plant must be made. The factors that should be considered in making the decision are:

i. Distance between the proposed site and the location where gas will be consumed should be as close as possible since gas pipes are expensive;

ii. Distance between the site and the supply of input materials (i.e., cow shed) should also be as close as possible to save input carrying efforts;

iii. Distance between the site and the location where the effluent can be stored (for example compost pits) should be as close as possible. Close distance

helps to ensure that the effluent can flow into the storage pit without much handling;

iv. Distance between the site and sources of water such as wells should be far enough to prevent contamination (say 10 to 15 m). However, it should be noted that if the water source is too far, it will take more time and effort to prepare the slurry since for a given volume of dung an equal volume of water should be added;

v. Distance between the sites and trees/bamboos should be far enough to prevent damage to the structures from the roots of the plants;

vi. Ground water depth should be investigated. Construction will be relatively easy at locations where the ground water table is low.

vii. The ultimate bearing pressure of the foundation should be adequate to support the load of the biogas plant and the slurry inside.

viii. The direction of the prevailing wind should be considered so that the smell from the biogas plant will not be a nuisance to residential areas.

At any particular site it may not be possible to fulfill all of the above criteria. However, efforts should be made to meet as many of the above listed criteria as possible such that the cost is lowered and the plant operation becomes less cumbersome (Karki *et al.*, 2005).

2.23.2 Design considerations

Digester Design:

i. Operating volume:

The operating volume of the digester is simply the volume of slurry in the digester (Ahmadu, 2009).

The operating volume of the digester (V_o) is determined on the basis of the chosen retention time (RT) and quantity of substrate input quantity (Q_S), and is given as (Ahmadu, 2009):

$$v_o = Qs \times RT\ [m^3/day \times number\ of\ days \quad (2.9)$$

The retention time is the interval of time during which the biomass is allowed to decompose in the digester. The retention time, in turn, is determined by the chosen digester temperature and the amount of biomass resource available. Kossmann *et al*., (2001) noted that for a plant of simple design, retention time should amount to at least 30 days.

$$Substrate\ input\ (Qs) = Biomass\ (B) + Water\ (W)\ (\frac{m^3}{day}) \quad (2.10)$$

In most agricultural plants, mixing ratio of dung to water varies from between 1:1 to 2:1.

ii. Total volume:

The total volume of the digester (VT) should be greater than the operating volume. This is to give room for the biogas produced and the rise of the slurry during fermentation. The operating volume of the digester must not exceed 90% of the total volume of the digester (Ahmadu, 2009). The total volume is thus given as:

$$V_T = V_o/0.8 \qquad (2.11)$$

iii. Digester dimensions:

Having determined the total volume of the digester, a ratio for the dimensions can be adopted, depending on the chosen geometric shape of the digester. For a cylindrical digester, the chosen geometry for this work,

$$V_T = \pi r^2 h_d \qquad (2.12)$$

Where V_T = Total volume of digester

r^d = radius of digester

h_d = height of digester

2.24 Lignocellulosic Biomass Conversion

Lignocellulosic material can be utilized to produce various energy products and other potential products. There are many processes that could be applied to convert lignocelluloses to different energy products such as biofuels and biogases. Such processes include anaerobic digestion, fermentation, incineration, pyrolysis, gasification and others (Galbe and Zacchi, 2012). Figure 2.3 presents some of the

potential products that could be produced through different processes using lignocellulosic materials as feedstock.

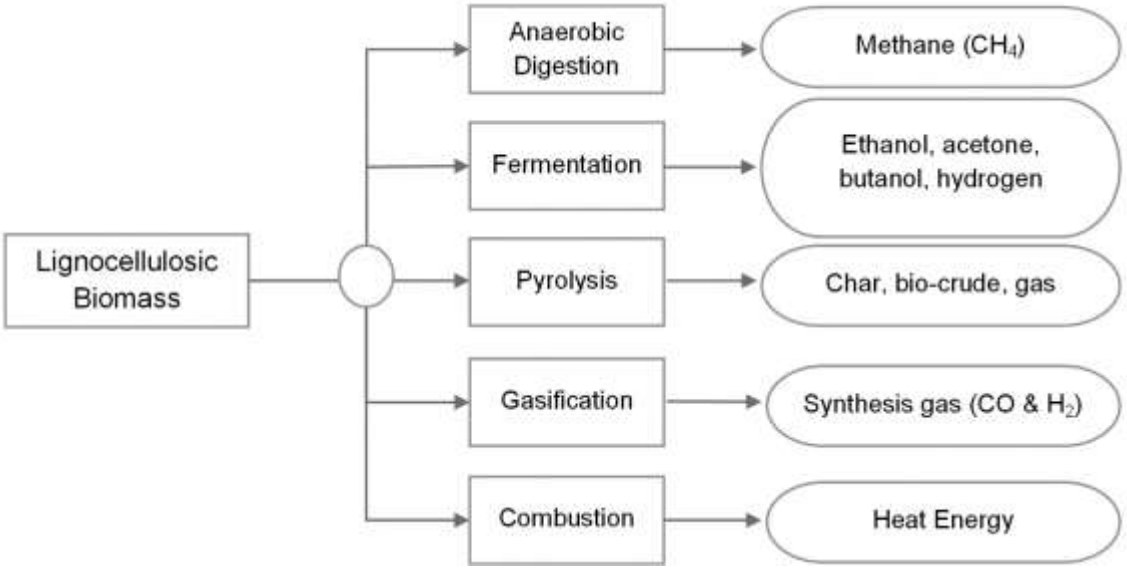


Figure 2.3: Potential products obtained from lignocellulosic materials through various processes.

However, there remain some obstacles impeding the production of energy from the abundant supply of biomass worldwide. The main challenge is the lack of low-cost technology that would make energy production from biomass an economically feasible process (Brodeur *et al*., 2011). For this reason, it is important to consider the economical aspect of new technologies and methods developed to improve the energy production from lignocellulosic biomass.

The focus of this paper is the use of AD as a means of producing biogas (biomethane) from lignocellulosic biomass. AD is very common as it relies on the use of microorganisms as an economical energy recovery process. However, as mentioned earlier, the nature of lignocellulosic material hinders the ability of such

microorganism to efficiently recover energy from lignocelluloses and thus a process enhancement in the form of pretreatment is necessary to achieve economical feasibility and process sustainability**.**

2.25 Pretreatment of Lignocellulosic Biomass

Biomass or plant waste has 3 major components which are cellulose, hemicellulose and lignin besides the extractives and minerals (Di Blasi *et al*., 1999). The content of the cellulose, hemicellulose and lignin normally will be in range 40 - 60 wt%, 20 - 40 wt%, and 10 - 25 wt% of the biomass in dry basis (McKendry, 2002). Rowell *et al*., (2005) stated that major carbohydrate portion in woods is a combination of cellulose (40 - 45 %) and hemicelluloses (15 - 25 %) which so-called holocellulose and usually covered 65 - 70 % of dry basis biomass weight. Cellulose is a major part of polysaccharides that is present in plants accompanied by hemicellulose (Browning, 1967).

Pretreatment of the substrate is needed either for making it easier to handle at the biogas plant or for altering its structure for easy degradation, hence enhancing its methane potential. There are different pretreatment methods that can be used depending on the types of substrates and the goals of the pretreatment. The most suitable pretreatment methods for agricultural, municipal, and industrial solid wastes are discussed in this section. The principal feedstocks used as substrate in the AD process in the work presented herein are lignocellulosic in nature. The bioconversion of lignocellulosic biomass to bio-energy in the form of methane via AD may be

limited by its hydrolysis as the digestible cellulose and hemicelluloses are covered by a sheath of insoluble lignin (Weiland, 2010). In the same way, the advantages and disadvantages of these pretreatments have also been carefully reviewed (Karimi and Taherzadeh, 2016). It is worth mentioning that some hydrolytic substances produced via certain pretreatments methods may be too toxic to the enzymatic biocatalyst and the anaerobic consortium which can lead to poor process yields, and even cessation of the AD process (Jönsson and Martín, 2016).

However, the complexity and variability of the lignocellulosic structure hinder the biodegradation, particularly the hydrolysis of the complex organic matter to turn into soluble compounds, which is the rate limiting step of the degradation (Cesaro *et al.*, 2012). This structural resistance can be broken by physical, chemical and biological pretreatment methods (Niemistö *et al.*, 2013) or by their combinations (Li *et al.*, 2015). The purpose of pretreatment is to change lignin and hemicellulose structures, reduce cellulose crystallinity, and increase the porosity of the materials (Kumar *et al*., 2009), (Figure 2.4 and Table 2.3). The physical pretreatments include mechanical (grinding, milling, ultrasonic and microwave radiations, gas explosions) and thermal treatment methods (hydrothermal treatment, steam explosion and freezing). There are chemical and biological methods as well (Wei *et al.*, 2015).

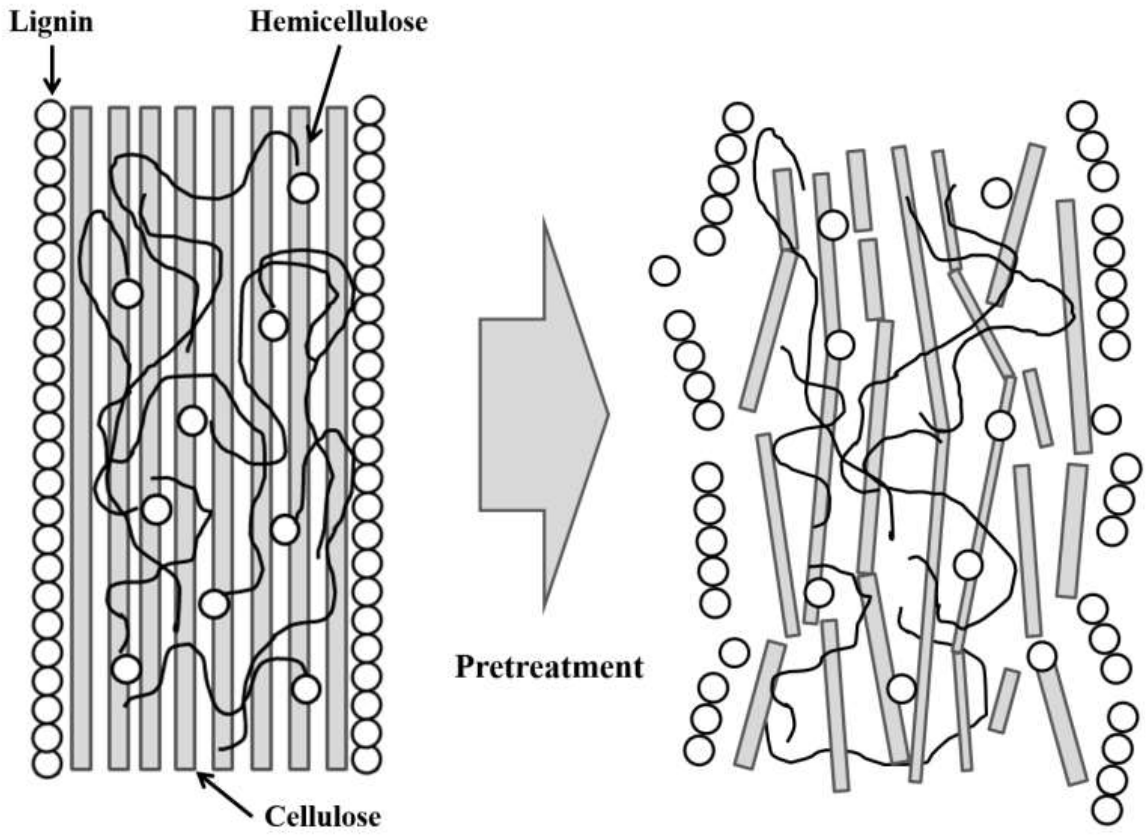


Figure 2.4: The effect of pretreatment of lignocellulosic material (Kumar *et al*., 2009)

Table 2.3: Lignocellulose contents of common agricultural residues and wastes.

Lignocellulosic materials	Cellulose (%)	Hemicellulose (%)	Lignin (%)
Hardwood stems	40-55	24-40	18-25
Softwood stems	45-50	25-30	25-35
Nut shells	25-30	25-30	30-40
Paper	85-99	0	15
Wheat straw	30	50	15
Rice straw	32.1	24	18
Sorted refuse	60	20	20
Leaves	15-20	80-85	0
Cotton seeds hairs	80-95	5-20	20
Newspaper	40-55	25-40	18-30
Fresh bagasse	33.4	30	18.9
Solid cattle manure	1.6-4.7	1.4-3.3	2.7-5.7
Coastal Bermuda grass	25	35.7	6.4
Switch grass	45	31.4	12.0

Source: Compiled from Betts *et al*., 1991; Sun and Cheng, 2002.

2.25.1 Physical pretreatment

2.25.1.1 Mechanical pretreatment

Mechanical pretreatment is required for biomass with high total solids content such as forest residue, crop residue, chicken feather wastes) to reduce the particle size and crystallinity of the feedstock. It can also reduce viscosity in biogas reactors making

mixing easier. During mechanical treatment milling is applied to cut the lignocellulosic biomass into smaller pieces which can effectively work on reducing the crystallinity and the degree of polymerization resulting in increasing available surface area for the attachment of degrading enzymes (Palmowski and Muller, 2000). The milling process can be performed on wet or dried basis based on the mill type. Colloid mill and fibrillator can only work properly for wet materials, such as wet paper and paper pulps while roller mill, extruder and hammer mill are usually used for dry materials. Furthermore, mill ball can work on either dry or wet materials (Walpot, 1986).

2.25.1.2 Thermal pretreatment

Thermal treatment refers to pretreatment methods performed at higher temperatures. The hemicelluloses part is the first to solubilize at temperatures above 150-180°C followed shortly thereafter by the lignin part (Garrote, 1999). During the break down of hemicelluloses acids will also be formed, which then will act as catalysts in the further break down and hydrolysis accelerating the solubilization of hemicellulose's oligomers (Gregg and Saddler, 1996). However, the risk of formation of inhibitory products, such as phenolic and heterocyclic compounds, furfural and HMF, especially in acidic conditions is elevated in heat pretreatment (Ramos, 2003). These inhibitory or toxic products have an adverse effect on the microorganisms during the subsequent bioconversion processes. Therefore, pretreatment at temperatures of 250°C and above should be avoided due to production of unwanted products and pyrolysis reactions (Brownell, 1986).

2.25.1.3 Steam explosion

Steam Explosion (SE) is one of the most effective methods for the pretreatment of lignocellulosic biomass. The substrate is put in a vessel and is exposed to steam at high temperature and pressure for normally 5-30minutes which hydrolyzes the glycosidic bonds in the substrate. After that, the steam is released and the substrate is cooled down quickly which makes water in the substrate to “explode", and opens up the structure of the lignocelluloses in the cell wall of the substrate and makes the biomass inside available to the bacteria (Bauer *et al.* 2009). The biomass undergoes explosive decompression by this swift reduction of pressure (Mood *et al.* 2013). The high efficiency of the steam explosion treatment is due to the thermo-mechano-chemical destruction applied in the method. Steam-pretreatment has been used to hydrolyze the hemicellulose and cellulose of softwood for enhanced bioethanol production (Söderström *et al.,* 2003). Fernández-Bolaños *et al.,* 2001 have shown that steam-explosion improved the accessibility of the cellulose and increased the enzymatic hydrolysis yield of seed husks of olive stones. Kaar *et al.,* (1998) identified the optimum conditions of the steam explosion cycle to pretreat sugarcane bagasse for conversion into ethanol and pointed out that steam explosion processing optimums are highly feedstock dependent, since different carbohydrates compositions dictate different conditions.

The anaerobic fermentation characteristics of green and dried corn straw pretreated by steam explosion method were investigated by Xu *et al.* (2012) who showed that the fermentation cycle of green straw is shorter than that of the dried one by 4-7 days.

Wang *et al.* (2011) used the steam exploded pretreatment technology to process corn stalk, and demonstrated that the biogas production per unit of pretreated corn stalk increased 16.8% ~ 63.2% than the unexploded corn stalk. Varga *et al.* (2004) proved that steam pretreatment removed the major part of the hemicellulose from the solid material, made the cellulose more susceptible to enzymatic digestion and increased the enzymatic conversion (from cellulose to glucose) of corn stover more than four times, compared to untreated material. The results of Wang *et al.* (2014) indicated that the anaerobic digestion of the silage remains after high-solids ethanol fermentation from unwashed steam exploded corn stover was able to improve overall content utilization and extract a greater yield of lignocellulosic biomass compared to ethanol fermentation alone.

Steam explosion pretreatment is usually defined by a severity factor that is calculated from the temperature and duration of the process. The relation between the steam explosion severity, duration, and temperature are depicted by Equation (13) (Amin *et al.*, 2017). A similar trend to the Salix woodchips was observed for birch wood chips (Vivekanand *et al.,* 2013), where an approximately two-fold increase in methane yield was achieved compared to untreated woodchips due to the steam explosion with the severity of 4.5 at temperature 220^{o}C. For agricultural biomass such as wheat straw, the different severity of steam explosion had shown no positive impact on methane yield, but the degradation rate was found to be increased. The severity factor of steam explosion for the majority of feedstocks usually lies within the range of 3.14– 3.56 (Amin *et al.*, 2017).

$$logR_o = log\left\{ t\, e^{\left(\frac{T-100}{14.75}\right)}\right\} \quad (2.13)$$

Where,

logRo = the severity factor as a function of treatment time;

T = Temperature in ^oC;

t = is the residence time in (min); and

14.75 = the activation energy where the process obeys first-order kinetics and the Arrhenius temperature dependence

2.25.2 Chemical Pretreatment

Chemical pretreatment has been investigated using a range of different chemicals, mainly acids and bases of different strengths under different conditions. Unlike alkali pretreatment, acid pretreatment does not disrupt lignin but is thought to work by breaking down hemicellulose and disrupting ether bonds between lignin and hemicellulose (Knappert *et al*., 1981).

2.25.2.1 Acid hydrolysis

Acid hydrolysis is categorized into two groups depending on the acid concentration; dilute acid pretreatment or high concentration acid pretreatment. According to the literature reviewed, dilute acid treatment is among one of the most effective methods for lignocellulosic biomass (Digman, 2010). Dilute acid treatment typically carried out, either at high temperatures (T > 160°C) and continuous flow with low solids loading and short retention times (for example 5 min), or low temperatures (T ≤ 160°C) and batch process with high solids loading at longer retention times (for

example 30-90 min) (McMillan, 1994). There is a variety of acids reported in literature that have been applied to a wide range of feedstocks, including softwood, hardwood, herbaceous crops, agricultural residues, wastepaper, and municipal solid waste. Among the acids, (that is dilute sulfuric acid, dilute nitric acid, dilute hydrochloric acid, dilute phosphoric acid, and peracetic acid) dilute sulfuric acid has been broadly applied due to its low cost and high effectiveness.

When dilute acid is added to the biomass and the mixture is kept at 160-220°C for a few minutes, this treatment offers good performance in the breakdown of hemicelluloses recovering monomeric sugars and soluble oligomers from the cell wall into the hydrolyzate. Consequently, the removal of the hemicellulose fraction increases the porosity of the material enhancing the digestibility (Chen, 2007). However, lignin is not significantly removed in this process. Therefore, this method is more suited for biomass with low lignin content (Yang and Wyman, 2004). Several studies showed that in order to achieve maximum hemicellulose recovery, particular attention should be paid to the applied treatment time, since there is only a relatively short time interval in which the hemicellulose degradation can occur to a considerable extent while the sugar decomposition is still small. Furthermore, it was found that conditions which gave maximum hemicelluloses removal and recovery in the hydrolyzate did not always result in the highest enzymatic digestibility (Cara, 2008). The drawback of this method is the risks of further degradation of hemicelluloses to furfural and hydroxymethyl furfural, which then have an inhibitory effect on the subsequent microbial processes.

2.25.2.2 Alkaline pretreatment

As previously mentioned, lignocellulosic materials are resistant to hydrolysis due to their structure and composition. Alkali addition causes swelling of lignocelluloses (Kong *et al*., 1992) and partial lignin solubilisation. Alkaline pretreatment is one of the major chemical pretreatment techniques used. This pretreatment refers to application of various bases, including sodium hydroxide, potassium hydroxide, calcium hydroxide (lime) (Kim and Holtzapple, 2005), aqueous ammonia and ammonium hydroxide. Alkaline pretreatment mainly results in delignification, together with solubilization of a remarkable amount of hemicelluloses. It is successful in removing acetyl and the various ironic acid substitutions on hemicelluloses which otherwise may decrease the accessibility of enzymes to hemicellulose and cellulose surfaces.

The efficiency of alkaline treatment extensively depends on the properties of the lignocellulosic material treated and on the treatment conditions. Generally, alkaline pretreatment is more successful on the substrates with low lignin content such as hardwoods and agricultural residues than hardwoods with higher lignin content (Kim and Holtzapple, 2005). Alkaline pretreatment is based on saponification of intermolecular ester bonds cross linking lignin and hemicelluloses resulting in a decreased degree of polymerization (DP) and crystallinity, the disruption of the lignin structure and the separation of linkages present between hemicelluloses and lignin. Among the different alkaline solutions investigated for the treatment of

lignocelluloses, aqueous ammonia and lime (calcium hydroxide) pretreatments are considered to be the most effective and inexpensive methods.

2.25.3 Biological pretreatment

A significant drawback with mechanical, thermal and chemical pretreatment techniques is the requirement of high energy input for an improved biomass conversion. Moreover, these methods are generally carried out using expensive instruments and chemicals. In contrast, utilizing microorganisms to enhance the biodegradability of organic matter and consequently methane production, offers advantages such as low-capital cost and low energy demand. In addition, these methods are environmentally sound. On the other hand, biological treatment methods require long resident times because the rate of the biological hydrolysis is usually very low (Sun and Cheng, 2002). Lignin is known as a major factor that determines the extent of biomass degradation in anaerobic conditions, and biological pretreatment methods have been considered as effective and cheap methods of delignification.

Generally, improvement in methane production by fungi is explained by the disruption of cell wall structure. Additionally, lignin degradation also increases the surface area of the cellulose to develop its susceptibility to microbes and enzymes. Microorganisms, such as brown, white and soft rot-fungi, are engaged to degrade hemicelluloses and lignin, but due to its high resistance only very small amount of cellulose will be degraded (Sun and Cheng, 2002). Among the large amounts of fungi

which work to degrade lignocellulosic materials, a white rot fungus, *Ceriporiopsis subvermispora*, is identified as the superior biopulping fungus that can degrade lignin without intensively breaking the cellulose. Other examples for fungi, used for the biological treatment are *Phanerochaete chrysosporium*, *Trametes versicolor*, *Trametes hirsuta* and *Bjerkandera adusta* (Sun and Cheng, 2002). Table 2.4 shows advantages and disadvantages of pretreatment.

Table 2.4: Advantages and disadvantages of different pretreatment technologies

Process	Advantages	Disadvantages
Milling	Increases surface area Makes substrate easier to handle Often improves fluidity in digester	Increased energy Demand High maintenance costs Sensitive to stones
Steam explosion	Breaks down lignin Solubilises hemicellulose Cost effective Yield higher glocuse	High heat and electricity demand Effective at certain temperature
Extrusion	Increases surface area	Increased energy demand High maintenance costs Sensitive to stones etc.
Hot water (TDH)	Increases the enzyme accessibility Effective at certain temperature	High heat demand
Acid	Solubilises hemicellulose	High cost of acid Corrosion problems Formation of toxic substances
Alkali	Breaks down lignin	High alkali concentration in digester High cost of chemical
Microbial	Low energy consumption	Slow No lignin breakdown
Enzymatic	Low energy consumption	Continuous addition required High cost of enzymes

(adapted from Taherzadeh and Karimi (2008a)

2.25.3.1 Mushrooms and mushroom biology

It has been well known that the 20th century has been an explosive time for the accumulation of knowledge. Modern technology for human civilisation is expanding

every day. However, human beings still face and will continue to face three basic problems: shortage of food; pollution of the environment; and diminishing quality of human health, due to the continued increase of the world population. The 20th century began with a world populated by 1.6 billion people and ended with 6billion inhabitants-- with most of the growth occurring in the developing countries. The growing world population is increasing by about 80 million people per year. At the present, about 800 million people in the world are living in poverty. On the other hand, it has been observed that over 70% of agricultural and of forest products has not been put to total productivity, and have been wasted in processing. Macrofungi (mushrooms) not only can convert these huge lignocellulosic biomass wastes into human food, but also can produce notable immune enhanced products, which have many health benefits. Another significant aspect of mushroom cultivation is using the biota in creating a pollution-free environment.

Oyster mushrooms are known to have medicinal properties such as antitumor, antiviral, antineoplastic, antimutagenic, antilipemic, antioxidant (Yashvant *et al*., 2012) and contain good amount of protein, vitamins, minerals, low fat, and crude fiber. Oyster mushroom cultivation is of economic importance in the area of agricultural waste recycling, animal feed, soil remediation, nutrition (Adedokun *et al.*, 2006; Emuh, 2010), economic use of land, income generating (Spore, 2006) and health. Globally, huge volumes of wastes are generated through agricultural, forestry, industrial processes and their accumulation causes environmental pollution. Many agricultural wastes such as banana leaves, corn husk, corn cobs, palm fruit shaft, cotton wastes, sawdust, wheat straw, cassava peel, rice straw, cocoa pods and coconut

husk have been used as substrates (growth medium) for mushroom production (Adedokun *et al*., 2006; Amuneke *et al*., 2011; Stanley *et al*., 2011; Gume *et al*., 2013; Adedokun, 2014).

The advantages of mushroom cultivation can be summarized as:

i. Wastes such as cereal straws are largely burnt by the farmers, which causes air pollution. However, these raw materials can actually be used for the cultivation of mushrooms. This kind of bioconversion exercise can greatly reduce environmental pollution.

ii. Mushroom cultivation can be a labour intensive activity. Therefore, it will serve as means of generating employment, particularly for rural women and youths in order to raise their social status. It will also provide additional work for the farmers during winter months when the farming schedule is light.

iii. It will provide the people with an additional vegetable of high quality, and enrich the diet with high quality proteins, minerals and vitamins which can be of direct benefit to the human health and fitness. The extractable bioactive compounds from medicinal mushrooms would enhance human's immune systems and improve their quality of life.

iv. Mushroom cultivation is a cash crop, whose harvested fruiting bodies can be sold in local markets for additional family income or exported for an important source of foreign exchange that will definitely improve the economic standards of the people.

v. Some warm mushrooms, for example *Volvariella volvacea* (Straw mushrooms) and *Pleurotus sajor-caju* (Oyster mushrooms) are relatively fast growing organisms and

can be harvested in 3 to 4 weeks after spawning. It is a short return agricultural business and can be of immediate benefit to the community.

Anaerobic digestion is an approach that can reduce the environmental risks of the waste. It can also be done by producing biogas, where stored energy in organic waste would change to usable energy. Several studies have been conducted in the field of biogas production along with various biomass and potential surveys oriented towards the production of biogas from different materials. For instance, co-digestion of cow manure and food waste balances the nutrients in an anaerobic digester, and thus providing a more stable environment for the growth of anaerobic bacteria (Banks *et al.,* 2011). Kozlowski *et al.* (2019), economically evaluated the possibility of using dairy waste for the production of electricity and heat. The study reported that the generated waste from the dairy could produce approximately 14.785MWh electricity and 57.815GJ of heat. This supports the construction of biogas plants that can generate electrical power of 1.72 MW.

In another study, thermophilic anaerobic digestion of cattle manure and pasteurised food waste was assessed in batch and high volume lab scale digesters (Zarkadas *et al.*, 2015). During this study, it was found that, the specific methane production increased by about 86%, and a reduction in volatile solid (VS) by about 35.2% when compared to the monodigestion of cattle manure. To enhance the performance of the anaerobic digester, various pre-treatment techniques can be employed. Song and Zhang (2015) conducted the experiments by pre-treating wheat straw with H_2O_2 at different concentrations viz., 1%, 2%, 3%, and 4%. The pretreated feed stock was co-digested

with the dairy cattle manure at different ratios. It was concluded that, wheat straw treated with 3% H_2O_2 was the optimal concentration. Also, the methane yield was found to be higher with the co-digestion of treated wheat straw than untreated wheat straw or dairy cattle manure alone. Most recently, a few articles have been published on the anaerobic co-digestion of livestock manure with the organic wastes are cow manure with barley (Akyol *et al.*, 2016), cow manure with sugar beet by-product (Aboudi *et al.*, 2016), dairy manure with tomato residues and corn stover (Li *et al.*, 2016), and sheep dung with waste paper (Li *et al.*, 2018). The results of all these studies significantly improved the biogas production. Table 2.5 presents the major findings of some of the authors on biogas yield with feed materials.

Table 2.5: Literature on Few Feed Materials and their Results

Authors	Feed Materials used	Results
Busch *et al.* (2009)	Maize, Grass, Sugar Cane	The process was extremely stable and no malfunction has had dictated so far. The biogas obtained has high methane (>72%) and low H_2S concentration (<100ppm)
Kalra and Panwar (1986)	Husk and Straw	The Straw alone produced 456% more gas than Husk alone 167% more than the mixture. The Husk has very small gas potential
Somayaji and Khanna (1994)	Rice and Wheat Straw in CD	Maximum gas production at 100% Rice Straw and 40% wheat straw substitution in CD

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Materials

The waste materials used in the study are cow dung and groundnut shells. Groundnut shells were 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 the 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 shells were sun dried for about fourteen (14) days in order to reduce the moisture content and for ease of handling as depicted in figure 3.1. Dried groundnut shells were further crushed mechanically using pestle and mortar for size reduction, milled into powder form and finally sieved with about 1.18μmm sieve tray.

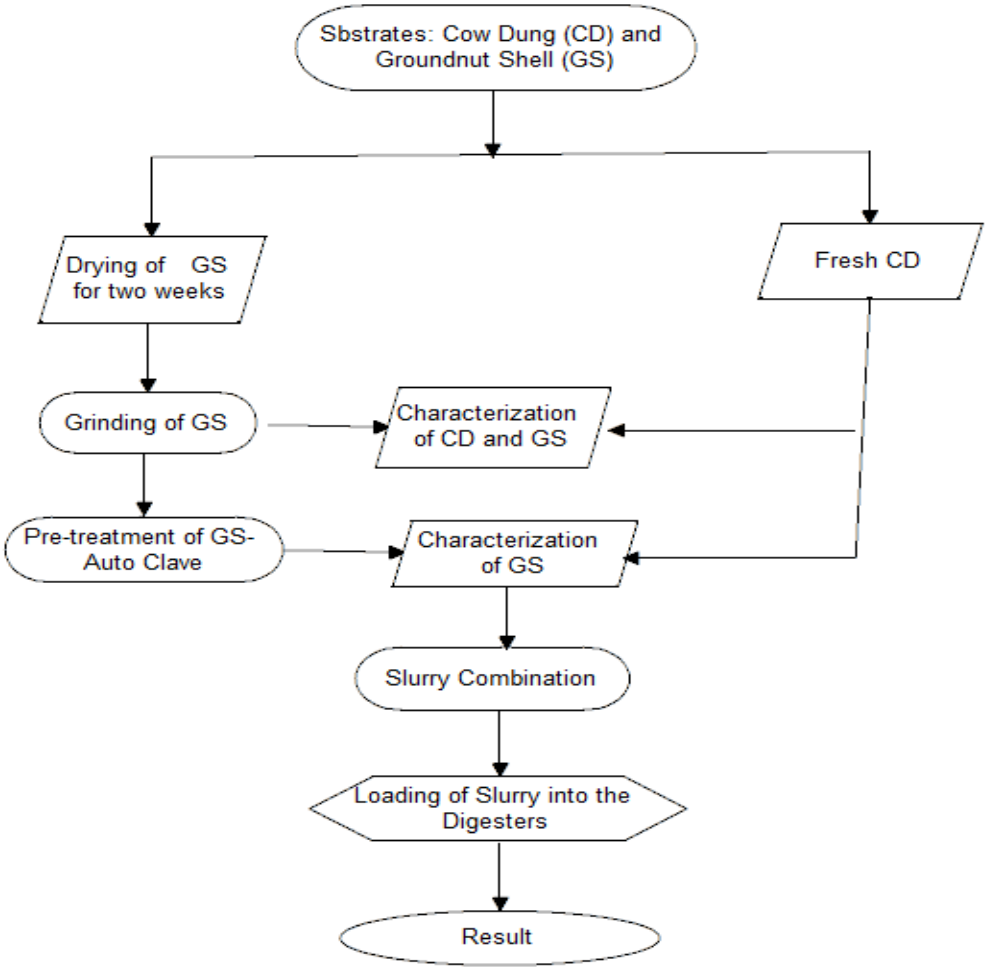


Figure 3.1: Research Flow Chart

The following equipments were used in the study;

i. Digital weighing balance: to determine the weight of the samples.

ii. pH meter: to measure the *p*H of the digested materials daily throughout the retention period.

iii. Measuring cylinder: to measure the volume of water displaced by the biogas generated.

iv. Mixing tank: a big plastic container for mixing the substrate.

v. Thermometer: for measuring the temperature.

vi. Mortar and pestle: for size reduction.

vii. Sieve: for sieving purposes.

viii. Funnel: for feeding the slurry into the digester so as to minimize spillage.

ix. Waterproof sacks for conveying of the substrates.

x. Shovels: for ensuring proper mixing and packing of the substrates.

xi Nose mask: for prevention of inhalation of particulate and odor.

xii Protective gloves: were worn to protect the hands from contamination

xiii Autoclave for steam explosion

xiv Distillation apparatus

xv Muffle furnace

xvi Water bath

xvii Digestion Apparatus

3.2 Pre-treatment Process

Pre-treatment is the first step towards effective conversion of lignocellulocis materials to biogas, which makes up one third of the total production cost and remains one of the barriers preventing commercial success. In this study, steam explosion which is one of the physical forms of pre-treatment and biological pre-treatment were used.

3.3 Lignocellulosic Content Determination

The lignocellulosic content determination was carried out according to (Datta, 1981) method as modified by Arora *et al.* (2013). About 10g of lignocellulocis shredded material was submerged in 100ml distilled water, and placed in an oven at 100^{o}C in a

water bath for 2 hours and sieved through a tare crucible. The residue was put in an oven dried at 90^0C till constant weight. Weight was measured as the water soluble. The dried residue was transferred and submerged in a 100 ml of 0.5 M H_2SO_4 and maintained in a water bath for 2 hours 100^0C, the contents was then sieved, dried and weighed as described above and loss in weight was considered as the hemicelluloses content. For cellulose and lignin estimates, 10 ml of 7.2% (v/v) H_2SO_4 was added to the above dried residue and placed in rotary shaker at 200rpm for 1hour at 30^0C. The incubated mixture was diluted up to 4% H_2SO_4 and placed in an autoclaved at 1.06kg/cm^2 for 40 min. The filtered content was dried and weighed. The loss in weight was recorded as cellulose and the remainder residue was considered as lignin. The same procedure was performed on the culture material to determine the loss in hemicelluloses, cellulose and lignin.

3.4 Preparation of the Substrate for Oyster Mushroom Cultivation

The experiment was carried out in the Animal production Laboratory, Soil science Laboratory and Civil Engineering Laboratory of Federal University of Technology, Minna. The groundnut shell was spread to dry and impurities were removed, milled into powdered form. Ten (10 kg) of the substrates was measured then pasteurized by partly immersing them in hot water (90°C for 4 h). After heat treatment, the substrate was soaked in water for 24hours to moisten them. Subsequently it was stalked on steep cemented floor so as to remove excess moisture from the substrates to get 65% moisture level. The substrate was fermented for 3 days by covering them with polythene sheets before bagging. After fermentation, (1 kg) was loaded into each

polypropylene bag (17cm×33cm×5cm) and each bag was inoculated with 10 g of the spawn, Plates IV. The substrates (now bagged and inoculated) were incubated in a darkroom for 3 weeks on a shelf. During this period, daily temperature and humidity of the incubation room were taken twice daily. The bags were fully colonized by the mushroom mycelia within 17 to 30 days. Next the bags were moved to another room for fructification. The two ends of the bags were cut open with a blade and placed side by side on the shelf provided for this purpose. The humidity of the bags during the cropping (fructification) stage was accomplished by spraying of water in the form of fine mist from a nozzle three times a day. Temperature and humidity of the cropping room were also monitored two times a day. The first primordial (pin heads) appeared 7 to 10 days after opening the bags depending upon the substrate. Matured mushroom were harvested by twisting gently to uproot from the base. The mushrooms generally mature in two to three days after the appearance of the pin heads.

Plate IV: Polypropylene bags filled with substrates for mushroom cultivation.

3.5 Analysis of Growth Rate of Oyster Mushroom

The yield of oyster mushroom was determined by recording the number and size of cap of the fruit bodies after sprouting. The following parameters of growth and yield were measured.

3.6.1 pH

The pH of the samples was determined using the electrometric method with 1: 2.5 sample solutions as used by Page *et al*. (1982). Five grams (5 g) of the air dried substrate was weighed into a 50 ml beaker. Distilled water (12.5 ml) was added. The suspension was stirred vigorously for 20 minutes. The suspension was allowed to stand for 30minutes by which time most of the suspended particles had settled out of the suspension. The pH meter was then calibrated with blank pH of 7. The *p*H meter electrodes were then inserted into the partly settled suspension. The *p*H values were read from the *p*H meter and the results recorded.

3.6.2 Rate of mycelia growth:

The fungal threads (comparable to plant roots) that appears as a network of white filaments which join together to form pinheads which develop into mushrooms. After spawning, a line was drawn across the bags using a permanent marker at where the spawns had settled to serve as a reference point for the measurement of the rate of mycelia formation. A measuring rule was used to measure the distance travelled by the mycelia in the transparent bags at 5 day intervals. The rate of mycelia formation was then calculated by subtracting the new measurements from the previous measurements at each 5 day intervals.

3.6.3 Time for total mycelia formation

The colonization of the substrate by the mycelia within the bags was monitored by measurement at five days intervals. The number of days the mycelia fully colonized the substrate after the day of spawning was then recorded. The colonization was seen by the formation of white mycelia throughout the substrates within the bags.

3.6.4 Time for primordia formation

After the bags were slit open, the formation of primordia was observed every two days intervals and the number of days it took for first primordia formation was observed and recorded.

3.6.5 Weight of harvested mushroom

The total weight of mushrooms harvested from the various treatments was measured using the electronic balance. The weight of the harvested mushrooms at two days intervals were weighed and recorded. The total weight of the harvested mushrooms 30 days after cropping was then calculated by simple addition.

3.6.6 Length of stalk

The length of the stalk was measured using the ruler. Five fruits were randomly selected using simple random technique and the lengths of the stalks were measured from the tip of the stalk to the base of the caps. This was done for each harvest within 20 days and the average determined.

3.6.7 Perimeter of the cap

The perimeter of the caps was measured using a thread and the measuring rule. The thread was used to trace the perimeter of the caps of the five randomly selected fruits. The length of the thread that covered the perimeter of the caps was then measured on the tape rule and the value recorded. This was done for each harvest within 20 days and the average calculated.

3.6.8 Moisture content of harvested mushroom

Five samples of the fruit body of the mushrooms were randomly selected and the moisture content was determined. After the weight of the empty Petri-dish and their covers were recorded, the samples were placed into the Petri-dish and weighed again. The weight of the Petri-dish plus the samples was then recorded. The Petri-dish plus the samples were then placed into the oven and the temperature set at 105^{o}C for 24hours. After this period, the Petri dish plus the samples were removed and placed in a desiccator for 30 minutes. The weight of the dry sample plus the Petri-dish was then measured and recorded. The percentage moisture of the substrates was calculated according to the formula:

$$MC\ \% = \frac{(wt\ of\ dry\ sample + Petri\ dish) - (wt\ of\ empty\ Petri\ dish)}{(wt\ of\ fresh\ sample + Petri\ dish) - (wt\ of\ empty\ Petri\ dish)} \times 100\% \qquad (3.1)$$

3.6.9 Biological efficiency

Total weight of the fruiting bodies harvested from the substrates within 30 days of fruiting was measured as total yield of the mushroom. The biological efficiency (yield

of mushroom per kg substrate on dry weight basis) was calculated by the formula proposed by Chang *et al*. (1981).

$$Biological\ Efficiency\ (B\ E\ \%) = \frac{Fresh\ weight\ of\ mushroom}{Dry\ weight\ of\ substrates} \times 100 \qquad (3.2)$$

3.7 Steam Explosion

As stated earlier, the steam explosion process is performed at high temperatures and pressure of about 240°C 33.4bar respectively and lasts for a few minutes. The pressure is released and biomass cools down quickly thereafter. This sudden drop in pressure causes intracellular water to evaporate very rapidly causing a phenomenon known as steam explosion or phase explosion (Figure 3.2 and appendix A - E). The main purpose of this treatment is to get 80-100% of the hemicellulose fraction solubilized making the cellulose fraction accessible to enzymatic hydrolysis (Grethlein and Converse, 1991). In addition, depolymerization of minor parts of cellulose and lignin can also be achieved**.**

Figure 3.2: Steam explosion equipment

3.8 Anaerobic Digester Set-Up

A 4,000 cm^3 plastic container was obtained from Kure's market, Minna, Niger State, washed and all stains removed. Two holes were drilled; one at the centre with about 1.25 cm diameter, and the other drilled at the side of the container with a diameter of 1.25 cm. A reinforced flexible hose pipe of 100 cm 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 (arodyte) in order to prevent any form of leakages was connected to 2000 cm^3capacity containers which served as the water chamber. The 1.25 cm diameter side hole was fitted with ⅜inch flexible hose pipe, male and female socket and ½inch plug where the sample was

taken for pH. The pH was measured daily using a digital pH meter. The sample to be analyzed were collected into a dry bottle from the digester and then analyzed. The probe of the pH 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 1.25 cm diameter but of 1.10 cm diameter where the thermometer probe was fitted tightly with arodyte adhesive gum. The temperature reading was taken between 2 pm and 4 pm daily throughout the period of the experiment and also the ambient temperature.

Also, a hole of 1.25 cm diameter was drilled at ¾ side of the water chamber and fitted with a reinforced flexible hose pipe of 80 cm diameter from the water chamber connected to 1000 cm^3 capacity containers – water collector. The weight of gas produced was equivalent to the amount of water displaced in the water chamber (Archimedes' principle of floatation). The displaced water was collected in the water collector. The volume of water displaced in the water collector was measured daily (between 2 pm and 3 pm) using a ruler that was attached to the water collector. Before final sealing of the digester and the water chamber, the slurry was stirred properly to avoid lump, and poured into Bio-digester and distil water poured into water chamber. Each treatment was replicated Nine times; Plate's V, VI and appendix F show digester experimental set up.

Plate V: Schematic Diagram of Anaerobic Digestion Set-up

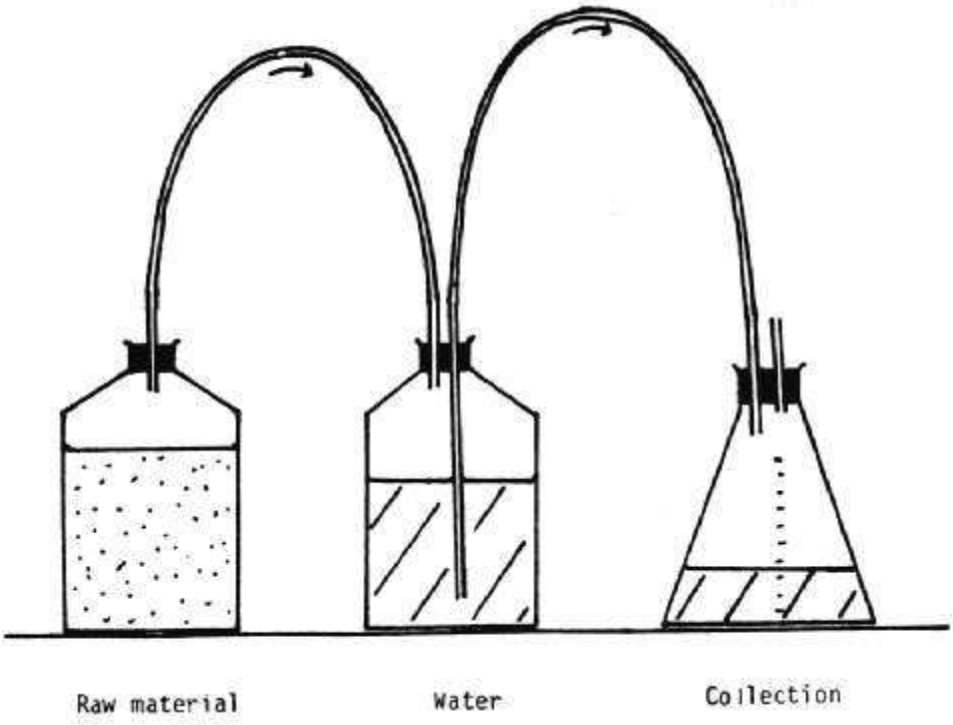


Plate VI: Schematic Diagram of Anaerobic Digestion Set-up

3.9 Fermentation Procedures for the Biological and the Physical Pre-treatment

a) 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.

b) 100% of cow dung and 0% of groundnut shell were mixed with water for Digester A.

c) 75% of cow dung and 25% of groundnut shell were mixed with water for Digester B.

d) 50% of cow dung and 50% of Groundnut shell were mixed with water for Digester C.

e) 25% of cow dung and 75% of groundnut shell were mixed with water for Digester D.

f) 0% of cow dung and 100% of groundnut shell were mixed with water for Digester E

g) The slurry was stirred properly to avoid lump, and poured into Bio-digester A, B, C, D and E respectively for biological pre-treatment and Bio digester F, G, H, I and J respectively for steam explosion (physical pre-treatment).

h) The fermentation was allowed for a period of 30 days under ambient temperature (psychrophilic).

i) 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.

j) The temperature of the medium was taken once daily

3.10 Proximate Analysis

3.10.1 Determination of moisture content (MC)

The hot oven air method of Association of Official Analytical Chemists (AOAC, 2010) was adopted for this analysis. Porcelain crucibles was washed and dried in an oven at 100^{o}C for 30 min. 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^{o}C for 4h. The samples were removed from the oven after this and were cool and weighed. The drying was resumed and all the crucibles with the samples were 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;

$$MC\% = \frac{W_1 - W_2}{W_1} \times \frac{100}{1} \qquad (3.3)$$

Where

W_1 = weight of the original sample

W_2 = weight of final dried sample

3.10.2 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.

These are the procedures that were used in determining the total solid;

i. 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.

ii. The crucibles were weighed (W_2) before use.

iii. The laboratory oven was switch on and allowed to reach a temperature of 105°C. This temperature was maintained throughout the experiment

iii. Substrates were added to the crucibles (W_3) and gently 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 2 hours.

iv. The crucibles plus substrate residues were allowed to cool in a desiccator to balance temperature. The desiccator was properly lubricated with grease and this is to prevent moisture from entering the desiccator as the test glassware cools.

v. The crucibles plus substrate (material) residue were weighed using electronic precision balance (W_1)

Equation (3.4) was used to determine the percentage of total solids.

$$TS\%_s = \frac{W_1 - W_2}{W_1 W_2} \times 100 \qquad (3.4)$$

Where

%TS = Percentage total solid

W_1 = Weight of dried crucible + dried residue

W_2 = Weight of crucible

W_3 = Weight of wet sample (substrate) + crucible

3.10.3 Determination of volatile solids (VS)

The volatile solid is the solid remaining after evaporation or filtrate are dried, weighed, and ignited at 600°C as shown in appendix G. The following procedures were followed in the determination of volatile solid of the substrates.

i. The residues obtained from total solids determination were ignited at 600°C for duration of 30minutes using a muffle furnace.

ii. The crucibles and black mass of carbon were allowed to cool partially in air before it was transferred to the desiccator for complete cooling.

iii. The samples were weighed once temperature balance is reached (W_4)

The percentage volatile solid (VS) were determine using Equation (3.5).

$$\%VS = \frac{W_1 - W_4}{W_1 W_2} \times 100 \quad (3.5)$$

Where,

%VS = Percentage volatile solids

W_4 = Weight of crucible + weight of residue after ignition

3.11 Ultimate Analysis

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

3.11.1 Determination of carbon content

This was determined using the Walkey and Black method. Ten gram (10 g) of GS each of the finely ground substrate was weighed into 500 ml conical flasks(appendixes H-K). Potassium dichromate (10 ml) was poured inside the flasks

and the mixture was swirled. H_2SO_4 (20 ml) was added and the flasks swirled again for 1min in a fume cupboard. Each mixture was allowed to cool for 30 min after which 200 ml of distilled water, 1 g 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 3.6;

$$(C)\% = \frac{B-T \times 133 \times 0.003 \times 100}{W} \qquad (3.6)$$

Where;

B = Blank titre value

T = Sample Titre value

W = Weight of waste sample

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

3.11.3 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 sample are present as nitrogen. Using a conversion factor of 6.25, the actual percentage of protein in the sample was calculated using equation 3.7: Micro-Kjedahl digestion/distillation apparatus and 50 ml Kjeldahl flasks were utilized in carrying out the analysis.

$$crude\ protein\% = \%\ nitrogen\ \times F \qquad (3.7)$$

Where:

F = conversion factor (6.25)

Digestions: Each (2 g) was weighed into Kjeldahl flasks (appendixes H-K). 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, 15 ml) 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 30 min; heated for another 30 min 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 25 ml volumetric flask and made up to the mark with distilled water.

Distillation of the protein: A 50 ml receiver flask containing 5 ml boric acid (methyl red and blue indicator) was placed under the condenser of the distillation apparatus so that the tip was 2 cm inside the indicator. A 10 ml 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 (35 ml) 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 3.8;

$$\%\ nitrogen\ (N) = \frac{Titre\ \times 0.0014\ \times 250}{Weight\ of\ original\ sample} \times 100 \qquad (3.8)$$

3.12 Daily Monitoring of Operational Parameters

In order to study and determine the most feasible local environmental conditions to optimally operate the developed biogas facilities, various physical and chemical parameters were monitored to check the status of the digester. Monitoring of the plant was carried out every day between 10.00AM and 5.00PM. Readings were taken to record the digester pH (appendix L) and temperatures and also the ambient temperature.

3.13 Measurement of Gas Production for the Substrates Digested

The gas holder was calibrated with the aid of a rule (appendix F) to enable the reading of the daily gas production of the anaerobic digesters. Produced biogas measurement

was done each day shortly before sunset. The biogas produced was taken as the total volume of the water displaced. The base diameter of the gas holder was 7 cm.

$$The\ base\ area, A = \pi\frac{d^2}{4} = 38.48\ cm^2 \qquad (3.9)$$

The height of the displaced water was read off on the rule attached to the gas holder for calibration.

Let this height (h) = x, which varies.

Volume of biogas at atmospheric pressure is obtained as the volume of cylinder above water level, given by

Volume, V = $\pi d^2h/4$ = Ah where h =x

Substituting for A from above,

$$V = 38.48 \times cm^3 \qquad (3.10)$$

Where V=volume of biogas

x = height of the water

3.14 Kinetic Modelling of Biogas Generation

To evaluate kinetics of biogas production with regards to prediction of biogas production, Modified Gompertz equation was used to model cumulative biogas production. The constants A, U and λ was determined using the non-linear regression approach with the aid of the solver function of the MS Excel ToolPak. This equation was utilized by researchers to study the cumulative methane production in biogas production. Zwietering *et al.* (1990) applied this equation to study bacteria growth. Budiyono *et al.* (2010) utilized this modified equation to describe biogas yield from cattle manure.

The biogas production kinetics for the description and evaluation of methanogenesis will be carried out by fitting the experimental data of biogas production to various kinetic equations. Biogas production rates of Groundnut shell co-digested with cow dung will be simulated using linear plots. The linear equation of the biogas production rate in the ascending and descending limb is expressed by Equation 3.11 (Kumar *et al*., 2004; Lo *et al*., 2010). It is assumed that biogas production rate will increase linearly with increase in time and after reaching a maximum point after sometime it would decrease linearly to zero with increase in time.

$$y = a + bt \qquad (3.11)$$

Where,

y = biogas production rate in dm^3/gm/day;

t = time in day for digestion;

a = (dm^3/gm/day) and

b = (dm^3/gm/day) are the constants obtained from the intercept and slope of the plot of y vs t.

For the ascending limb, b is positive and it is negative for the descending limb. The exponential plot for the ascending and descending limb can be presented by Equation 3.12 (De Gioannis *et al*., 2009). Here it is assumed that biogas production rate will increase exponentially with increase in time and after reaching the high point it would decrease to zero exponentially with increase in time.

$$Y = a + b\exp(ct) \qquad (3.12)$$

Where,

y = biogas production rate in dm^3/gm/day;

t = time in day for digestion;

a and b = (dm^3/gm/day) are the constants

c = constant (day^{-1}).

For the ascending limb, c is positive and it is negative for the descending limb. In addition, cumulative biogas production was simulated using logistic kinetic model, exponential rise to maximum and modified Gompertz kinetic model. Logistic kinetic equation is shown in Equation 3.13:

$$C = \frac{a}{1 + b \exp(-kt)} \tag{3.13}$$

Where,

C = cumulative biogas production (dm^3/gm);

k = kinetic rate constant (day^{-1});

t = hydraulic retention time (Days);

a , b are the constants.

Exponential rise to maximum is presented in Equation 3.14 (De Gioannis *et al*., 2009; Lo *et al*., 2010):

$$C = A\,(1 - \exp(-kt)) \tag{3.14}$$

Modified Gompertz kinetic model equation is a modified form of the Gompertz equation which is commonly used to simulate the cumulative biogas production (Lo *et al*., 2010). This model assumes that cumulative biogas production is a function of hydraulic retention time. The modified Gompertz equation can be presented as follows (Budiyono *et al.*, 2010; Yusuf *et al*., 2011):

$$Y = Aexp\{-\exp\left[\frac{\mu\, e}{A}(\lambda - T) + 1\right]\} \quad (3.15)$$

Where

Y = Cumulative of specific biogas production (ml)

A = Biogas production potential (ml)

μ = Maximum biogas production rate (d^{-1})

λ = Lag phase period

T = Cumulative time for biogas production (days)

e = Mathematical constant (2.718282)

3.15 Procedure to Fit the Data in to Solver Equation

i Solver menu was selected under the tools icon.

ii A new pop up window appeared

iii A target cell was labeled by typing $G: $4

iv "Equal to " the min function was selected to minimize the value in cell G4

v In the labeled box, the cells were changed to G1: G3

vi Solver values was varied for A, C and K to minimize the sum of Chi Squared.

vii A box appeared to choose solve or cancel

viii Solve option was clicked on the menu and initial values were altered to fit the data

ix A new pop up appeared asking if to keep the values or revert to the original values, and keep solver solution was selected.

x A, C and K values were inserted in cells G1:G3

xi A graph of column B against Column C was plotted, and two curves were matched very closely. But if they do not, then a better guesses for A, C and K should be selected to start with.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Feedstock Characterisation

The results of the physico-chemical analyses of the substrates prior to anaerobic digestion are shown in tables 4.1a, 4.1b and appendix M. The result of chemical analyses showed that steam explosion reduces the total solid of groundnut shell from 87.90% to 79.42%, while; the volatile solid was increased from75.11% 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 the factors affecting the anaerobic process; it affects methane yield and production rates.

Table 4.1a: 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 (C)	42.00	62.02	61.90
Nitrogen Content (N)	0.38	0.50	0.70

Table 4.1b: 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

Table 4.1b showed lignocelluloses content of the substrate: Hemicellulose before and after pre-treatment are 40.20% and 34.11% respectively, Cellulose are 30.50% and 28.80% while lignin before and after pre-treatment are 35.39% and 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 *et al.,* 2012). Cellulose is a main structural component in a plant cell.

4.2 Monitoring of Operational Parameters

1. pH before and after digestion.

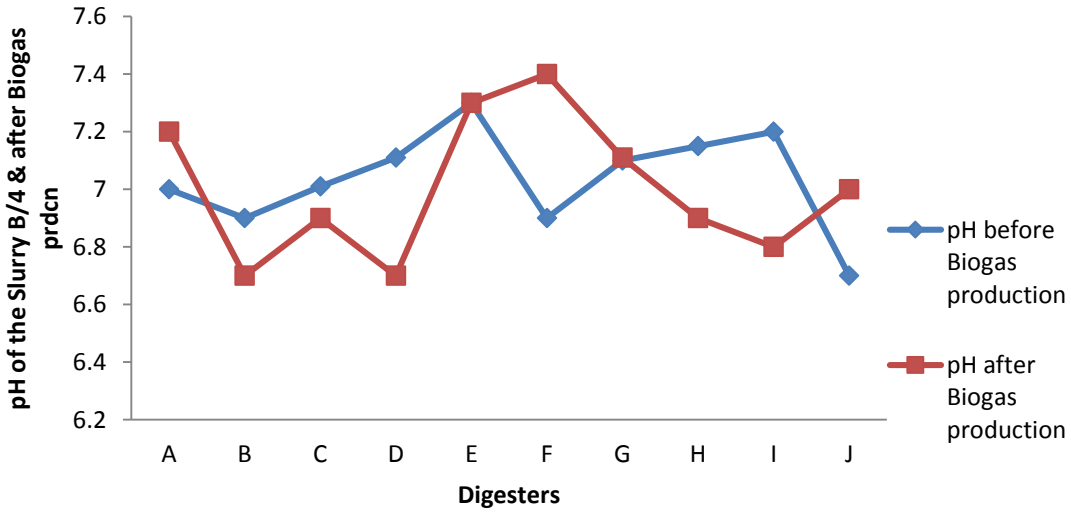


Figure 4.1: pH of the Slurry before and after Biogas production

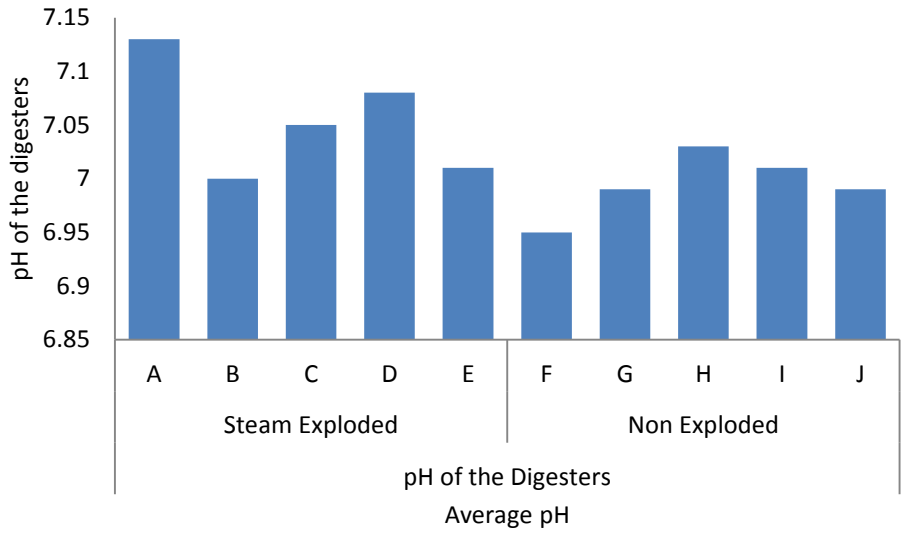


Figure 4.2: Average pHs of the Digesters

From Figure 4.1, 4.2, and appendixes N-O, it was revealed that the acidity in the digester caused the very low yield of biogas in the first 7 days of retention. The result was compared to some other results obtained previously by (Musa and Raji, 2016) from the analysis of biogas from three organic wastes, (Nwanko *et al.*, 2017) who generated biogas from kitchen waste and cow dung, and (Otun *et al.*, 2015) who evaluated the production of biogas from the co-digestion of animal, food and fruit waste.

During the early stage of decomposition, the acid-forming bacteria were found to be breaking down the substrate with volatile fatty acids produced. This changed the values of the general acidity for the digesting material with the value of the pH falling below neutral (Ajiboye *et al.*, 2018). As the weeks went by, the organic acids produced during acetogenesis (majorly acetic acid) were acted upon by methanogenic bacteria and hence broken down into methane and carbon dioxide; the major

constituents of biogas. The pH begins to rise as the acetic acid is converted into biogas. It should be noted that pH affects the growth of microbes during anaerobic fermentation/digestion. Otun *et al* (2015)reported that it is important to maintain the pH of an anaerobic digestion process between 6 - 8, in order not to inhibit the growth of methanogens.

4.3. Digester Temperature during Biogas Production

Figure 4.3 and appendix P shows the averages slurry temperature trend of the mixed substrates. The ambient temperature varied from 28˚C and 40˚C with the mean temperature at 34 ± 1.58˚C, this fluctuation is as a result of climatic conditions, which in turn affects the slurry at each stage of digestion. The mesophilic (21˚C - 38˚C) is the temperature range that was identified from the slurry temperature. This is similar to the result observed by Otun *et al* (2015) and Nwanko *et al* (2017). From the results obtained, anaerobic bacteria thrive best at a mesophilic temperature of about 34˚C (Okewale *et al.*, 2018). Temperature is observed by many biogas researchers as a critical condition for anaerobic digestion, as methanogenic bacteria operate most efficiently at temperatures 30˚C - 40˚C (Deepanraj *et al.*, 2014). The ambient temperature affects the rate of digestion due to the direct contact of the outside walls of the digester and the atmosphere (Okewale *et al.*, 2016).

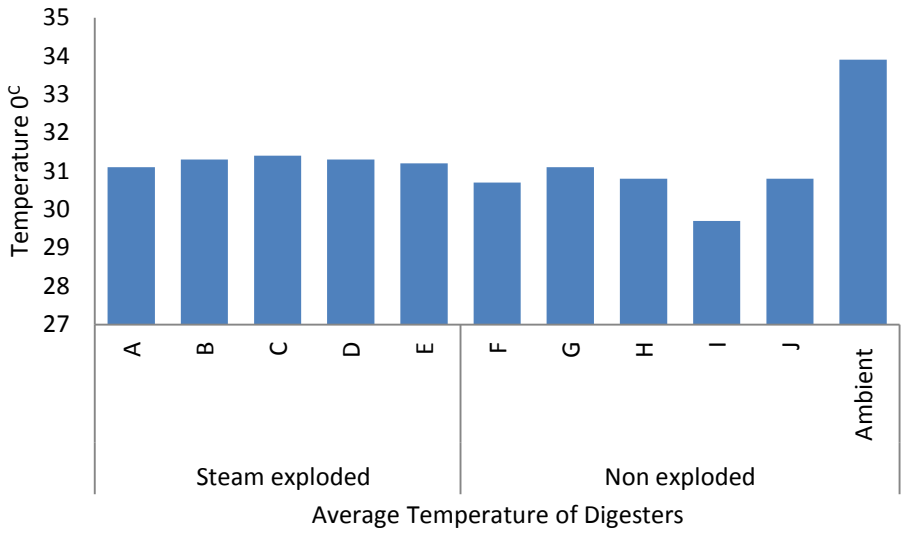


Figure 4.3: Average Temperature of Digesters and Ambient Temperature (^{0}C)

4.4 Volume and Cumulative Biogas Production (cm^3)

Figure 4.4 and appendixes Q - S reveals that biogas production was delayed till the fourth day, which could be related to the fact that most cows feed on fibrous materials and microorganisms require a longer time to degrade fibrous materials. This finding corroborates well with previous reports by Babatola, (2008) in Akure, and Ukpai and Nnabuchi (2012) in Abakaliki, both in Nigeria. The absence of biogas production in the first three days could result from multiple carbon sources in the cow dung (substrate). As one carbon source is exhausted due to an anaerobic condition, the microbial cells divert their energy source for growth to a new carbon supply (Tyagi *et al.,* 1981). A close examination of the findings of this research shows that biogas production was less and gradual in the first week of the investigation as shown in figure 4.5a and 4.5b. This suggests that the biogas producing microorganisms are in the lag phase of growth, where acclimatization or adaptations of the cells take place.

It can also be deduced from this that biogas production rate is equivalent or dependent on the growth of methanogens. From the second week of the study, results indicated a progressive increase in biogas production, which continued to the third week of the study. This indicates that the methanogens are in their exponential stage of growth. However, this differs from the findings of Rabah *et al. (*2010) in Sokoto and that of (Abubakar and Ismail, 2012), where biogas production experienced a decline in the late fourth week. These differences observed may be due to the different breeds of cows found in the different locations. Also, climatic factors, the nature or quality of feed or pasture that the cows were exposed to, are factors that could contribute to the differences in the rate of biogas production (Abubakar and Ismail, 2012)**.**

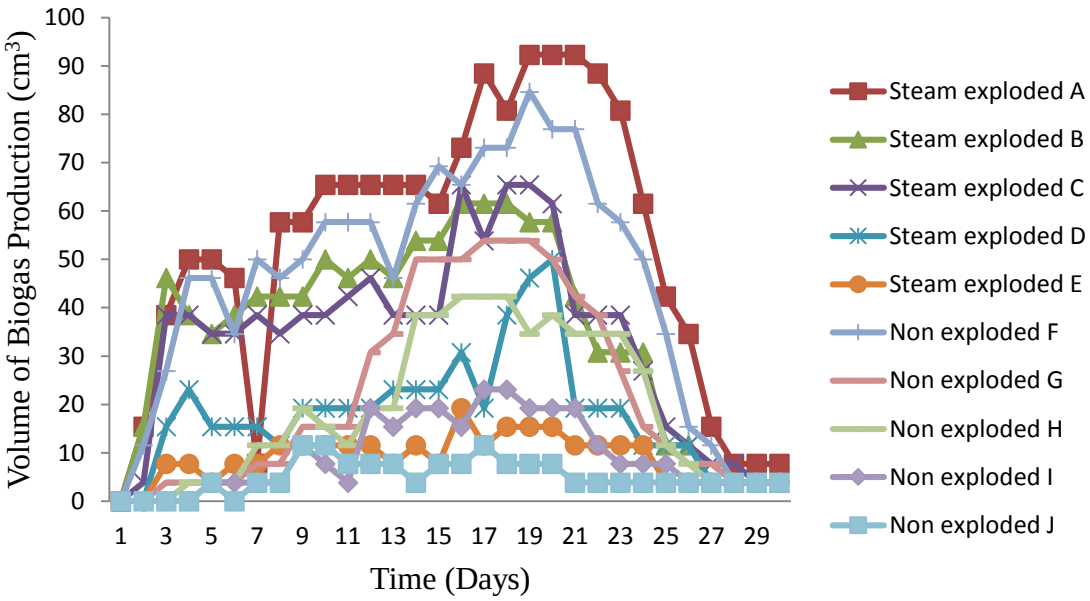


Figure 4.4: Volume of Biogas Production (cm^3)

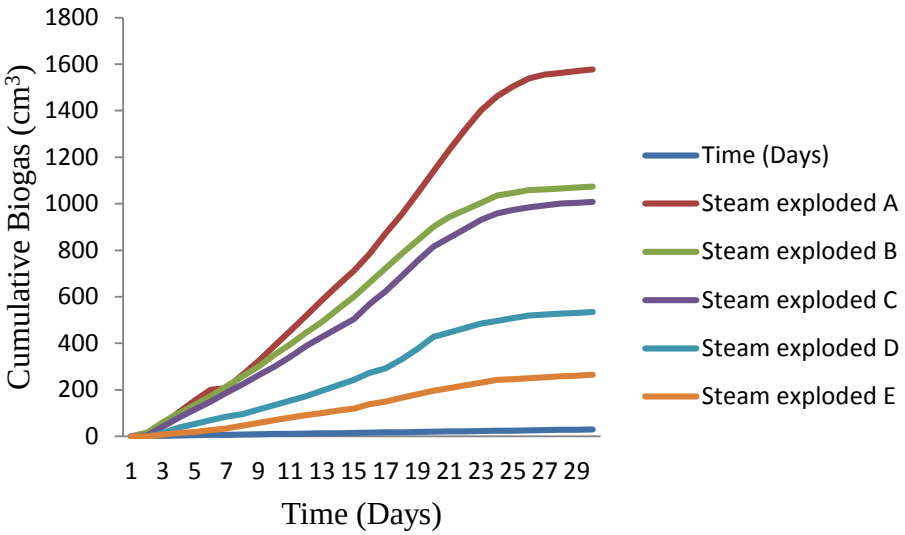


Figure 4.5a: Cumulative Biogas Production for Steam exploded (cm^3)

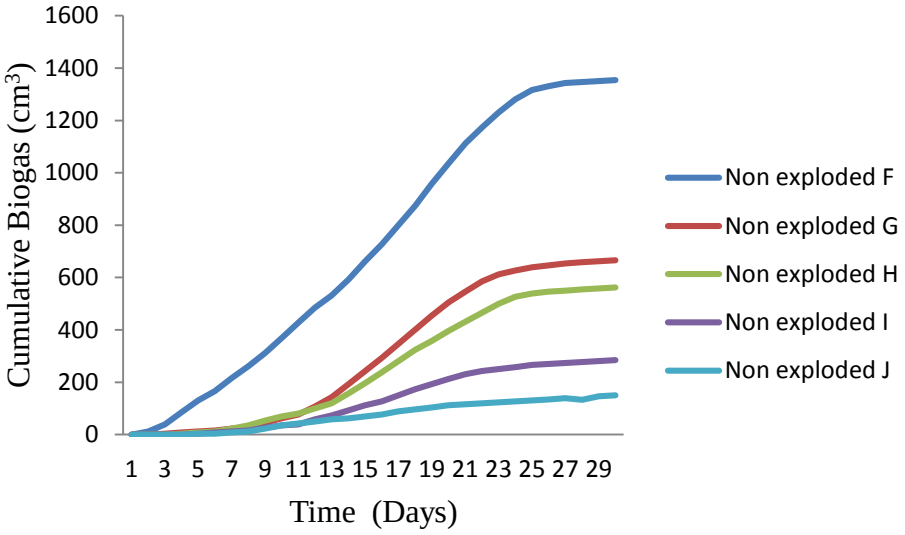


Figure 4.5b: Cumulative Biogas Production for Non exploded (cm^3)

4.5 Analysis of the Biogas Production with the Modified Gompertz Model

The study of biogas production from cow dung and groundnut shell was conducted in digesters labeled A-J. Biogas production was monitored and measured until biogas production reduced significantly. The modified Gomperzt equation was then used to

fit the cumulative daily biogas production which was observed to adequately describe the biogas production from the co-digestion of the substrates.

The biogas produced is a function of bacterial growth in batch digesters, modified Gompertz equation relates cumulative biogas production and the time of digestion through biogas yield potential (A), the maximum biogas production rate (μ) and the duration of the lag phase (λ). To analytically quantify parameters of the reactors growth curve, a modified Gompertz equation was fitted to the cumulative biogas production data as shown in Figures 4.6a-j, Table 4.2a-j and Table 4.3. From the Figures, digesters A and F have the highest biogas production potential of 58cm^3 and 30 cm^3 at a biogas production rate of 92.35 cm^3 and 84.66 cm^3 with a lag phase of 21 days and 19 days respectively. Digesters A and F contained 100% of CD which is an indication that they are a good source of catalyst to increase the volume of biogas production.

Modified Gompertz Model

P*EXP(-EXP(((R*2.7188282)/P)*(L+T)+1))

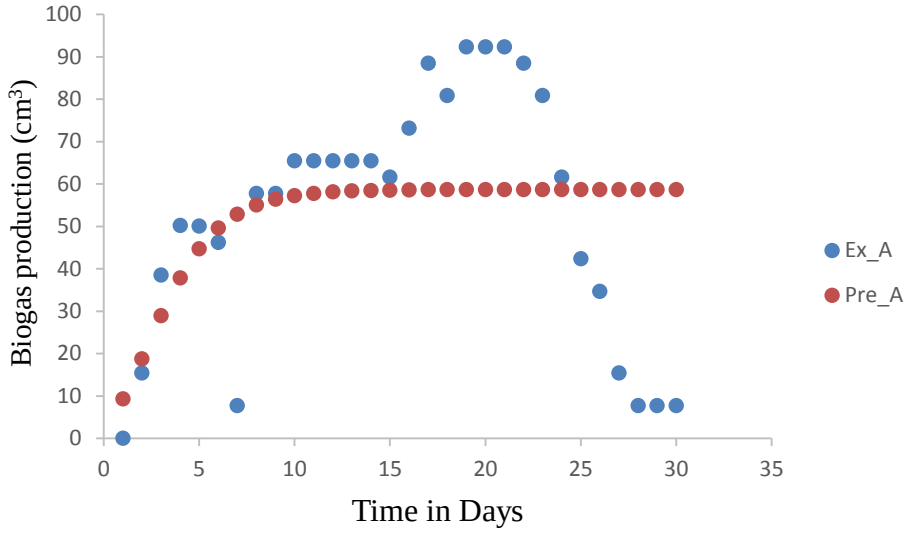


Figure 4.6a: 100% CD

Table 4.2a: Parameter Estimates MODEL A (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	58.676	6.024	46.315	71.037
R	-10.324	9.230	-29.261	8.614
L	- 1.82	2.816	-5.962	5.592

Model B

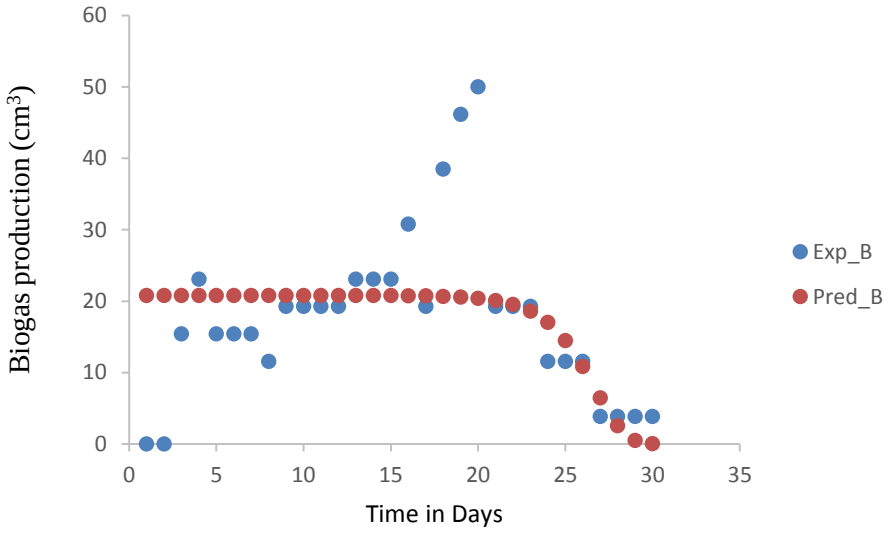


Figure 4.6b: 75%CD & 25%GS

Table 4.2b: Parameter Estimates MODEL B (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	44.631	3.047	38.379	50.883
R	11.399	7.234	-3.444	26.241
L	- 26.464	1.351	-29.236	-23.693

Model C

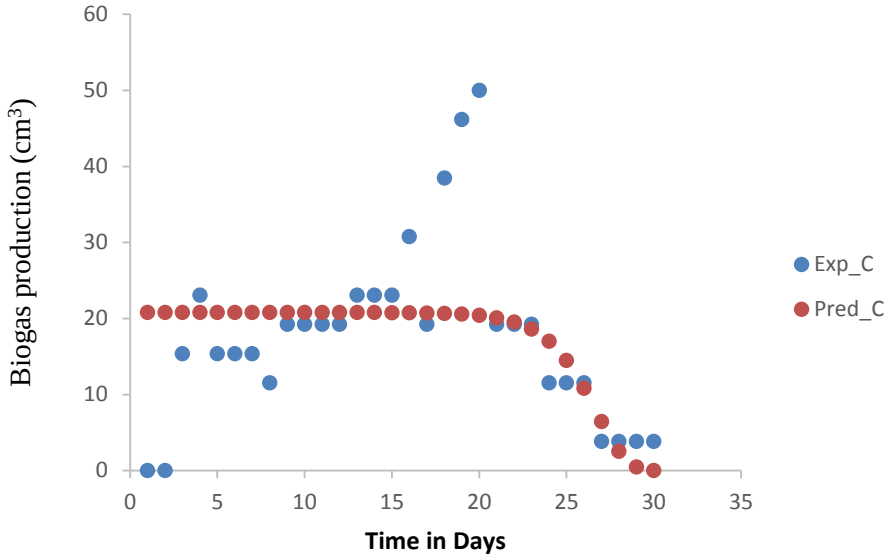


Figure 4.6c: 50%CD & 50%GS

Table 4.2c: Parameter Estimates MODEL C (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	40.683	3.270	33.973	47.392
R	12.306	10.297	-8.822	33.434
L	- 26.667	1.510	-29.765	-23.570

Model D

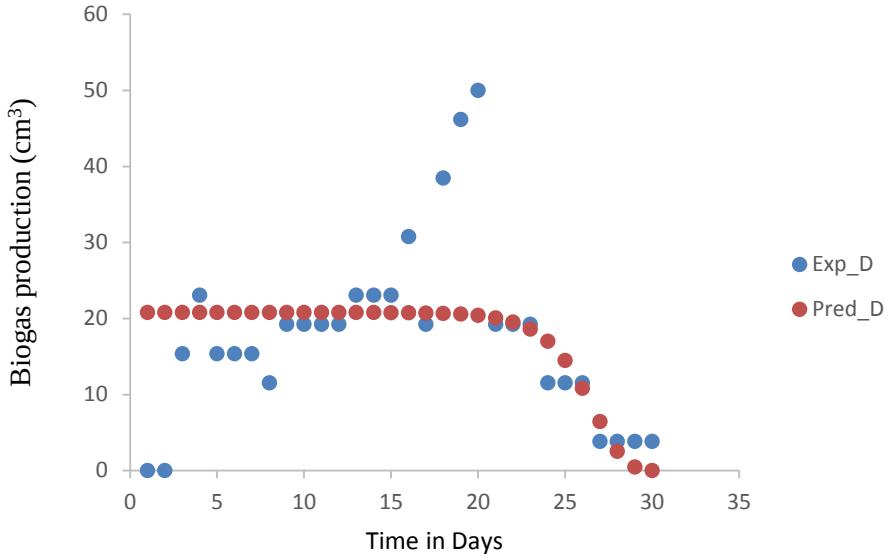


Figure 4.6d: 25%CD & 75%GS

Table 4.2d: Parameter Estimates MODEL D (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	20.795	2.462	15.742	25.847
R	4.489	3.541	-2.776	11.754
L	- 28.434	2.893	-34.370	-22.498

Model E

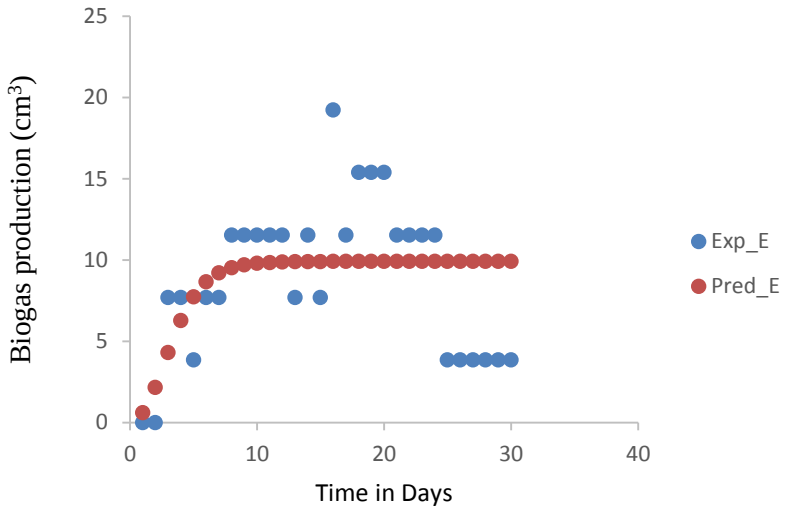


Figure 4.6e: 100%GS

Table 4.2e: Parameter Estimates MODEL E (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	9.920	.906	8.060	11.779
R	-2.207	1.871	-6.046	1.631
L	-1.046	2.074	-5.302	3.210

Model F

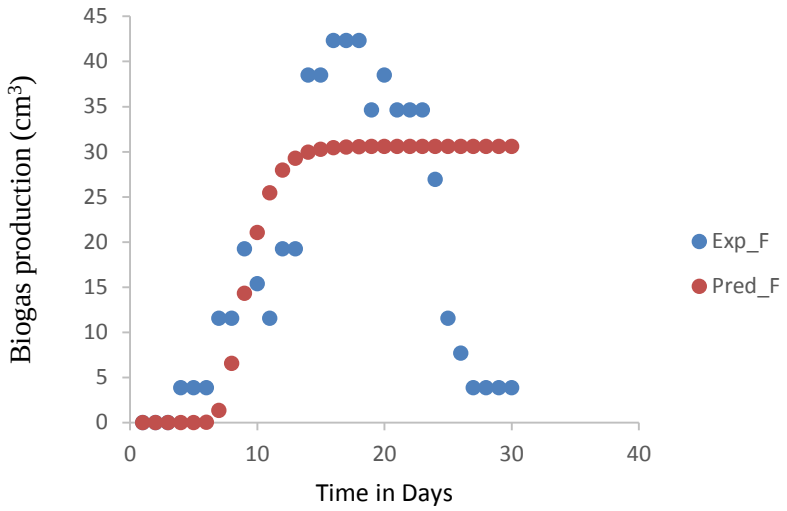


Figure 4.6f: 100%GS

Table 4.2f: Parameter Estimates MODEL F (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	53.578	3.997	45.376	61.779
R	23.815	24.147	-25.731	73.361
L	-26.707	1.245	-29.263	-24.152

Model G

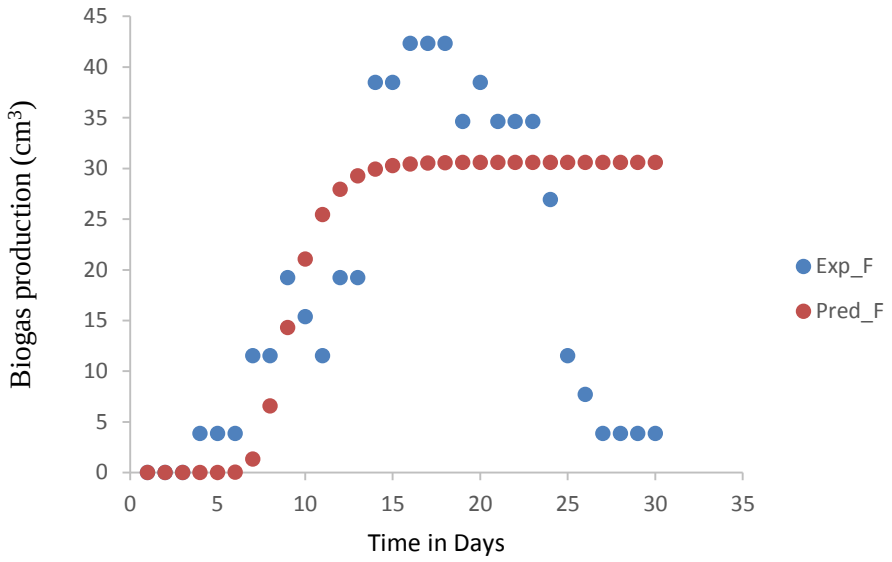


Figure 4.6g: 25%CD & 75%GS

Table 4.2g: Parameter Estimates MODEL G (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	30.604	4.094	22.204	39.003
R	-7.966	9.340	-27.129	11.197
L	-7.198	2.445	-12.215	-2.180

Model H

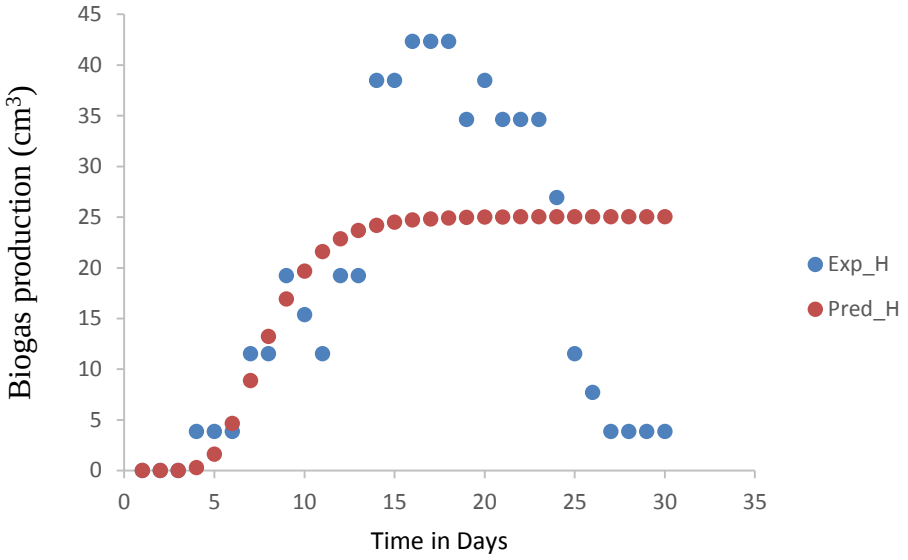


Figure 4.6h: 50%CD & 50%GS

Table 4.2h: Parameter Estimates MODEL H (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	25.034	3.213	18.442	31.627
R	-4.473	4.113	-12.912	3.966
L	-5.012	2.767	-10.689	0.665

Model I

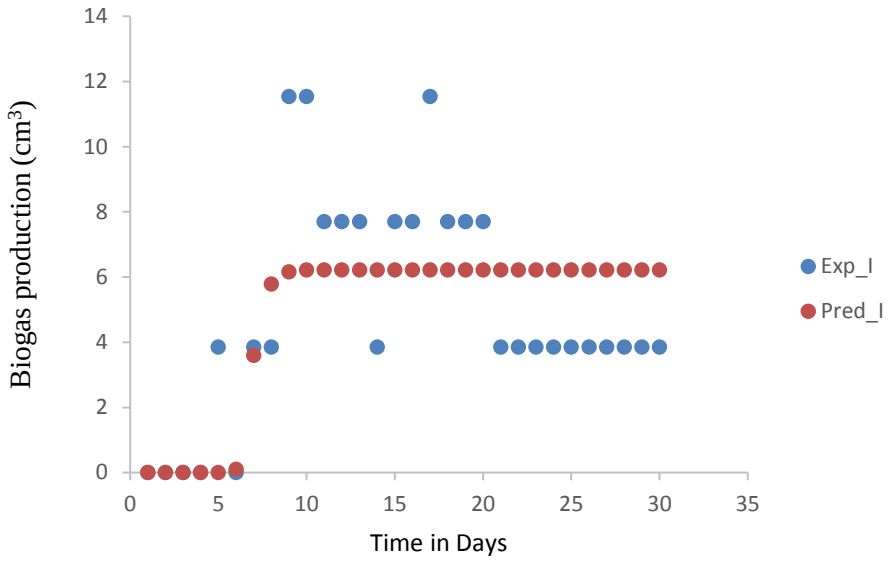


Figure 4.6i: 25%CD & 75%GS

Table 4.2I: Parameter Estimates MODEL I (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	12.555	1.544	9.388	15.722
R	-2.584	2.506	-7.726	2.558
L	-5.240	2.546	-10.464	-0.17

Model J

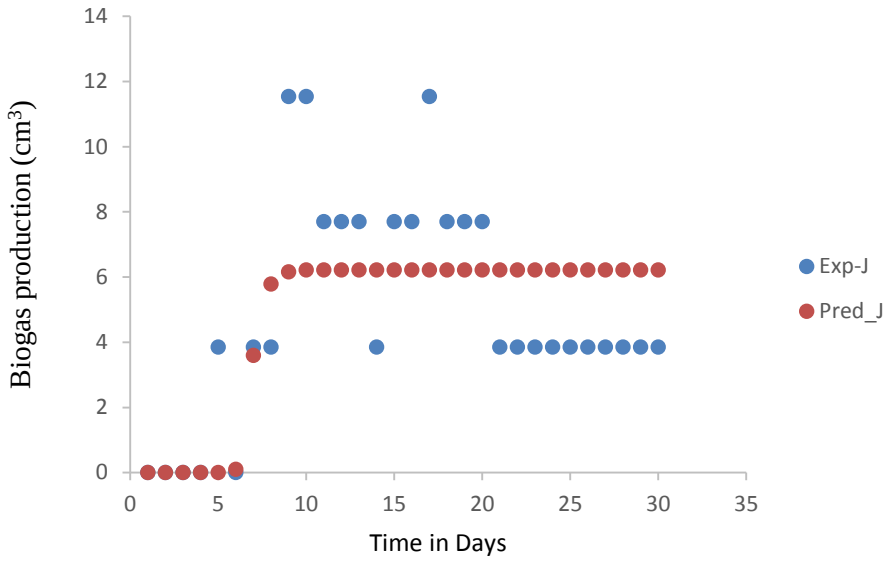


Figure 4.6j: 100%GS

Table 4.2j: Parameter Estimates MODEL J (95% Confidence Interval)

Parameter	Estimate	Std Error	Lower Bound	Upper Bound
P	6.224	0.558	5.078	7.369
R	-4.623	10.001	-25.144	15.898
L	-6.207	1.829	-9.960	-2.453

Table 4.3: R-Squares (Measures the goodness of fit)

A	B	C	D	E	F	G	H	I	J
0.204	0.587	0.453	0.249	0.274	0.476	0.352	0.368	0.374	0.444

CHAPTER FIVE

5.0 CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

From the results of the study conducted, the following conclusions can be made:

Biogas digesters of 4,000 cm^3 were designed modifying the Ajoy Karki's Biogas model fabricated using locally available materials and tested under the existing weather condition in Minna. The biogas digesters constructed in this study were used for the anaerobic digestion of cow dung, groundnut shell as well as co-digestion of cow dung and groundnut shell respectively. The study has shown that biogas can be produced from cow dung and groundnut shell (as observed in previous studies). 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. Kinetics of biogas production was studied here by applying modified Gompertz equation and it was found that the data predicted by the model are quite close to the experimental data with ±10% error.

On a global scale, the study addresses and contributes to planetary health (health, places, and planet) directly or indirectly. The biogas yield was dependent on the temperature of the environment where the digesters were placed. The pH on the other hand was affected by the Carbon: Nitrogen (C:N) ratio of the mixed substrates. An increase in the amount of gas produced with respect to the retention time of twenty-five (25) days, yielding a reasonable amount of gas. The quantity and quality of biogas produced after the 25^{th} day makes the biomass to be regarded as the best

mixture and the best C:N ratio for optimized biogas production. In conclusion, the mixed substrates produced biogas faster than other substrates in mixture when compared to findings from literatures. The cost of production of one digester was ₦17,250.00; this is considered a price affordable to the lower and middle class. One of the limitations to the research and adoption of technology is the availability and regular supply of feedstock.

5.2 Recommendations

1. The work done herein can be used to produce good quality biogas for use locally and internationally.

2. This technology should be encouraged in the rural areas where our forest resources are stretched due to over dependency on wood.

3. The technology again can play a major role in achieving the United Nations Sustainable Development Goal of climate action.

4 The use of biogas will not only serve as a source of fuel but will also help in the management of waste. The biomass generated after digestion can be used both as animal feed and to improve soil fertility. It is therefore recommended that large scale production of biogas from wastes should be undertaken by all as the wastes around you today can become our wealth tomorrow

5.3 Contribution to Knowledge

The research has established that biogas can be produced from co digestion of cow dung and groundnut shell in a plastic digester of size 4,000 $cm^{3.}$ It is also established that steam exploded substrate produced more gas than when non exploded. Considering the relatively low cost of the substrates in addition to controlling environmental pollution, the use of groundnut shell and cow dung as substrate for biogas production is concluded a worthwhile venture and substrates are best efficient in biogas production when used in its crude form

REFERENCES

Abbasi, T., Tauseef, S. M., & Abbasi, S. A. (2011). *Biogas energy*: Springer Science & Business Media, 2, 304-306.

Aboudi K., Álvarez-Gallego C. J., & Romero-García L. I. (2016). Evaluation of methane generation and process stability from anaerobic co-digestion of sugar beet by-product and cow manure. *Journal of Bioscience and Bioengineering*, 121(5), 566-72.

Abubakar, B., & Ismail, N. (2012). Anaerobic digestion of cow dung for biogas production. Asian Research Publishing Network. *Journal of Engineering and Applied Sciences*, 7(2), 169-172.

Abubakar, M. M. (1990). Biogas generation from animal wastes. *Nigeria Journal of Renewable Energy*, 1, 69 – 73.

Adedokun, O. M., Ayodele, V. I., & Fasidi, I. O. (2006). Spawn production and growth of Pleurotus tuber regium (fries) singers on agricultural wastes. *Bioscience Research Communications*, 15(6), 437 – 444.

Adedokun, O. M. (2014). Oyster mushroom: Exploration of additional agro-waste Substrates in Nigeria. *International Journal of Agricultural Research*, 9(1), 55-59.

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

Ahmadu, T. O. (2009). Comparative Performance of Cow Dung and Chicken Droppings for Biogas Production, M.Sc Unpublished Thesis Submitted to the Department of Mechanical Engineering, Ahmadu Bello University, Zaria.

Ajiboye, A.V., Lasisi, K. H. & Babatola, J. O. (2018). Evaluation of the Effect of Sodium Hydroxide Solution on Biogas Yield of Anaerobic Digestion of Poultry Waste and the Digestate. *International Journal of Energy and Water Resources*, 2, 23-31. https://doi.org/10.1007/s42108-018-0003-2

Akinbomi, J. F. K., Ilori, M. O., Oyebisi, T. O., Akinwumi I. O., & Adeoti O. (2000). Biogas Energy use in Nigeria: Current Status, Future prospects and policy implication. *Renewable and Sustainable Energy Reviews*, 5, 97-112.

Akyol, C., Ozbayram, E. G., Ince, O., Kleinsteuber, S. & Ince, B. (2016). Anaerobic co-digestion of cow manure and barley: Effect of cow manure to barley ratio on methane production and digestion stability. *Environmental Progress & Sustainable Energy*, 35(2), 589-595.

Amin, F. R., Khalid, H., Zhang, H. U., Rahman, S., Zhang, R., Liu, G., & Chen, C. (2017). Pretreatment methods of lignocellulosic biomass for anaerobic digestion, 7, 72.

Amuneke, E. H., Dike, K. S., & Ogbulie, J. N. (2011). Cultivation of Pleurotus ostreatus: An edible mushroom from agro base waste products. *Journal of Microbiology and Biotechnology Research*,1, 1-14.

Anonymous (2011). Biogas – Document Transcript www.slideshare.net/arushdeep/biogas-7329849. Retrieved 18/02/18.

Association of Official Analytical Chemists (AOAC), (2010). Official methods of Analysis. Association of Analytical Chemists. 14th ed. Washington, USA, 22209.

Arora, K., Sharma, S., & Sharma, A. (2013). Comparative Evaluation of Chemicals and Botanicals based Dehydration of Biogas Slurry, *Journal of Industrial and Scientific Research,* 35, 131-153.

Arthur, W., Jerry, M. & David, B. (2013). *The biogas handbook: Science, production and applications*. Oxford Cambridge Philadelphia, New Delhi. Woodhead publishing Limited.

Babatola, J. O. (2008). Comparative study of biogas yield pattern in some animal and household wastes. *African Research Review*, 2(4), 54-68.

Banks, C. J., Salter, A. M., Heaven, S. & Riley, K. (2011). Energetic and environmental benefits of co-digestion of food waste and cattle slurry: A preliminary assessment. *Resources, Conservation and Recycling*, 56(1), 71-9.

Bauer, A., Leonhartsberger, C., Amon, B., Boesch, P., Friedland, A., & Amon, T. (2009). Analysis of methane potentials of steam-exploded wheat straw and estimations of energy yields of combined ethanol and methane production. *Journal of Biotechnology*, 142, 50 - 55.

Betts, W. B., Dart, R. K., Ball, A. S., & Pedlar, S. L., (1991). Biosynthesis and Structure of lignocellulose. In Betts (eds) Biodegradation: Natural and Synthetic Materials. Springer-Verlag, Berlin, Germany, 139-155.

Berglund, M., & Börjesson, P. (2006). Assessment of energy performance in the life-cycle of biogas production. *Biomass and Bioenergy*, 30**,** 254-266.

Bhatt, A., & Tao, L. (2020)**.** Economic perspective of biogas production via anaerobic digestion. *Bioengineering*, 7, 74.

Bond, T., & Templeton, M. R. (2011). History and future of domestic biogas plants in the developing world. *Energy for Sustainable Development,* 15**,** 347-354.

Boon, D., & Mah, R. (2006). Transitional bacteria. In: Anaerobic Digestion of Biomass. *Renewable Energy*. 35.

Braun, R. (2007). Anaerobic digestion: a multi-faceted process for energy, environmental management and rural development, In: Improvement of Crop Plants for Industrial End Uses, P. Ranalli (Ed.), 335-416, Springer, ISBN 978-1-4020-5485- 3, Dordrecht, The Netherlands.

Brodeur, G., Yau, E., Badal, K., Collier, J., Ramachandran, K. B., & Ramakrishnan, S. (2011). "Chemical and Physicochemical Pretreatment of Lignocellulosic Biomass: A Review," *Enzyme Research*, 2011, 1–17.

Brownell, H. H. (1986). Steam-explosion pretreatment of wood: Effect of chip size, acid, moisture content and pressure drop.*Biotechnology and Bioengineering 28*, 792-801.

Browning, B. L. (1967). *Methods of wood chemistry*, Volumes I & II, John Wiley & Sons, Inc., New York, USA.

Budiyono, I. N., Widiasa, S. J., &Sunarso, O. (2010). The Kinetic of Biogas Production Rate from Cattle Manure in Batch Mode. *International Journal of Chemical and Biological Engineering*, 10(1), 68-75.

Busch G., Grobmann J., Sieber M., & Burkhardt M. (2009). A new and sound technology for biogas from solid waste and biomass. *Water air Solid Pollution Focus*, 9, 89-97.

Cara, C. 2008). Conversion of olive tree biomass into fermentable sugars by dilute acid pretreatment and enzymatic saccharification.*Bioresource Technology*, *99*, 1869-1876.

Cesaro, A., Naddeo, V., Amodio, V., & Belgiorno, V. (2012). Enhanced Biogas Production from Anaerobic Codigestion of Solid Waste by Sonolysis, *Ultrasonics Sonochemistry*, 19(3), 596-600. http://dx.doi.org/10.1016/j. ultsonch.2011.09.002.

Chang, S. T., Lau, O. W., &Cho, K. Y. (1981). The cultivation and nutritive value of Pleurotus sajor-caju. *European Journal of Applied Microbiology*, 12, 58 – 62.

Chen, Y. (2007). Potential of Agricultural Residues and Hay for Bioethanol Production, *Applied Biochemistry and Biotechnology, 142*, 276-290.

Chima, C. N., Jude, N. O., Justina, C. O., & Ekpewerechi, S. A. (2013). Biogas Potential of Organic Waste in Nigeria. *Journal of Urban and Environmental Engineering*. 7(1), 110-116.

Conti, F., Wiedemann, L., Sonnleitner, M., Saidi, A., & Goldbrunner, M. (2019). Monitoring the mixing of an artificial model substrate in a scale-down laboratory digester. *Renewable Energy*, 132, 351–362. http:// dx.doi.org/ 10.1016/ j.renene.2018.08.013.

Cows to Kilowatts, (2005). *Nigeria: Turning Waste into Energy and Fertiliser* Innovative for Sustainable Development: Local Case Studies from Africa. http://www.un.org/esa/sustdev/publications/africa_casestudies/kilowatts.pdf. Retrieved 2nd February, 2018.

Dahlquist, E. (2013). Technologies for Converting Biomass to Useful Energy: Combustion, Gasification, Pyrolysis, Torrefaction and Fermentation.

Datta, R. (1981). Acidogenic fermentation of lignocelluloses acid yield and conversion of components. *Biotechnology bioengineering*, 2167-2170.

De Bruyn, J., & Hilborn, D. (2007). Anaerobic digestion basics. Ontario Ministry of Agriculture, Food and Rural Affairs. www.ontario.ca/nasm-omafra. Retrieved 18/02/18.

Deepanraj, B., Sivasubramanian, V., & Jayaraj, S. (2014). *Solid Concentration Influence on Biogas Yield from Food Waste in an Anaerobic Batch Digester.* Proceeding on International Conference and Utility Exhibition on Green Energy for Sustainable Development, Pattaya, 19-21, 1-4.

De Gioannis, G., Muntoni, A., Cappai, G., & Milia, S. (2009). Landfill gas generation after mechanical biological treatment of municipal solid waste. Estimation of gas generation rate constants. *Waste Management*, 29, 1026–1034.

Department of Environment, Food and Rural Affairs (DEFRA). (2013). Statistics on waste managed by local authorities in England in 2012/13, London.

Di Blasi, C., Signorelli, G., Di Russo, C., & Rea, G. (1999). "Product distribution from pyrolysis of wood and agricultural residues, *Industrial & Engineering Chemistry Research*, 38(6), 2216-2224. DOI: 10.1021/ie980711u

Digman, M. F. (2010). Optimizing on-farm pretreatment of perennial grasses for fuel ethanol production, *Bioresource Technology, 101*, 5305-5314.

Dioha, E., & Nfor, B. (2017). Operating and Financing a Family Biogas Plant. *Journal of Energy Technologies and Policy*, 2(5), 25-37.

Duke, J. A. (1981). *Handbook of legumes of world economic importance*. Plenum Press. New York.

Energy Commission of Nigeria (ECN)&United Nations Development Programme (UNDP), (2005). Renewable Energy Master Plan. Final Draft Report.

ECN, (1998). Economic Commission of Nigeria (ECN) World solar programme 1996 2005, Projects of the Government of Nigeria: Projects Documents ECN Abuja, 1998.

Edelmann, W., Joss, A., & Engeli, H. (1999). Two Step Anaerobic Digestion of Organic Solid Wastes. In: 11 International symposiums on Anaerobic Digestion of Solid Wastes. *International Association of water quality*, Barcelona, Spain, 150 – 153.

Emuh, F. N. (2010). Mushroom as a purifier of crude oil polluted soil. *International Journal of Science and Nature*, 1(2), 127-132.

Esan, A. A. (2008). Developing Global Network for promoting renewable energy policy and Legislation in Nigeria, Energy Commission of Nigeria, NATCOM-UNESCO. The National Workshop on Creating Legislative Framework and Awareness for Use of Alternative Energy for Sustainable Development in Nigeria. Calabar, Nigeria, 24-26.

Eurostat. (2014). Generation of waste by the Construction sector, by country, year, and waste category, in kg per inhabitant and tonnes. Retrieved from http://epp.eurostat.ec.europa.eu/portal/page/portal/waste/waste_generation_and_management/generation/construction

Ezeah, C., Fazakerly, J. A., & Roberts, C. L. (2013). Emerging trends in informal sector recycling in developing and transition countries. *Waste Management.* 33, 2509-2519.

Food and Agricultural Organization (FAO). (2007). The roles of agriculture in development: policy implications and guidance. Research programme summary report 2007. Socio-economic analysis and policy implications of the roles of agriculture in developing countries, Roles of Agriculture Project Phase II. Rome.

Fernández-Bolaños, J. (2001). Steam-explosion of Olive Stones: Hemicellulose Solubilization and Enhancement of Enzymatic Hydrolysis of Cellulose, *Bioresource Technology*, 79(1), 53-61. http://dx.doi.org/10.1016/S0960-8524(01)00015-3.

Fergusen, R. M. W., Villa, R., & Coulon, F. (2014). Bioengineering options and strategies for the optimization of anaerobic digestion processes. *Environmental Technology*, 3(1), 1–14. http:// doi.org /10.1080 /09593330.2014.907362

Fulford, D. (1998). Running a Biogas programme. A hand book: ‘’*How biogas works*.” Intermediate Technology Publication 103 – 105. Southampton Row, London, W, C, IB 4H, UK, 33 – 34.

Galbe, M., & Zacchi, G. (2012). “Pretreatment: The key to efficient utilization of lignocellulosic materials,” *Biomass and Bioenergy*, 46, 70–78.

Garrote, G. (1999). Hydrothermal processing of lignocellulosic materials, *European Journal of Wood and Wood Products*, 57, 191-202.

Gashaw, A. (2014). Anaerobic co-digestion of biodegradable municipal solid waste with human excreta for biogas production: A review. *American Journal of Applied Chemistry*. 2(4), 55-65.

Godi, N. Y., Zhengwuvi, L. B., Abdulkadir, S., & Kamtu, P. (2013). Effect of cow dung variety on biogas production**.** *Journal of Mechanical Engineering Research. 5*(1), 1-4.

Greentumble, (2015). Waste incineration: Advantages and disadvantages. http://greentumble.com/waste-incineration-advantages-and-disadvantages/. Accessed 29 August 2017.

Gregg, D., & Saddler, J. (1996). A techno-economic assessment of the pretreatment and fractionation steps of a biomass-to-ethanol process.*Applied Biochemistry and Biotechnology 57-58*, 711-727.

Grethlein, H. E., & Converse, A. O. (1991). Common aspects of acid prehydrolysis and steam explosion for pretreating wood, *Bioresource Technology 36*, 77-82.

Gujer & Zehnder, (1983). Biogas production from co-digestion of corn cobs, groundnut shell and sheep dung. *Biomass and Bioenergy*, 4(3), 11 – 21

Gume, B., Muleta, D., & Abate, D. (2013). Evaluation of locally available substrates for cultivation of some edible mushrooms. *African Journal of Microbiology Research*, 7, 2228-2237.

Hawkes, D. I. (1979). Factors Affecting net Energy production from Mesophilic Anaerobic Digestion: Proc. At International Conference on Anaerobic Digestion venue of conf. St. Joseph's At Michigan, U. S.A.

Humenik, F. J., and Hanna, G. P. (2007). Respiratory relationships of a symbiotic algal-bacterial culture for wastewater nutrient removal. *Biotechnology Bioengineering*, 12, 541 –560

Ibrahim, M. D., & Imrana, G. (2016). Biogas production from lignocellulosic materials: co-digestion of corn cobs, groundnut shell and sheep dung. *Imperial Journal of Interdisciplinary Research*, 2(6) 5-11.

Igboro, S. B. (2011). Production of Biogas and Compost from Cow Dung in Zaria, Nigeria, unpublished PhD Dissertation Presented to the Department of Water Resources and Environmental Engineering, Ahmadu Bello University, Zaria Nigeria.

Ilochi, E. E., & Nwachukwu, A. (1989). Symposium *Biotechnology for Energy*. Dec 1621.

Isa, B., Post, J., & Furedy, C. (2014). Solid Waste Management and Recycling; Actors, Partnerships and Policies in Hyderabad, India and Nairobi, Kenya; Kluwer Academic Publishers: Dordrecht,London, UK.

Isci, A., & Demirer, G. N. (2007). Biogas production potential from cotton wastes. *Renewable Energy*, 32**,** 750-757.

Iweka, S., Owuama, K., Chukwuneke, J., & Falowo, A. (2021). Optimization of biogas yield from anaerobic co-digestion of corn chaff and cow dung digestate: RSM and Python approach. Heliyon, 7, e08255.

Jewell, W., Cummings, R., & Richards, B. (1993). Methane fermentation of energy crops. Maximum conversion. Kinetics and insitu biogas purification, *Biomass and Bioenergy*, 5(3 – 4), 261 – 278.

Jönsson, L. J., & Martín, C. (2016). Pretreatment of lignocellulose: Formation of inhibitory byproducts and strategies for minimizing their effects. *Bioresource Technology*, 199, 103-112.

Kaar, W. E., Gutierrez, C. V., & Kinoshita, C. M. (1998). Steam Explosion of Sugarcane Bagasse as a Pretreatment for Conversion to Ethanol.*Biomass and Bioenergy*, 14(3), 277-287. http://dx.doi.org/10.1016/S0961-9534(97)10038-1

Kalra, M. S., & Panwar, J. S. (1986). Anaerobic digestion of rice crop residues. *Agricultural Wastes*, 17, 263–269.

Karak, T., Bhagat, R. M., & Bhattacharyya, P. (2012). Municipal Solid Waste Generation, Composition, and Management: The World Scenario. Critical Reviews in Environmental Science and Technology, 42(15), 1509-1630.

Karimi, K., & Taherzadeh, M. J. (2016). A critical review of analytical methods in pretreatment of lignocelluloses: Composition, imaging, and crystallinity. *Bioresource Technology*, 200, 1008-1018.

Karki A. B., Shrestha N. J., & Bajgain S. (2005). *Biogas as Renewable Energy Source in Nepal*: Theory and Development. Nepal, BSP. Obtainable on www.snvworld.org

Kim, S., & Holtzapple, M. T. (2005). Lime pretreatment and enzymatic hydrolysis of corn stover, *Bioresource Technology, 96*, 1994-2006.

Khalid, A., Arshad M., Anjum, M., Mahmood T., & Dawson L. (2011): The Anaerobic digestion of solid organic waste. *Waste Management*, 31 (2011) 1737-1744.

Knappert, D., Grethlein, H., & Converse, A., (1981). Partial acid hydrolysis of poplar wood as a pretreatment for enzymatic hydrolysis. *Biotechnology and Bioengineering Symposium* 11, 67–77.

Knox, A. (2005). An Overview of Incineration and EFW Technology as Applied to the Management of Municipal Solid Waste (MSW) (PDF). University of Western Ontario.

Kofoworola, O. F. (2007). Recovery and recycling practices in municipal solid waste management in Lagos, Nigeria. *Waste Management*. 27, 1139- 1143.

Kong, F., Engler, C., & Soltes , E., (1992). Effects of cell-wall acetate, xylan backbone and lignin on enzymatic hydrolysis of aspen wood.*Applied Biochemistry and Biotechnology*, 34–35, 23–35.

Konya, R. S., Zitte, L. F., & Ugwulor, Q. N. (2013). Characterization of waste and their recycling potentials: A case study of East-West road, Port Harcourt. *Journal of Applied Science and Environmental Management*. 17(2), 233-238.

Koppar, A., & Pullammanappallil, P. (2008). Single-stage, batch, leach-bed, thermophilic anaerobic digestion of spent sugar beet pulp. *Bioresources technology*, 99 (8), 2831-2839

Kossmann, W., Pontz, U., Habermehl, S., & Hoerz, T. (2001). Biogas Digest, VolII: Biogas – Application and product Development, 4 - 11, 19 -25 and 25 -80 http://www.gtz.de/ de/dokumente/en- biogas–vol .2pdf

Kozlowski, K., Pietrzykowski, M., Czekala, W., Dach, J., Kowalczyk-Jusko, A., Jozwiakowski, K., & Brzoski, M., (2019). Energetic and economic analysis of biogas plant with using the dairy industry waste. *Energy Rep*, 183, 1023–1031.

Kumar, P., Barrett, D. M., Delwiche, M. J., & Stroeve, P., (2009). Methods for Pretreatment of Lignocellulosic Biomass for Efficient Hydrolysis and Biofuel Production, *Industrial Energy Chemistry Resources* 48 (8), 3713-3729. http://dx.doi.org/10.1021/ie801542g

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. *Atmospheric Environ*ment, 38, 4921– 4929.

Li, J., Zhang, R., Siddhu, M. A. H., He, Y., Wang, W., Li, Y., Chen, C., & Liu, G. (2015). Enhancing Methane Production of Cornstover Through a Novel Way: Sequent Pretreatment of Potassium Hydroxide and Steam Explosion,

Bioresource Technology, 181(4), 345-350. http://dx.doi.org/10.1016/j.biortech.2015.01.050.

Li, W., Siddhu, M. A., Amin, F. R., He, Y., Zhang, R., Liu, G., & Chen, C. (2018). Methane production through anaerobic co-digestion of sheep dung and waste paper. *Energy Conversion and Management*, 15(156), 79-87.

Li, Y., Zhang, D., Li, G., Lu, J., & Li, S. (2016). Solid state anaerobic co-digestion of tomato residues with dairy manure and corn stover for biogas production. *Bioresource technology*, 1(217), 50-56

Linke B., Heiermann M., &Mumme, J. (2006). Results of monitoring the pilot plants Pirow and Clausnitz. In: Rohstoffe, F.N. (Ed.), Solid-State Digestion–State of the Art and Further R&D Requirements, (24), 112–130.

Lo, H. M., Kurniawan, M. E. T., Sillanpaa, T. Y., & Chiang, C. F. (2010). Modelling biogas production from organic fraction of MSW co-digested with MSWI ashes in anaerobic bioreactors. *Bioresources Technology*, 101, 6329-6335.

Maishanu, S. M., Musa, M., & Sambo, A. S. (1990). Biogas technology: The output of the Sokoto Energy Research Centre, *Nigeria Journal of. Solar Energy*, 9, 183 – 194.

Mano Esteves, E. M., Naranjo Herrera, A. M., Peçanha Esteves, V. P., & Morgado, C. R. V. (2019). Life cycle assessment of manure biogas production: A review. *Journal of Cleaner Production* 219, 411–423. https://doi.org/10.1016/j.jclepro.02.091

Maramba, F. (1978), Production and Utilization of Biogas in Rural Areas of Industrialized and Developing Countries, Schriftenreihe der gtz, 97, 63, after; Biogas and Waste Recycling - The Phillipine Experience; Metro Manila, Phillipines.

Massé, D. I., Talbot, G., & Gibert, Y. (2011). In farm biogas production: A method to reduce GHG emissions and develop more sustainable livestock operations, *Animal Feed Science and Technology,* 166, 436-445.

Mata-Alvarez, J., Mace, S., & Llabres, P. (2003). Anaerobic digestion of organic solid wastes. An overview of research achievements and perspectives. *Bioresource Technology*, 74 (1), 3-16.

McKendry P. (2002). Energy production from biomass (part 1): overview of biomass, *Bioresource Technology*, 83(1), 37-46.

McMillan, J D. (1994). Pretreatment of Lignocellulosic Biomass, In *Enzymatic Conversion of Biomass for Fuels Production*, 292-324, American Chemical Society, Washington DC.

Monnet, F. (2003). An Introduction to Anaerobic Digestion of Organic Waste, being a Final Report Submitted to Remade, Scotland available on http://www.remade.org.uk/media/9102/an introduction to anaerobic digestion Accessed on 30 May, 2019.

Mood, S. H., Golfeshan, A. H., Tabatabaei, M., & Jouzani, G. S. (2013). Lignocellulosic biomass to bioethanol, a comprehensive review with a focus on pretreatment. *Renewable Sustainable Energy Revelution*, 27, 77–93.

Mshandete, A. M., & Parawira, W. (2009). Biogas Technology Research in Selected Sub-Saharan Africa. *African Journal of Biotechnology*, 8(2), 116-125. Available online at http://www.academicjournals.org/AJB. (Accessed 10 September, 2018).

Mukumba, P., & Makaka, G. (2015). Biogas production from a field batch biogas digester using cow dung as a substrate. *International Journal of Engineering Science and Technology*, 4, 1–10.

Murphy J., Braun R., Weiland P and Wellinger A (2011). Biogas from Crop Digestion. IEA Bioenergy Task 37 publication. Available from http://www.iea-biogas.net_download/publi-task37/Update_Energy_crop_2011.pdf (Accessed 19 January, 2018).

Musa, B., & Raji, H. M. (2016). Quantitative and Qualitative Analysis of Biogas Produces from Three Organic Wastes. *International Journal of Renewable Energy Research*, 6, 299-305.

Niemistö, J., Saavalainen, P., Pongrácz, É., & Keiski, P. L. (2013). Assessment of Lignocellulosic and Waste-based Feedstocks, *Journal of Sustainable Development of Energy, Water and Environment Systems*, 1(2), 58-77. http://dx.doi.org/10.13044/j.sdewes.01.0005 (Accessed 10 September, 2018).

Nigerian National Petroleum Corporation (NNPC), (2005). Nigeria to earn US$150m from Biofuels initiatives. http://www.nnpc.group.com/news/ biofuels.html. Retrieved 14/06/2018.

Ngumah, C. C. (2013). Biogas potential of organic waste in Nigeria. *Journal of Urban and Environmental Engineering*, 7(1).

Nwaigwe, K. N., & Enweremadu, C. C. (2015). Analysis of chemical oxygen demand (COD) removal rate using upflow bioreactor with central substrate dispenser (UBCSD). In Proc.4th International Conference on Advances in Engineering Sciences and Applied Mathematics, 67–70. Malaysia.

Nwankwo, C. S., Eze, J. I., & Okoyeuzu, C. (2017). Design and Fabrication of 3.60m^3 Household Plastic Bio Digester Loaded with Kitchen Waste and Cow

Dung for Biogas Generation. *Scientific Research and Essays*, 12, 130-141. https://doi.org/10.5897/SRE2017.6516

Nwosu, C., Omokhudu, G., & Ahijo, T. (2016). An Assessment of Open Dumps and Landfill Management in the Federal Capital Territory, Nigeria- Using Scotland as a Case Study for Structural Development. *Journal of Environment and Earth Science*. 6 (7), 78-91.

Obasioha, B. O. (2015). Waste Management Progress in Nigeria's Delta State. www.bioenergyconsult.com/waste-management/Nigeria.

Obileke, K., Mamphweli, S., Meyer, E., Makaka, G., Nwokolo, N., & Onyeaka, H. (2019). Comparative study on the performance of an above ground and underground fixed dome biogas digester. *Chemical Engineering Technology*, 43, 68–74.

Ofoefule, A. U., Nwanko, J. I., & Ibeto, C. N. (2008). Biogas production from paper waste and its blend with cow dung. *Advances in Applied Science Research.* 1(2):1-8.

Ogwueleka, T. C. (2009). Municipal solid waste characteristics and management in Nigeria. *Iran journal of environmental health science and engineering*. 6 (3), pp. 173-180.

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, Switzerland online at http://www.scientific.net(Accessed 10 September, 2018).

Okewale, A. O., Omoruwuo, F., & Ojaigho, R. O. (2016). Alternative Energy Production for Environmental Sustainability. *British Journal of Renewable Energy*, 1, 18-22. https://doi.org/10.1051/rees/2016040

Okewale, A. O., Omoruwou, F., & Anih, C. E. (2018). Production of Biogas from Co-Digestion of Cow Dung, Saw Dust and Maize Husk. *Advances in Chemical Engineering and Science*, 8, 113-123. https:// doi.org/ 10.4236/ aces.2018.83008

Okoro- Shekwaga, K. C., & Horan, N. (2015). Suitability of Anaerobic digesters for West Africa: Nigeria a case study. *International journal of Environmental Science and Development*. 6 (2), 155-164.

Otitoju, T. A. (2014). Individual attitude towards recycling of municipal solid waste in Lagos, Nigeria. *American Journal of Engineering Research.* (7), 77-88.

Otun, T. F., Ojo, O. M., Ajibade, F. O., & Babatola, J. O. (2015). Evaluation of Biogas Production from the Digestion and Co-Digestion of Animal Waste, Food Waste and Fruit Waste. *International Journal of Energy and Environmental Research*, 3, 12-24.

Oumarou, M. B., Dauda, M., Abdulrahim, A. T., & Abubakar, A. B. (2012). Municipal solid waste generation, recovery and recycling: A case study. *World journal of Engineering and Pure and Applied Sciences*. 2(5), 143.

Page, D. I., Hickey, K. L., Narula, R., Main, A. L., & Grimberg, S. J. (1982). Modeling anaerobic digestion of dairy manure using the IWA anaerobic digestion model No. 1 ADM1. *Water Science & Technology*. 58(3), 1-8.

Palmowski, L. M., & Müller, J. A. (2000). Influence of the size reduction of organic waste on their anaerobic digestion, *A Journal of International Association on Water Pollution Research 41*, 155-162.

Patinvoh, R. J. (2017). Dry fermentation of manure with straw in continuous plug flow reactor: Reactor development and process stability at different loading rates.*Bio resources. Technology*,224 197-205.

Pearson, D. (1976). *The Chemical Analysis of Food*. (7th Edn.). Churchill Livingstone. New York, 11-12, 14-15.

Phillips, J., & Williams, I. D. (2022). Leading the World: *A Review of Household Recycling in Wales*. Recycling, *7*, 46. https://doi.org/ 10.3390/ recycling7040046.

Rabah, A. B., Baki, A. S., Hassan, L. G., Musa, M., & Ibrahim, A. (2010). Production of *Biogas* using abattoir waste at different retention. *Science World Journal*, 5 (4), 23-26.

Ramos, L. P. (2003). The chemistry involved in the steam treatment of lignocellulosic materials, *Química Nova 26*, 863-871.

Renewable Energy Policy Network for the 21st Century (REN21). (2017.). Renewables Global Status Report (Paris: REN21 Secretariat) ISBN 978-3-9818107-6-9.

Richards, B., Cummings, R., White, T., & Jewell, W. (2010). Methods for kinetic analysis of methane fermentation in high solids biomass digesters.*Biomass and Bioenergy*,1(2), 65 – 66.

Rowell, R. M., Pettersen, R., & Tshabalala, M. A. (2012). 3 Cell wall chemistry. In: Rowell R (ed) Handbook of wood chemistry and wood composites. CRC Press, Boca Raton, FL, 33.

Rowell, R. M., Pettersen, R., Han, J. S., Rowell, J. S., & Tshabalala, M. A., (2005). Cell wall chemistry, Handbook of wood chemistry and wood composites, CRC Press, Florida, USA, 35-74.

Sangodoyin, A. Y. (2017). Considerations on contamination of groundwater by waste disposal systems in Nigeria. *Environmental Technology*, 14(10), 957-964.

Sambo, A. S. (2010). Renewable Energy Development in Nigeria: http://www.worldfuturecouncil.org/fileadmin/user_upload/PDF/PkfAE._Bala-Nigerian_Energy_Commission_Renewal_Energy_Development_In_Nigeria.pdf. Accessed April 2020.

SECOT Co. Ltd. (2009). Environmental and health impact assessment (main report): Bang Pakong Power Plant Unit 1-2 Replacement Project (updated project description-version) (in Thai). Bangkok.

Sleat, R. & Mah, R. (2006). Hydrolytic bacteria. In: Anaerobic Digestion of Biomass. *Renewable Energy*, 15.

Stichting Nederlandse Vrijwilligers (SNV). (2017). Africa's largest biogas and clean cooking convention: Fuelling a silent green revolution. http://www.snv.org/update/africas-largest-biogas-and-clean-cooking-convention-fueling-silent-green-revolution. Accessed July 2019.

Söderström, J., Pilcher, L., Galbe, M., & Zacchi, G. (2003). Two-step Steam Pretreatment of Softwood by Dilute H_2SO_4 Impregnation for Ethanol Production, *Biomass and Bioenergy*, 24(6), 475-486. http://dx.doi.org/10.1016/S0961-9534(02)00148-4.

Somayaji, D., & Khanna, S., (1994). Biomethanation of rice and wheat straw.*World Journal of Microbiology Biotechnology*, 10(5), 521–523.

Song, Y. C., Kwon, S. J., & Woo, J. H. (2004). Mesophilic and thermophilic temperature co- phase anaerobic digestion compared with single-stage mesophilic and thermophilic digestion of sewage sludge, *Water Resources*,38(7), 1653 – 1662.

Song, Z., & Zhang, C. (2015). Anaerobic codigestion of pretreated wheat straw with cattle manure and analysis of the microbial community. *Bioresource technology*, 186, 28-35.

Spore, (2006). Information for Agricultural development in ACP countries. CTA Issue, 124(6), 23-27.

Sridhar, M. K. C., & Hammed, T. B. (2014). Turning waste to wealth in Nigeria: An overview. *Journal of Human Ecology*,46(2), 195-203.

Stanley, H. O., Umolo, E. A., & Stanley, C. N. (2011). Cultivation of oyster mushroom (Pleurotus pulmonarius) on amended corncob substrate. *Agriculture and Biology Journal of North America*, 2(8), 1239 - 1243.

Steffen, R., Szolar, O., & Braun, R. (1998). Feedstocks for Anaerobic Digestion. Available: http://www.adnett.org/dl_feedstock.pdf (Accessed 19 January, 2018).

Sun, Y., & Cheng, J. (2002). Hydrolysis of lignocellulosic materials for ethanol production: a review, *Bioresource Technology*, *83*, 1-11.

Taherzadeh, M., & Karimi, K. (2008). Pretreatment of Lignocellulosic Wastes to Improve Ethanol and Biogas Production: A Review. *International Journal of Molecular Sciences*, 9(9), 1621.

Taherzadeh, M. J., & Rajendran, K. (2014). *Factors affecting development of waste management.* Waste Management and Sustainable Consumption: Reflections on Consumer Waste, 67.

Taphee, G. B., & Jongur A. A. U. (2014). Productivity and efficiency of groundnut farming in Northern Taraba State. *Nigerian Journal of Agriculture and Sustainability*, 5(1), 45-56.

Themelis, N. J., & Verma, S. (2013). "The Better Option: Anaerobic Digestion of Organic Waste in MSW." Waste Management World January/February 2004.

Tyagi, V., Panwar, N., Rahim, N., & Kothari, R. (1981). Review on solar air heating system with and without thermal energy storage system. *Renew Sustain Energy Revolution,*16(4), 2289–303.

Ukpai, P. A., & Nnabuchi, M. N. (2012). Comparative study of biogas production from cow dung, cow pea and cassava peeling using 45 litres biogas digester. *Advances in Applied Science Research,*3(3), 1864-1869.

Umaru, I. (2010). Recycling of solid waste and the "Yan Bola" Underground Economy: A survey of Environmental Entrepreneurs in Central Nigeria. *Journal of Human Ecology*, 30 (1), 45-54.

UN (2007). United Nations, *The Millennium Development Goals Report* 2006. Published by United Nations Department of Economic and Social Affairs. New York, NY.

Uwaegbulam C. 2017. Enugu begins solid waste reforms, partners private sector. https://guardian.ng/property/enugu-begins-solid-waste-reform-partners-private- sector/. Accessed December 2019.

Vandevivere P., De Baere, L., & Verstraete, W. (2002). Types of anaerobic digesters for solid wastes, in Biomethanization of the Organic Fraction of Municipal

Solid Wastes, J. MataAlvarez, Editor. 2002, IWA Publishing: Barcelona. 111-140

Varga, E., Reczey, K. & Zacchi, G. (2004). Optimization of Steam Pretreatment of Corn Stover to Enhance Enzymatic Digestibility, *Applied Biochemical and Biotechnology*, 113-116, 509-23. http://dx.doi.org/10.1385/ABAB:114:1-3:509

Vivekanand, V., Olsen, E. F., Eijsink, V. G., &Horn, S. J. (2013). Different steam explosion conditions on methane potential and enzymatic saccharification of birch. Bio resources. Technology, 127, 343–349.

Walpot, J. I. (1986). Enzymatic hydrolysis of waste paper, *Conservation & Recycling 9*, 127-136.

Wang, X. T., Cai, H. Z., & Liu, L. S. (2011). Experimental Study on Anaerobic Fermentation of Steam Explosion Pretreated Corn Stalk, *Advanced Materials Research*, 183-185, 1975-1978. http:// dx.doi.org/ 10.4028/ www.scientific.net/AMR.183-185.1975

Wang, Z., ZheLv, Z., Du, J., Mo, C., Yang, X. & Tian, S. (2014). Combined Process for Ethanol Fermentation at High-solids Loading and Biogas Digestion from Unwashed Steam-exploded Corn Stover, *Bioresource Technology*, 166, 282-287. http://dx.doi.org/10.1016/j.biortech.2014.05.044.

Ward, A. J., Hobbs, P. J., Hollimana, P. J., & Jones, D. L. (2008). Optimisation of the anaerobic digestion of agricultural resources.*Bioresource Technology*, 99(17), 7928-7940.

Wei, Y., Li, X., Yu, L., Zou, D., & Yuan, H. (2015). Mesophilic Anaerobic Co-digestion of Cattle Manure and Cornstover with Biological and Chemical Pretreatment, *Bioresource Technology*, 198, 431-436. http:// dx.doi.org/ 10.1016/j.biortech.09.035

Weiland, P. (2010). Biogas production: current state and perspectives. *Applied microbiology and biotechnology*, 85(4), 849-860.

World Bank, (2015). *The Little Green Data Book.*Washington, DC: World Bank.10.1596/978-1-4648-0560-8. License: Creative Commons Attribution CC BY 3.0 IGO.

World Bank, (2016). *The Little Green Data Book*Washington, DC: World Bank.10.1596/978-1-4648-0560-8. License: Creative Commons Attribution CC BY 3.0 IGO.

Xu, G. Z., Fan, S. Y., Zhang, B. L., & Liu, J. B. (2012). Anaerobic Fermentation Characteristics of Corn Straw Pretreated by Steam Explosion, *Advanced Materials Research*, 512-515, 334-337, http://dx.doi.org/ 10.4028/ www.scientific.net/AMR.512-515.334

Yang, B., & Wyman, C. E. (2004). Effect of xylan and lignin removal by batch and flowthrough pretreatment on the enzymatic digestibility of corn stover cellulose, *Biotechnology and Bioengineering 86*, 88-98.

Yavini, T. D., Taura, U. H., Mohammed, N., & Namo, J. M. (2014). Comparative study of mesophilic biogas production potentials of selected agro-wastes. *The International Journal of Engineering and Science,* 3(2), 1-6.

Yashvant, P., Ram, N., & Singh, V. K. (2012). Medical properties of Pleurotus Species (Oyeter mushroom): A Review. *World Journal of Fungal and Plant Biology*, 3(1), 01-12.

Yawalker, K. S., Agarwal, J. P., & Bodke, S. (1977). *Manures and Fertilizers*; Agriculture and Horticultural Publishing house, Nagpur, India (4th Edition).

Yusuf, M. O. L., Debora, A., &Ogheneruona, D. E. (2011). Ambient temperature kinetic assessment of biogas production from co-digestion of horse and cow dung. *Resources of Agriculture Eng*ineering, 57(3), 97-104.

Zarkadas, I. S., Sofikiti, A. S., Voudrias, E. A., & Pilidis, G. A. (2015). Thermophilic anaerobic digestion of pasteurised food wastes and dairy cattle manure in batch and large volume laboratory digesters: Focusing on mixing ratios. *Renewable Energy*, 80, 432-440.

Zwietering, M. H., Jongenburger, I., Rombouts, F. M., & Van't Riet K. (1990). Modeling of the bacterial growth curve. *Applied and Environmental Microbiology*, *56*, 187.

Appendices

Appendix A: Autoclaving of Groundnut Shell

Appendix B: Taken the weight of column bag filled with the substrate

Appendix C: Mushroom Spawn and Inoculation of the column bags with the spawn

Appendix D: Sprinkling of Water

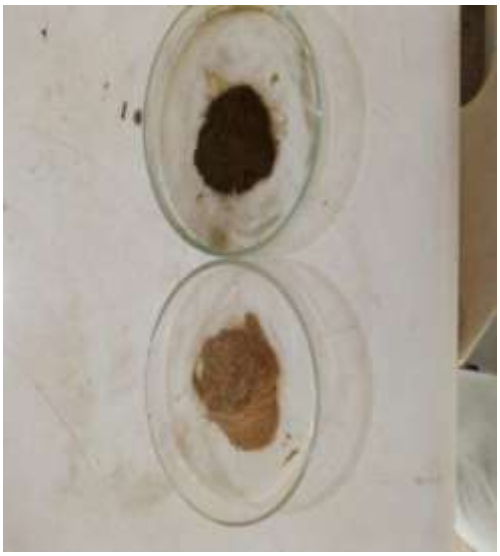
Appendix E: Loading of the Digesters

Appendix F: Digesters

Appendix G: Determination of VS: muffle furnace

Appendix H: Taken the weight of the samples for C and N determination

Appendix I: Determination of Nitrogen



Appendix J: Determination of Nitrogen- Digestion Apparatus

Appendix K: Water bath

Appendix L: pH Determination

Appendix M: Sample Calculations for Proximate Analysis

1. Moisture Content (MC)

$$\%MC = \frac{W_1 - W_2}{W_1} \times \frac{100}{1} \quad \text{(M1)}$$

Where

W_1 = wt of the original sample

W_2 = wt of final dried sample

$$MC = \frac{40.44 - 4.20}{40.44} \times 100 = 89.50$$

2 Determination of Total Solids (TS)

$$\%TS = \frac{W_1 - W_2}{W_3 - W_2} \times 100 \quad \text{(M2)}$$

Where

%TS = Percentage total solid

W_1 = Weight of dried crucible + dried residue

W_2 = Weight of crucible

W_3 = Weight of wet sample (substrate) + crucible

$$TS = \frac{47.76 - 44.80}{54.75 - 44.80} = \frac{1.96}{9.95} \times 100 = 19.61$$

3 Determination of Volatile Solids (VS)

$$\%VS = \frac{W_1 - W_4}{W_1 - W_2} \times 100 \quad \text{(M3)}$$

Where,

$\%VS$ = Percentage Volatile solid

W_4 = Weight of crucible + weight of residue after ignition

$$VS = \frac{47.75 - 46.28}{47.50 - 44.80} \times 100 = \frac{1.47}{2.7} \times 100 = 54.44$$

Calculation for Ultimate Analysis

1 Determination of Carbon Content

$$\% \text{ carbon} = \frac{B - T \times 133 \times 0.003 \times 100}{W} \quad \text{(M4)}$$

Where;

B = Blank titre value

T = Sample Titre value

C = Concentration of Fe solution

W = Weight of waste sample

2 Chemical Oxygen Demand (COD)

$$\% \text{ nitrogen (N)} = \frac{\text{Titre} \times 0.0014 \times 250}{\text{Weight of original sample}} \times 100 \quad \text{(M5)}$$

Appendix N: pH of the Slurry before and after Biogas production

Digester	pH before Biogas production	pH after Biogas production
A	7.00	7.20
B	6.90	6.70
C	7.01	6.90
D	7.11	6.70
E	7.30	7.30
F	6.90	7.40
G	7.10	7.11
H	7.15	6.90
I	7.20	6.80
J	6.70	7.00

Appendix O: pH of the Digesters

Time (Days)	Steam Exploded					Non Exploded				
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**I**	**J**
1	7.2	6.9	7	6.8	6.9	6.9	7	7	6.8	6.9
2	7.4	7.1	7.1	7.1	7	6.8	6.9	6.9	7	7
3	7.2	6.9	7	6.8	7	6.9	7	6.9	7	7
4	6.9	7	6.8	6.9	6.9	6.7	7	6.8	6.9	6.8
5	7.6	7.1	7	6.8	7.3	6.9	7	6.8	7	6.9
6	7	6.9	7.4	7.2	6.9	6.9	7	6.9	7	7.3
7	7.3	7.1	7.2	6.9	7.2	7.4	6.9	7	6.8	6.9
8	7.1	6.9	7	6.8	6.8	7.1	6.9	7.3	7.2	7
9	7.1	7	6.9	7.2	6.9	6.9	6.9	7	7	7
10	6.9	7.2	7.6	7.2	6.6	7	7.4	6.9	7	6.8
11	7.5	7.2	6.9	7	6.8	7	7.1	6.9	7	6.2
12	6.9	7	6.9	6.9	7	7.1	7.2	6.9	6.5	6.9
13	6.9	7.3	7	7.1	7	6.4	6.9	7	6.8	7
14	7.1	6.9	7	7.2	6.9	6.9	7.2	7.5	7.5	7
15	7.2	6.9	7.4	7.1	6.9	7.1	6.9	7.1	6.9	7
16	7.2	7.6	7	6.9	6.9	6.4	6.5	7.1	6.9	6.9
17	7	6.9	7.2	7.5	7.2	6.9	7	6.8	7	7
18	7.1	7	7.2	7.6	7.2	7.1	6.9	7	7	7.1
19	7.4	6.5	**6.2**	7.1	6.9	7	6.9	6.9	7.2	7.5
20	7	7.1	6.9	6.9	6.9	6.9	7	7.4	6.9	6.9
21	7.1	6.9	7	7	7	7	7	6.8	7	7
22	6.9	6.9	7.2	7.5	7	6.9	6.7	6.8	7	6.9
23	7	7.5	7.1	6.9	6.9	7	6.9	7	6.8	7
24	7	7.1	6.9	7	7	7	7	6.9	7.2	7.5
25	7.4	6.2	6.9	7.2	7.5	7.1	6.9	7	**7.6**	6.9
26	7	7.1	6.9	6.9	7.1	6.9	7	7.1	6.9	7
27	7.1	6.9	7	7	6.9	7.4	6.9	7.2	7.5	7
28	6.9	7.2	7.5	7	7	7	7.4	7.4	6.9	7.4
29	7.4	6.9	7.2	7.5	7.1	6.9	7	7	7	7
30	7	6.9	7.2	7.5	7.5	6.9	7.2	7.5	7	7
Average	**7.13**	**7**	**7.05** **7.05**	**7.08**	**7.01**	**6.95**	**6.99**	**7.03** **6.99**	**7.01**	**6.99**

Appendix P: Digesters and Ambient Temperature (^{0}C)

Time (Days)	Steam exploded					Non exploded					Ambient
	A	B	C	D	E	F	G	H	I	J	
1	30	31.1	29	30	30.1	31	30	28.9	32	31.1	33
2	31.1	29.8	30.1	32	31.1	28.8	31	30	28.9	29.9	32
3	32.1	31.1	33	31.1	29.9	30.1	32	31.1	29,8	33	34
4	33.2	31.1	30.1	31	33	31	30	28.9	32	31.1	33.2
5	32.1	31.1	33.2	30.1	32	32.1	32	31	30	28.9	33.9
6	35	34.1	32.1	35.1	34	30.1	32	31.1	32.1	32	37
7	32	30	31.1	33.1	34	29.3	30.1	32	31.1	33	35.8
8	29	32.2	34	31	32	31	30	28.9	32	31.1	36
9	32	31.1	33	31.1	30.2	31	30	28.9	29.9	30.1	34.8
10	33	32.1	32.1	32	34	30.1	32	31.1	33	28.7	35
11	31.1	33.2	30.1	32	33	32.1	32	30.1	32	31.1	36.6
12	33.3	31.1	33.2	30.1	32	30.1	32	31.1	28.9	29.9	35
13	29.8	30.2	32	30	31.1	29.3	30.1	32	31.1	33	33
14	31.1	30.1	31	30	28.9	31	30	28.9	30	28.9	32.5
15	32.2	34	31	32	29.8	30.1	32	31.1	29.8	30.1	34.6
16	31.2	32	30	31.1	33.2	29.8	30.1	32	30	31.1	33
17	32.2	34	31	32.1	32.1	32	30	33.2	29.8	30.1	35
18	30.2	32	30	33.2	31.1	30	31.1	30.1	32	31.1	34
19	29.2	32	30	31.1	32	30	31.1	32	30	31.1	33
20	30.1	32	31.1	29.8	30.1	30	33.2	32	30	33.2	33
21	29.8	30.1	30	33.2	30.1	32	31.1	32	30	31.1	34
22	28.8	30	31.1	32	30	33.2	32	30	31.1	28.6	33.2
23	30.1	30.1	32	31.1	29.8	30.1	30	31.1	30	31.1	32
24	32	30	31.1	29.8	30.1	30	31.1	32	30	33.2	34
25	28.9	32	30	33.2	29.8	30.1	30	31.1	29.8	30.1	32
26	30	31.1	32	30	31.1	32	30	33.2	30.1	32	33.9
27	32	30	31.1	30	31.1	30.1	32	31.1	30	31.1	33.2
28	29.8	30.1	32	31.1	30	31.1	32	30	31.1	30.2	32
29	32	30	33.2	29.8	30.1	30	31.1	30.1	32	31.1	34
30	30.1	32	31.1	32	30	33.2	32	30	33.2	28.2	35
Aver.	**31.1**	**31.3**	**31.4**	**31.3**	**31.2**	**30.7**	**31.1**	**30.8**	**29.7**	**30.8**	
	31.26					**30.63**					**33.9**

Appendix Q: Biogas Production

Time (Days)	Gas Production (cm) in the gas holder									
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**I**	**J**
1	0	0	0	0	0	0	0	0	0	0
2	0.4	0.4	0.1	0	0	0.3	0	0	0	0
3	1	1.2	1..0	0.4	0.2	0.7	0.1	0	0	0
4	1.3	1	1	0.6	0.2	1.2	0.1	0.1	0	0
5	1.3	0.9	0.9	0.4	0.1	1.2	0.1	0.1	0.1	0.1
6	1.2	1	0.9	0.4	0.2	0.9	0.1	0.1	0.1	0
7	1.5	1.1	1	0.4	0.2	1.3	0.2	0.3	0.1	0.1
8	1.5	1.1	0.9	0.3	0.3	1.2	0.2	0.3	0.1	0.1
9	1.5	1.1	1	0.5	0.3	1.3	0.4	0.5	0.3	0.3
10	1.7	1.3	1..0	0.5	0.3	1.5	0.4	0.4	0.2	0.3
11	1.7	1.2	1.1	0.5	0.3	1.5	0.4	0.3	0.1	0.2
12	1.7	1.3	1.2	0.5	0.3	1.5	0.8	0.5	0.5	0.2
13	1.7	1.2	1	0.6	0.2	1.2	0.9	0.5	0.4	0.2
14	1.7	1.4	1	0.6	0.3	1.6	1.3	1	0.5	0.1
15	1.6	1.4	1	0.6	0.2	1.8	1.3	1	0.5	0.2
16	1.9	1.6	1.7	0.8	0.5	1.7	1.3	1.1	0.4	0.2
17	2.3	1.6	1.4	0.6	0.5	1.9	1.4	1.1	0.6	0.3
18	2.1	1.6	1.7	1	0.4	1.9	1.4	1.1	0.6	0.2
19	2.4	1.5	1.7	1.2	0.4	2.2	1.4	0.9	0.5	0.2
20	2.4	1.5	1.6	1.3	0.4	2	1.3	1	0.5	0.2
21	2.4	1.1	1	0.5	0.3	2	1.1	0.9	0.5	0.1
22	2.3	0.8	1	0.5	0.3	1.6	1	0.9	0.3	0.1
23	2.1	0.8	1	0.5	0.3	1.5	0.7	0.9	0.2	0.1
24	1.6	0.8	0.7	0.3	0.3	1.3	0.4	0.7	0.2	0.1
25	1.1	0.3	0.4	0.3	0.1	0.9	0.3	0.3	0.2	0.1
26	0.9	0.3	0.3	0.3	0.1	0.4	0.2	0.2	0.1	0.1
27	0.4	0.1	0.2	0.1	0.1	0.3	0.2	0.1	0.1	0.1
28	0.2	0.1	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1
29	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
30	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Average	**1.41** **0.77**	**0.93**	**0.81**	**0.47**	**0.24**	**1.24** **0.66**	**0.58**	**0.49**	**0.27**	**0.13**

Appendix R: Volume of Biogas Production (cm^3)

Time (Days)	Steam exploded					Non exploded				
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**I**	**J**
1	0	0	0	0	0	0	0	0	0	0
2	15.39	15.39	3.85	0	0	11.54	0	0	0	0
3	38.48	46.18	38.48	15.39	7.7	26.94	3.85	0	0	0
4	50.02	38.48	38.48	23.09	7.7	46.18	3.85	3.85	0	0
5	50.02	34.63	34.63	15.39	3.85	46.18	3.85	3.85	3.85	3.85
6	46.18	38.48	34.63	15.39	7.7	34.63	3.85	3.85	3.85	0
7	7.72	42.33	38.48	15.39	7.7	50.02	7.7	11.54	3.85	3.85
8	57.72	42.33	34.63	11.54	11.54	46.18	7.7	11.54	3.85	3.85
9	57.72	42.33	38.48	19.24	11.54	50.02	15.39	19.24	11.54	11.54
10	65.42	50.02	38.48	19.24	11.54	57.72	15.39	15.39	7.7	11.54
11	65.42	46.18	42.33	19.24	11.54	57.72	15.39	11.54	3.85	7.7
12	65.42	50.02	46.18	19.24	11.54	57.72	30.78	19.24	19.24	7.7
13	65.42	46.18	38.48	23.09	7.7	46.18	34.63	19.24	15.39	7.7
14	65.42	53.87	38.48	23.09	11.54	61.57	50.02	38.48	19.24	3.85
15	61.57	53.87	38.48	23.09	7.7	69.26	50.02	38.48	19.24	7.7
16	73.11	61.57	65.42	30.78	19.24	65.42	50.02	42.33	15.39	7.7
17	88.5	61.57	53.87	19.24	11.54	73.11	53.87	42.33	23.09	11.54
18	80.81	61.57	65.42	38.48	15.39	73.11	53.87	42.33	23.09	7.7
19	92.35	57.72	65.42	46.18	15.39	84.66	53.87	34.63	19.24	7.7
20	92.35	57.72	61.57	50.02	15.39	76.96	50.02	38.48	19.24	7.7
21	92.35	42.33	38.48	19.24	11.54	76.96	42.33	34.63	19.24	3.85
22	88.5	30.78	38.48	19.24	11.54	61.57	38.48	34.63	11.54	3.85
23	80.81	30.78	38.48	19.24	11.54	57.72	26.94	34.63	7.7	3.85
24	61.57	30.78	26.94	11.54	11.54	50.02	15.39	26.94	7.7	3.85
25	42.33	11.54	15.39	11.54	3.85	34.63	11.54	11.54	7.7	3.85
26	34.63	11.54	11.54	11.54	3.85	15.39	7.7	7.7	3.85	3.85
27	15.39	3.85	7.7	3.85	3.85	11.54	7.7	3.85	3.85	3.85
28	7.7	3.85	7.7	3.85	3.85	3.85	3.85	3.85	3.85	3.85
29	7.7	3.85	3.85	3.85	3.85	3.85	3.85	3.85	3.85	3.85
30	7.7	3.85	3.85	3.85	3.85	3.85	3.85	3.85	3.85	3.85
Average	**52.59**	**37.79**	**33.61**	**17.83**	**9.52**	**45.15**	**22.19**	**18.73**	**9.49**	**5**
			30.27					**20.11**		

Appendix S: Cumulative Biogas Production (cm^3)

Time (Days)	Steam exploded					Non exploded				
	A	B	C	D	E	F	G	H	I	J
1	0	0	0	0	0	0	0	0	0	0
2	15.39	15.39	3.85	0	0	11.54	0	0	0	0
3	53.87	61.57	42.33	15.39	7.7	38.48	3.85	0	0	0
4	103.89	100.05	80.81	38.48	15.4	84.66	7.7	3.85	0	0
5	153.91	134.68	115.44	53.87	19.25	130.84	11.55	7.7	3.85	3.85
6	200.09	173.16	150.07	69.26	26.95	165.47	15.4	11.55	7.7	3.85
7	207.81	215.49	188.55	84.65	34.65	215.49	23.1	23.09	11.55	7.7
8	265.53	257.82	223.18	96.19	46.19	261.67	30.8	34.63	15.4	11.55
9	323.25	300.15	261.66	115.43	57.73	311.69	46.19	53.87	26.94	23.09
10	388.67	350.17	300.14	134.67	69.27	369.41	61.58	69.26	34.64	34.63
11	454.05	396.35	342.47	153.91	80.81	427.13	76.97	80.8	38.49	42.33
12	519.51	446.37	388.65	173.15	92.35	484.85	107.75	100.04	57.73	50.03
13	584.93	492.55	427.13	196.24	100.05	531.03	142.38	119.28	73.12	57.73
14	650.35	546.42	465.61	219.33	111.59	592.6	192.4	157.76	92.36	61.58
15	711.92	600.29	504.09	242.42	119.29	661.86	242.42	196.24	111.6	69.28
16	785.03	661.86	569.51	273.2	138.53	727.28	292.44	238.57	126.99	76.98
17	873.53	723.43	623.38	292.44	150.07	800.39	346.31	280.9	150.08	88.52
18	954.34	785	688.8	330.92	165.46	873.3	400.18	323.23	173.17	96.22
19	1,046.69	842.72	754.22	377.1	180.85	958.16	454.05	357.86	192.41	103.92
20	1,139.04	900.44	815.79	427.12	196.24	1,035.12	504.07	396.34	211.83	111.62
21	1,231.39	942.77	854.27	446.36	207.78	1,112.02	546.4	430.97	231.07	115.47
22	1,319.89	973.55	892.75	465.6	219.32	1,173.65	584.88	465.6	242.61	119.32
23	1,400.70	1,004.33	931.23	484.84	230.86	1,231.37	611.82	500.23	250.31	123.17
24	1,462.27	1,035.11	958.17	496.38	242.4	1,281.39	627.21	527.17	258.01	127.02
25	1,504.60	1,046.65	973.56	507.92	246.25	1,316.02	638.75	538.71	265.71	130.87
26	1,539.23	1,058.19	985.1	519.46	250.01	1,331.41	646.45	546.41	269.56	134.72
27	1,554.60	1,062.02	992.8	523.31	253.95	1,342.92	654.15	550.26	273.41	138.57
28	1,562.32	1,065.89	1,000.50	527.16	257.8	1,346.80	658	554.11	277.26	132.42
29	1,570.02	1,069.74	1,004.35	531.01	261.65	1,350.65	661.85	557.96	281.11	146.27
30	1,577.72	1,073.59	1,008.20	534.86	265.5	1,354.50	665.7	561.81	284.96	150.12