Effect of Co-Digestion of Leachate and Cotton Gin on The Production of Biogas from Cow Dung

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APPROVAL SHEET

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DECLARATION

It is hereby declared that this research study has been done for partial fulfillment of requirements for the degree of Masters of Sciences in Environmental Sciences. This work has not been taken from any publication. I hereby also declare that no portion of the work referred to in this thesis has been submitted in support of an application for another degree or qualification in this university or other institute of learning.

Mehak Fatima

"Every breath I take in my life and every little drop of blood running in my body, are dedicated to my family to whom I owe everything"

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"In the name of Allah the most Merciful and Beneficent"

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LIST OF ABBREVIATIONS

AEBD	Alternate Energy Development Board
PCRET	Pakistan Council for Renewable Energy Technologies
NGO	Non-Governmental Organizations
COD	Chemical Oxygen Demand
BTU	British Thermal Unit
NRCS	Natural Resource Conservation Service
MSW	Municipal Solid Waste
C: N	Carbon to Nitrogen Ratio
ICARDA	International Center for Agriculture Research in Dry Areas
VS	Volatile Solids
TS	Total Solids
GC-MS	Gas Chromatography Mass Spectrometry

ABSTRACT

Pakistan has been facing major hindrance in the economic development due to the energy crises. Luckily, Pakistan has massive biomass resources available as crop residues, food waste, sugarcane bagasse, wood, industrial waste, dung, feces, and poultry litter etc. which could be used to produce biogas, an alternative renewable fuel. The current research was conducted to study the potential to produce biogas from cow dung with the co-digestion of cotton gin with the objective of increasing the biogas production through the addition of municipal solid waste leachate in different proportion under mesophilic conditions. For this reason, batch digestion was performed in the laboratory of IESE, NUST. In each case six digesters having working volume of 1.2 liters were used. First three digesters were filled with cotton gin and cow dung in leachate and other three with cotton gin and cow dung in water. Same proportions (900ml) of liquid (leachate or water) was added in the digesters. The solid substrates (cotton gin and cow dung) were added in three different combinations (1:2.5, 1:5 and 1:7). To adjust the pH of digesters, Na_2CO_3 was used to buffer any sharp drop. Results showed that the highest percentage reduction in VS (80.3%) was found in the digesters containing cotton gin and cow dung filled as 1:2.5 in leachate thus producing maximum volume of gas. Combination 1 (1:1.25) cumulatively produced highest amount of biogas which was approximately 1350 ml/g of VS while digester with the same solid content ratios in water produced 1000ml/g of VS. Co-digestion in leachate and water resulted in 12.95 times and 10.83 times higher biogas production then raw (leachate and gin independently) respectively. Average methane content in biogas after attaining stability was 67.8% for combination 1 in leachate while 35.5% for the same combination in water. Percentage reduction in COD was highest in leachate. All three leachate samples showed

changes of 61.4% to 71.2% reduction of COD. From the results, it was concluded that leachate addition had a positive effect on biogas production from both type of solid waste but this effect was more considerable in the digester containing highest ratio of cotton gin.

Chapter 1

NTRODUCTION

1.1 Background

The dependence of growing population on energy and fuels has forced research and development organizations to investigate alternate energy resources. Coupled with the increasing threats of climate change, an effective energy resource is greatly needed. One of the many types of renewable energy that has been developed is converting biological materials into valuable fuels. This bioenergy can come in many forms that are comparable to the established fossil fuels in the modern market, and with the proper equipment, can be used as a sustainable replacement. A useful energy material is methane that is carbon-based gas primarily generated from biological reactions with microorganisms in oxygen deprived conditions through anaerobic digestion. Anaerobic digestion takes place when bacteria converts a biomass feedstock into various other organic compounds, ultimately ending in a mixture of carbon dioxide and methane, commonly called biogas. This mixture approximately composed of 40% carbon dioxide and 60% methane along with tiny fractions of trace gases. The anthropogenic carbon dioxide is a concern as greenhouse gas emissions but carbon dioxide released in this reaction is considered carbon neutral. Methane can further be purified and used for a purpose of generating heat or electricity (Ward et al., 2008). Anaerobic digestion serves a dual purpose in both providing methane and reducing volatile solids that lowers the risk of possible pollution at disposal. These solids can further be used as a soil amendment in agriculture.

1.2 Anaerobic Digestion

The process of anaerobic co-digestion involves mainly the conversion of volatile acids into methane. To achieve maximum biogas production efficiency, many of the reaction parameters that significantly affect the experimental results include temperature, pH, physical and chemical characteristics of substrates.

The main product of anaerobic digestion is methane. Methane is a useful energy material that is carbon-based primarily produced from biological reactions with microorganisms in oxygen deprived conditions through anaerobic digestion. Anaerobic digestion takes place when bacteria converts a biomass feedstock into various other organic compounds, ultimately ending in a mixture of carbon dioxide and methane, commonly called biogas. Biogas is a combination of approximately 40 % carbon dioxide and 60 % methane along with other gases present in traces. The anthropogenic carbon dioxide is a concern as greenhouse gas emissions but carbon dioxide released in this reaction is considered carbon neutral. Methane can be purified and used for a purpose of generating heat or electricity (Ward et al., 2008). Anaerobic digestion serves a dual purpose in both providing methane and reducing volatile solids that lowers the risk of possible pollution at disposal. These solids can further be used as a soil amendment in agriculture.

Co-digestion is a technique of combining multiple feed sources into the same anaerobic digestion system to increase overall methane content (Esposito et al., 2012). During the last two decades, different organic substrates have been studied showing synergic effect of the combined treatment (Macias-Corral et al., 2008; Esposito et al., 2011; Esposito et al.,2012). These co-substrates commonly include cotton wastes (Adl et al., 2012), organic solid waste (Esposito et al., 2011), sewage sludge, rice stalks (Iyagba et al., 2009), and Micro-algae (Perazzoli et al., 2016).

This method is currently being used worldwide with a variety of biodegradable substrates having different energy production potentials. The general objective is to maximize the production of biogas and meet today's energy demand through shifting to renewable energy however in this study a specific choice of low cost and free high energy potential substrates has been made. The study focuses on the potential of cotton gin for biogas production and municipal solid waste management through the use of leachate as a source of energy to reduce environment pollution. (Braun, 2002)

1.3 Use of Industrial Waste (Cotton Gin Trash)

Cotton waste is considered as an effective energy source, especially in countries wherce of energy among other agriculture waste products mainly in the parts of the world where cotton is grown massively such as China, Pakistan, US, Brazil, Turkey, Australia and India (Isci and Demirer, 2007). Among other cotton wastes, cotton gin trash has high lignocellulosic material (60%). Lignocellulose is a polysaccharide combination of cellulose, hemicelluloses, and lignin that has recalcitrant plant material (Placidoet al., 2013).

Cotton gin trash is a waste produced during the ginning process of cotton. The disposal of cotton gin trash is an issue as it is in large amounts and pose threat to the environment (Placido et al., 2013). Cotton gin trash has high ligno-cellulose that makes it a perfect candidate for a cheap and readily available carbon source (Sahu and Pramanik, 2015). Many studies have confirmed the hydrocarbon potential of cotton gin trash (Jeoh and Agblevor, 2001; Agblevor et al., 2006; Isci and Demirer, 2007).

The figure below show the area under the cotton cultivation along with the cotton yield from the year 2006 to 2017.

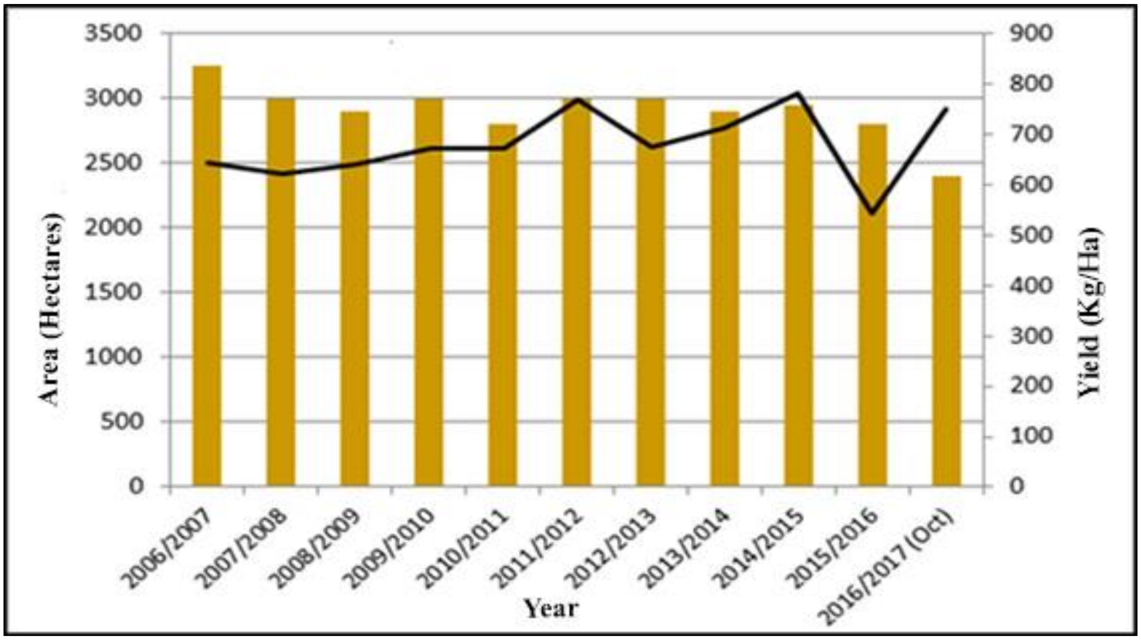

Figure 1: Pakistan cotton growing area and yield

1.4 Biogas in Pakistan

Pakistan is under a critical energy shortfall which is getting worse with each day passing. This burden on forested land and national energy grid can be reduced through the installation of biogas plants in rural areas of the country which can serve 70 percent of Pakistan's population living there. Government is providing grants and funds to build biogas plants and encourage the use of biogas as a major fuel. The production of main substrate, cow dung to run the biogas plant can be made available by the cattle farms, it is estimated that a family of 4 to 5 members can easily fulfill their domestic needs through 50kg of cow dung that produces 100 cubic feet of biogas daily. (Akhter, 2004) and per the estimates each farmer in Pakistan possesses 2-3 buffalos/cows (Sheikh, 2009). Akhter in his study found out that substrate from these cattle when mixed in an equal proportion of water will generate large amount of biogas. This biogas can then be utilized for domestic purposes such as cooking and heating and to produce electricity using a gas fired engine additionally the residual waste from the plant can be used in farms as fertilizer.

During the period of 1974 to 1987, the government of Pakistan had installed about 4,137 biogas plants. The first 100 biogas plants were installed by the government financial support which was withdrawn later. Soon after the withdrawal of government's fund that the installation of biogas plants slowed down drastically and only 6000 plants were set up by the completion of year 2006. However, 4000 biogas plants have been installed by 50 percent financial support from the beneficiaries of Pakistan Centre for Renewable Energy Technologies (PCRET, 2010)

Currently Pakistan is making great effort in the biogas technology. The biogas plants are being made available in local markets and the construction of the plants is taking place nationwide. Plants with movable gasholders, built-in immobile dome gasholder and cheap balloon/bags are three commonly used designs for the biogas plants operating in Pakistan (Amjid et al., 2011), figure 2 and 3 below show the units that are currently being operated. The use of biogas as fuel is common in the educational institution of Pakistan. Utilization as vehicle fuel has proven to be the most successful way to upgrade biogas waste. However, the high investment cost of the plants has made the development of this technology impossible without the support of the government to make it attractive and available in the market. (Technology times 2011)

Figure 2: Movable gasholder biogas plant installed by PCRETE

Figure 3: Fixed dome gasholder biogas plant installed by PCRETE

1.5 Hypothesis

The production of biogas is expected to increases with the addition of co-substrates. The addition of cotton gin and leachate will provide a synergism for the production of biogas from cow dung.

1.6 Objectives

1. To convert the MSW and cotton gin waste into energy.
2. To characterize the nature of cotton gin and leachate as the energy sources.
3. To identify the product (biogas), the ionization of methane into different species through mass spectrometry gas chromatography technique.

Chapter 2

LITERATURE REVIEW

This chapter comprises of few basic details and different factors affecting biogas. The core purpose of this chapter is to deliver important material which is helpful for the understanding of the results and discussion in the next chapter.

2.1 History of Biogas

In Assyria and Persia water bath was heated using biogas during the 10th century BC in Assyria and during the 16th century AD in Persia, said Lusk (2007).

The decay of organic matter could result in the evolution of flammable gasses as concluded by Helmont in the 17^{th} century. A study conducted by Volta in the year 1776 found out that there the volume of flammable gasses produced was directly proportional to the quantity of the organic matter undergoing the decay process. Sir Humphrey Davy in 1808 discovered the presence of methane in gases produced through the anaerobic digestion of cow dung. (Lusk, 2005)

2.2 Composition of Biogas

Biogas is a combination of gasses that are combustible that are formed as a result of anaerobic decomposition of organic matter due to the presence of bacteria in oxygen deprived conditions. The gas that is majorly produced is methane (CH4) along with carbon dioxide (CO2) in a relatively smaller amount. Other gases are also present in traces are listed in the table below (Zhu, 2008).

Table 1: General composition of biogas

Gases	% Composition
Methane (CH_4)	40-75%
Carbon dioxide (CO_2)	25-55%
Hydrogen sulfide (H_2S)	50-5000ppm
Ammonia (NH_3)	0-1%
Water (H_2O)	0-10%
Nitrogen (N_2)	0-5%
Oxygen (O_2)	0-2%
Hydrogen (H_2)	0-1%

Source: Zhu 2008

Composition of biogas depends upon the material feed and process nature (Rao, 2010).

Four constituents are required to produce biogas:

1) Organic Material
2) Microorganisms
3) Anaerobic Environments
4) Temperature

2.3 General Types of Substrates used

Typical substrates for the biogas process. Biogas can be produced from the co-digestion of the following raw materials (Murmansk, 2008):

- Food waste
- Sewage sludge

- Cotton waste from industry
- Animal manure.
- Agriculture waste
- Grass and herbs

2.3.1 Co-substrates: cotton gin and leachate

The key techniques for the optimization of biogas include design and treatment of the digester along with the Co-substrates that reduce the inhibitory action of main substrates. Cotton gin mixed with cow manure balanced the amount of macro and micronutrients, increasing the organic loading yields higher volume of methane per unit of digester volume in a relatively shorter retention time. In a similar study Corral (2008) in his experiment took advantage of cellulolytic microbes living in bovine dung to breakdown the carbon in the cotton gin. Finally, by synergizing and diversifying the microorganism the process of methanogenesis is enhanced. Biogas production is enchased by the appropriate and accessible feed. The source of the biomass greatly effects the biodegradability of the biomass. The current analysis also discoursed several kinds of pretreatments to eliminate the difficulties while filling the digesters. Different types of substrates are used for the anaerobic breakdown of biogas along with the design of digester having numerous process physical conditions also the nature of feed added for the optimization of biogas.

The landfill management can be well controlled by an emerging technology of leachate recirculation. The influence of the recirculation of leachate on the co-disposal of three major types of wastes (sediment dredging, municipal solid waste and sewage sludge) was studies using a lab column examination. Chemical factors (COD, total-P, pH, ammoniacal-N) and gas production (production rates, concentrations of CH_4 and CO_2 and total gas volume) were observed for eleven weeks. The recirculation resulted in a decrease in the duration of waste-stabilization and was helpful in improving gas production and enhancing leachate quality in terms of COD. The results showed that leachate recirculation has the potential to increase the efficiency and waste volume reduction rate of landfill sites (Sanphoti et al., 2006)

2.4 Types of anaerobic digestion processes

There are three major types in anaerobic digestion, which are given below (Torales, 2013);

2.4.1 Psychrophilic anaerobic digestion

This type of anaerobic digestion is mainly carried out in that region where temperature is low. Temperature ranges between 10-20 °C. Under this temperature range, digestion is very slow and time consuming. About 40-50 days are required for biogas generation.

2.4.2 Mesophilic anaerobic digestion

Temperature ranges for mesophilic digestion are between 25-40 °C. This type of digestion takes 25-30 days for bio degradation. Mesophilic digestion tends to be more sturdy and tolerant than thermophilic anaerobic digestion but biogas generation is less (Torales, 2013).

2.4.3 Thermophilic anaerobic digestion

Thermophilic digestion occurs between 50-65 °C. Digestion process is very fast in the thermophilic process. Biogas generation starts on 8th to 10th day of the run. High biogas production is achieved in this process but it is very costly because high degree of energy input is required. During this process, about 30-60% of the digestible solids are converted into biogas (buhr and Andrews, 1977).

2.5 Process of Biogas Production

The reactions of biogas production process through anaerobic digestion start with the hydrolysis reaction followed by the formation of organic acids, carbon dioxide and hydrogen and finally ends up in the formation of methane gas. The details of each step are given below.

2.5.1 Chemistry of anaerobic digestion process

1. Carbon, C → organic acids (R •COOH) → CH4 + CO2
2. Nitrogen, N→ amino acids [R • (NH2) • COOH] →NH3+ amine.
3. Sulphur, S→ H2S organic+ S compounds
4. Phosphorus, P→ PH3 +organic P compounds (Vindis et al., 2009).

There is a streamlined flow metahne production from biogas. The process is depicted in the flow diagram below.

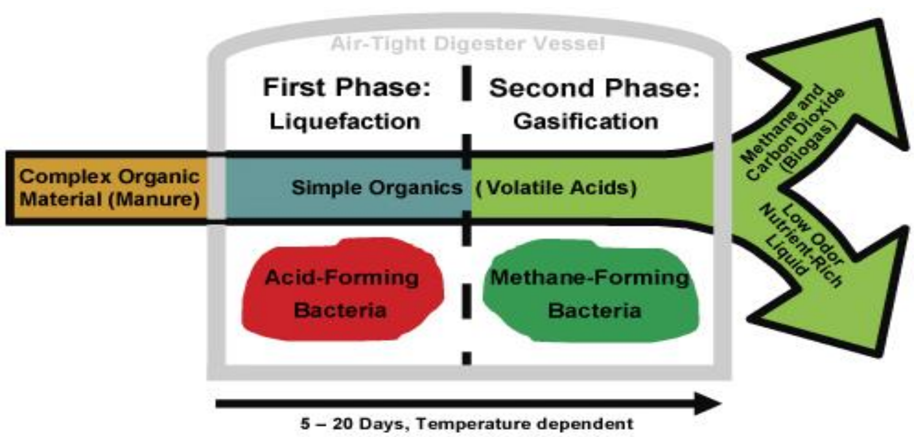

Figure 4: Anaerobic digestion process in a digester

2.5.2 Steps in anaerobic digestion process

Anaerobic digestion a chain processes. It is a well-known process because of effective stabilization and efficient performance that results in profitable production of biogas. Anaerobic digestion takes place in four steps, which are listed below.

1. ***Hydrolysis:*** In first step the organic materials are depolymerized, in which the complex polymers e.g.; proteins, carbohydrates and lipids are break down into monomers e.g.; cellulose, protease, lipase and amylase, by extracellular enzymes. Polysaccharides are converted into sugar. Lipids are converted into small or long chain fatty acids. (Gunaseelam, 1997)
2. ***Acidogenesis:*** In this process, microorganisms use fatty acids, amino acid and sugars as substrates to produce organic acids like acetic, propionic, butyric and small chain fatty acids, alcohols, H2 and CO^2. (Vindis *et al.*, 2009).
3. ***Acetogenesis:*** There is no clear distinction between Acetogenesis and Acidogenesis reaction. In this step, the Acetogenesis bacteria degrade the hydrogen sinks acids like propionic, butyric and valeric acids into formate, acetate, CO2 and hydrogen. (Rojas *et al.,* 2010).
4. ***Methanogenesis:*** H_2/CO_2 and acetic acid are mainly utilized by methanogens to form methane and carbon dioxide. Methanogens utilize limited amount of substrate like methanol, alcohols and formate to produce methane

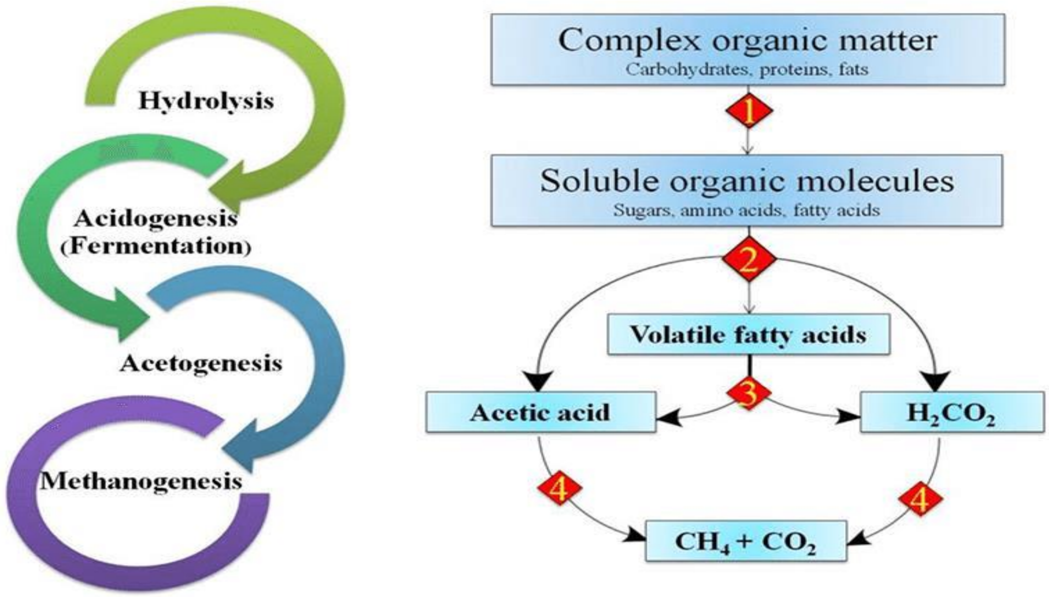

Figure 5: Steps in anaerobic digestion process

2.6 Parameters effecting the Biogas Production

Many factors affect the anaerobic digestion process, controlling these factors can enhance the anaerobic process (sambo et al., 1995). These factors include:

- Type of organic waste
- Waste particle size
- Temperature
- pH
- Presence of toxic material
- Hydraulic retention time
- Solid retention time
- Carbon to nitrogen ratio
- Digester loading rate
- Mixing

Not all types of waste can be converted to biogas. Anaerobic bacteria cannot degrade lignin, Waste that contain huge amount of sulfur or nitrogen result in large amount of unwanted ammonia and hydrogen sulfide that is not our requirement. The waste that is not completely soluble in water can degrade very slowly (Marchaim, 2009). A brief description of the above listed parameters is given below

2.6.1 Waste particle size

Size reduction is very important parameter in anaerobic digestion. Pumping of waste after size reduction is much easier to degrade as compared to raw waste. We can avoid blockage of pipes after shredding. Shredding not only helps in pumping but also increase the surface area for bacteria in reaction. Through shredding biodegrading can easily accomplish. It also helps in getting the consistent feed (okishio et al., 2010).

2.6.2 Temperature

Temperature plays very important role in anaerobic digestion. Biogas production depends on temperature variation. With increase in temperature, biogas production increase.

2.6.3 pH

pH plays a vital role in anaerobic digestion. Methane producing bacteria are directly affected with the hydrogen ion concentration. Digestion is completely prohibited by excess of acidity. Bacteria produce methane in the pH range of 6-8 (Yadvika et al., 2004). At the start acid forming bacteria begin to produce acids which break down into methane by methanogens. If acids formation is still increasing and exceed the consumption level of methanogens, then these acids decrease the pH. When pH decreases, the carbon dioxide contents start to increase while methane production decreases. The narrow range mentioned above is mainly due to the varying optimal pH levels for the varying microbes involved in the system (Kim et al., 2003). The optimal level for methanogenesis is 7, while the optimum level for acidogenesis and hydrolysis is in the range of 5.5-6.5. (Cantu, 2014).

2.6.4 Hydraulic retention time

Hydraulic retention time means how many days the material remains in the digester. This retention time plays an important role in anaerobic digestion because it tells us the time factor for bacteria growth which on other hand tells the conversion of organic material into methane (Jilani et al., 2007). For thermophilic environment the required hydraulic retention time is about 15 to 25 day while that required mesopilic conditions ranges between 30 – 50 days (Demetrides, 2008)

2.6.5 Solid retention time

The solid retention time is one of the important factors in digestion process. It is equal to the amount of solids maintained in the digester divided by the amount of solid drained. Conversion of volatile solids into methane depends on solid retention time. If solid retention time is very low, then it shows that there is no sufficient time for bacteria to grow. Bacteria loss in effluent increases. If bacteria loss rate exceeded than bacteria growth rate, wash out occurs (Okishio et al., 2010).

2.6.6 C: N ratio

Carbon to nitrogen ratio is also a main factor that affects the anaerobic digestion process. Carbon provides energy to microbes while nitrogen enhances the microbial growth. If we do not maintain the quantity of nitrogen, then microbial growth is inhibited which have adverse effect on methane production. If carbon contents increase and nitrogen contents decreases this will result in increase in energy level which inhibits microbial growth. Carbon is utilized 30 times more than the nitrogen. Therefore, carbon to nitrogen ratio should be maintained at 30:1 **(**Emerson et al., 2003).

2.6.7 Digester loading rate

Volatile components are those, which can be digested. It depends on the type of feed or waste, which is fed into the digester because it will dictate the biochemical activity in the digester. Loading rate keeps the balance between the acid formation and methanogensis. If the acid formation increases, then the pH level decreases. This has adverse effect on methanogensis and also effect the carbon to nitrogen ratio. The digesters used in this experiment had a working volume capacity of 1.2 liters and a total of eight (8) digesters were used, four (4) for vegetable waste and the other four (4) for cow dung. Each digester was filled with 600 grams of waste either vegetable waste or cow dung as solid portion and 600 ml of liquid portion, the liquid portion consisted of different proportion of leachate with water. This feeding of the digesters was done at the start of the experiment and no further addition was made after that (Sangeen, 2016).

2.6.8 Mixing

Mixing plays an important role in achieving the optimum anaerobic process. Mixing keeps uniformity in substrate concentration and in temperature. Mixing avoids solid deposition in anaerobic reactor (Jilani et al., 2007).

2.7 Design Parameters of a Generalized Biogas Plant

The general parameters used while setting up a biogas plant are given in the table 2.

Table 2: Design parameters of biogas plant

Parameter	Range
Temperature	20-45 °C (mesophilic condition)
pH	6.5-9.5
Total Solid	8-10%
C / N ratio	25-30:1
Volatile Solids	≥ 80%
Moisture Content	≥70%
Particle Size	10-12mm

Source: Jilani et al, 2007

2.8 Leachate Management for Biogas Plant

The landfill management can be well controlled by an emerging technology of leachate recirculation. The influence of the recirculation of leachate on the co-disposal of three major types of wastes (sediment dredging, municipal solid waste and sewage sludge) was studies using a lab column examination. Chemical factors (COD, total-P, pH, ammoniacal-N) and gas production (production rates, concentrations of CH_4 and CO_2 and total gas volume) were observed for eleven weeks. The recirculation resulted in a decrease in the duration of waste-stabilization and was helpful in improving gas production and enhancing leachate quality in terms of COD. The results showed that leachate recirculation has the potential to increase the efficiency and waste volume reduction rate of landfill sites. (Sanphoti et al., 2006)

Gillani (2012) carried out a research on biogas production from cow dung, vegetable residues and co-digestion of these wastes in anaerobic conditions under thermophilic temperature in batch and continuous mode of digestion. He reported that in batch mode, biogas production from cow dung, vegetable waste and co-digestion was 47.7 L, 65 L and 56L per 1 kg of waste respectively. Average methane content in the biogas produced was 50%, 62% and 55% respectively. In continuous mode of digestion cow dung produced 44 L/kg of waste, vegetable waste produced 71.9 L/kg and co-digestion of both wastes generated 55 L/kg. The average methane content was around 50%, 55% and 52% respectively.

Azhar (2012) conducted experiment on biogas production using food waste under thermophilic conditions in batch mode of digestion and reported that food waste produced 0.04 cubic meter of biogas per kg of waste having 60% methane content in it. Afeeq et al. (2012) also studied biogas production from food waste under thermophilic condition in batch mode of digestion. Their result showed that the cumulative gas produced under these conditions was about 38.5 m3 with gas production rate of 61.61 liter per kilogram of waste.

Chapter 3

METHODOLOGY

In this chapter, the methodology of the whole experimental work like setup of the digester for biogas production, the operational parameters under which the digestion was carried out and different other analysis of the feedstock material done before and after the digestion process will be discussed.

The adopted methodology for the entire experimental work done is as follows,

1. Collection of waste
2. Setup and feeding of the Digesters
3. Estimation of moisture content, dry matter and volatile solid (before digestion)
4. Maintenance of Temperature and pH (during experiment)
5. Biogas collection system (during experiment)
6. Estimation of moisture content, dry matter and volatile solid (after digestion)

3.1 Field Survey and Waste Collection

The typical waste used in the experiment were cow dung, cotton gin and leachate which were obtained from their sources manually using gloves and brought to the laboratory. Cow dung was collected from an Afgan Abadi located near NUST H-12 Islamabad. Leachate from a landfill site, and cotton gin was brought from cotton ginning industry of same city. All three of the substrates are shown in figure 6 below a b c

Figure 6 The ingredients of the feed: (a) Leachate, (b) Cotton Gin and (c) Cow Dung

3.2 Layout of Biogas Reactor

The lab–scale reactor was designed as a single batch digester, made up of 8 up-flown anaerobic digesters of 1.2 L capacity each, had been used in this study. Substrates were feed into the digesters to 3/4 of their total length whereas the rest 1/4 was left for the collection of gas. The complete-mix condition was maintained through mixing and shaking of the digester while adding the mixture. Once the digesters were adjusted with a pipe attached at the top to allow the up flow of gas from the digester into the inverted gas collection bottle through water displacement method. The digesters were placed and affixed at a certain depth in a water tank made up of glass having dimensions of 24 cm by 16 cm to make sure that the digesters had enough contact with water, they were affixed with tapes and wires. Water bath was prepared by filling the tank to $3/4^{th}$ of its height. Mesophilic temperature of water bath was maintained at 34˚C by placing an aquarium heater. To make sure that temperature was stable, the thermostat fixed within the heater was adjusted at 34˚C. The test run was done. The gas production was monitored daily and observations were made. The gas production from leachate was noticed on the next day whereas that of cotton gin was observed at the fourth day. Production rates for the rest of six digesters having the substrates in different ratios were observed on variable days. Followed by the first run, the digesters were then washed and dried for the consecutive run

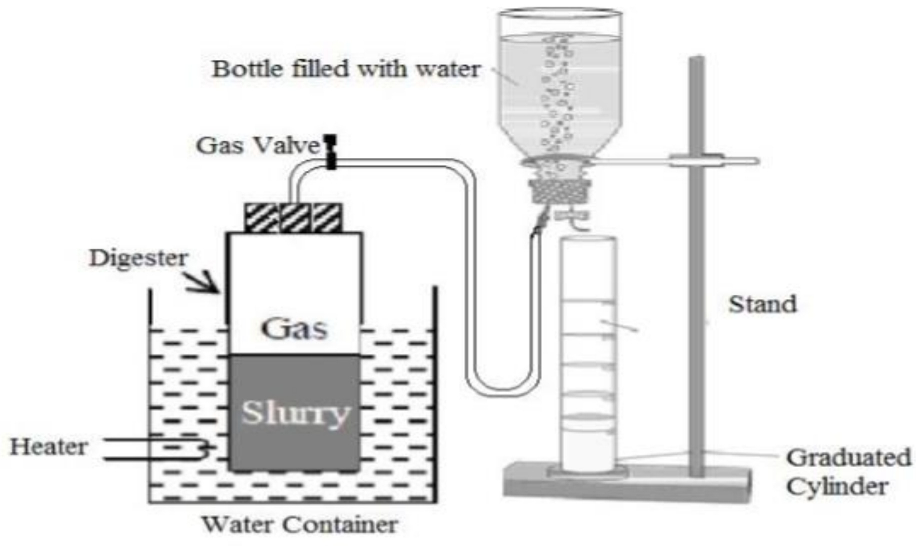

Figure 7 Schematic diagram of experimental setup

General specifications of a biogas reactor are given in the table below.

Table 3: Specifications of the biogas reactor

Specification of Biogas Plant	
Plant type	Reactor with eight fixed digesters
Batch type	Batch feeding
Type of waste	Cotton Gin, Leachate and cow dung
Temperature	mesophilic conditions (20 ^{o}C-45^{O}C)
pH	6.5-9.5
Length of inlet	5.08 cm
Volume of digester	1.2 L

3.3 Preparation of Digester Feed

Triplicate for both controls were prepared. Control for cotton gin was prepared by pre-weighing 100 grams of cow dung with the addition of 600 ml water. Second control contained 600 ml of leachate to analyze the biogas production from raw leachate without dilution and addition of the main substrate (cow dung).

Samples for the pilot run were prepared in a fixed solid to liquid ratio of 1:6, however the solid weight was prepared in different ratios as mentioned in the table below.

Table 4: Substrates prepared for the test run

Experiment	Ratio (Solid to Liquid)	Cotton Gin (g)	Cow Dung (g)	Total Solid Mass (g)	Liquid Volume (ml)	
					Leachate	Distilled Water
W1	1:2.5	38.6	71.4	100	0	600
W2	1:5	16.7	83.3	100	0	600
W3	1:7	12.5	87.5	100	0	600
L1	1:2.5	38.6	71.4	100	600	0
L2	1:5	16.7	83.3	100	600	0
L3	1:7	12.5	87.5	100	600	0

Where:

1. W1 Combination (1:2.5) in Water
2. W2 Combination (1:5) in Water
3. W3 Combination (1:7) in Water
4. L1 Combination (1:2.5) in Leachate
5. L2 Combination (1:5) in Leachate
6. L3 Combination (1:7) in Leachate

After the completion of the test run it was observed that the amount of substrate was not sufficient since the digesters were not filled up to 75% of their length thus to fulfill the requirement of the optimum design the amount of substrate in each digester was increased as shown in the table 5 below.

Table 5: Substrates prepared for the main run

Experiment	Ratio	Total solid Mass, 150 (g)		Liquid volume (ml)	
		Cotton Gin	Cow Dung	Leachate	Distilled water
W1	1:2.5	42.9	107.1	0	900
W2	1:5	25	125	0	900
W3	1:7	18.8	131.2	0	900
L1	1:2.5	42.9	107.1	900	0
L2	1:5	25	125	900	0
L3	1:7	18.8	131.2	900	0

3.4 Total Feed Added in the Reactor

All the prepared samples were added in the digester on the same day. The amount of substrate in each digester is shown in the table below.

Table 6: Total feed added in the digesters

Run no.	Total amount of cow dung (g)	Total amount of cotton gin (g)	Leachate(ml)
Test run	484.4	135.6	1800
Main run	726.6	173.4	2700

3.5 Temperature Control

For temperature control a 1000 W electric immersion rod with thermostat was used to maintain the temperature to 34˚C.

3.6 pH control

For pH measurement, pH meter (Wagtech international) was used. The pH of all the eight digesters was set to greater than 6.5 before starting the experiment. For pH adjustments Sodium Carbonate (Na_2CO_3) was used.

3.7 Measurement of Biogas Volume

To measure the volume of a biogas liquid displacement method was used. In liquid displacement method, gas measuring equipment is constructed from glass or plastic jars, bottles or cylinders. In this method volume of gas is measured by the amount of liquid displaced from the tightly sealed liquid bottle into the open container by the gas released from the digester (Parajuli, 2011).

a **b**

Figure 8: (a) and (b): Gas Collection through water displacement method

3.8 C: N Ratio

The estimated values for carbon to nitrogen ratio in substrate used during the experiment were derived from extensive literature review. No experiment was conducted. The values of carbon and nitrogen that were obtained by ultimate analysis in literature were used in calculations for estimating C/N ratio in experiment. C: N ratio was calculated by multiplying the % carbon for each component by the sum of parts by mass of that ingredient and adding it in the carbon total for components. Then divided the amount of N into the amount of C, which gave the C: N ratio where N = 1. The analysis was done by following standard methods in ICARDA manual (1996). The C/N ratio for substrates are given below.

Table 7: The C: N ratio of the substrates

Sr No.	Substrate	C: N ratio
1	Cow dung	14-25:1
2	Leachate	14:1
3	Cotton gin	21:1

3.9 Moisture Content in Solids

After preparing the substrate for digestion a sample has been taken for the analysis of moisture content. A silica crucible was used for this purpose. The crucible was washed and dried in hot air at 105oC and ignited in furnace at 550°C for 1 hour, so that all the organic matter present in it is exhausted. The empty crucible was then weighed. A representative sample was taken in a crucible and weighed again. After weighing the sample was placed in an oven Model WTC Blender for drying. This is done to get the exact solid mass. The sample was in an oven placed for 24 hours. Once the sample was dried and cooled in the desiccators it was weighed again. To measure the moisture, content a simple calculation is done (APHA, 2005).

$$\text{Moisture Content} = \frac{W_w - W_d}{W_w} \times 100$$

where:

Mn = percentage of moisture content of material

W_W = wet weight of the material

Wd = weight of the material after drying.

3.10 Dry Matter Content

Along with moisture content the dry matter was obtained in the same experiment. When the sample was placed in an oven for 24 hours, it evaporates all the water content present in it and the crucible is only left with the solid dry matter. After weighing the crucible with dry content in it, simple calculation is done to get the dry matter content present in the representative sample (Demetriades, 2008).

$$\%\ \text{Dry matter} = \frac{(A3 - A1)}{(A2 - A1)} \times 100$$

A1 = Empty crucible weight, g

A2 = Empty crucible weight + wet sample, g

A3 = Empty crucible weight + dried sample, g

3.11 Volatile Solids

After getting the dry solid content, the dried samples were placed in the muffle furnace Model NEY M 525 Series III at 550oC for 5 hours. In this way, all the volatile organic matter is removed from the sample and only the ash content or fixed solids are left. Again, the crucible with ash content was placed in the desiccators to cool it down weighed for the final time. The readings were putted in the formula below to get the organic content present in the sample (APHA, 2005).

$$\% \text{ Volatile solids} = \frac{(A2 - A3) - A1}{A2 - A1} \times 100$$

A1 = Empty crucible weight, g

A2 = Empty crucible weight + dried sample, g

A3 = Empty crucible weight + ash content, g

3.12 COD Measurement

The method used for the determination of COD in the samples was closed reflux titrimetric. The sample was first centrifuged and then diluted up to 10 % in 100 ml distilled water before COD analysis. 10 ml of it was oxidized by 3.5ml of sulfuric acid in COD vial. The sample was then refluxed in a strong acidic solution with a known excess of potassium dichromate ($K_2Cr_2O_7$). After digestion, the remaining unreduced $K_2Cr_2O_7$ was titrated with ferrous ammonium sulfate to measure the amount of $K_2Cr_2O_7$ used up and the oxidizable material was calculated in terms of oxygen equivalent. The standard reflux duration was 2 hours. Duplicates were prepared for all samples before the analysis so that their average value could be recorded (APHA, 2005).

3.13 Flame of Biogas

The gas ignition was carried out to observe the biogas flame. In case of complete combustion, a dark blue flame is produced which is almost invisible in daylight.

3.14 Quantitative Analysis of Biogas using GC-MS Technique

Ghani and Idris (2009) analyzed biogas contents using gas chromatography mass spectrometry, therefore the composition was determined for biogas samples by gas chromatography. The gases to be measured by this method were CO_2, CH_4, NH_4 and H_2. They were determined using PE Nelson 1020 Personal Integrator. (Perkin Elmer, Norwalk, Connecticut, USA). The instrumental details and the analysis conditions are given in table below.

Table 8: Information about GC-MS used for detailed gas composition analysis

Parameter	Value
Model of GC-MS	GC2010plus-SHIMADZU
Data handling	PE Nelson 1020 Personal Integrator
Column temperature	160°C
Carrier gas	Helium
Sample size	0.25 ml (gas)
Total analysis time	8 Min
Gas flow rate	30ml/min
Detector type	Thermal conductivity

Gas samples were analyzed using the GC-MS. The samples were injected through syringe and the results were displayed on the monitor in the form of peaks as depicted in the Figure (a, b, c and d) below.

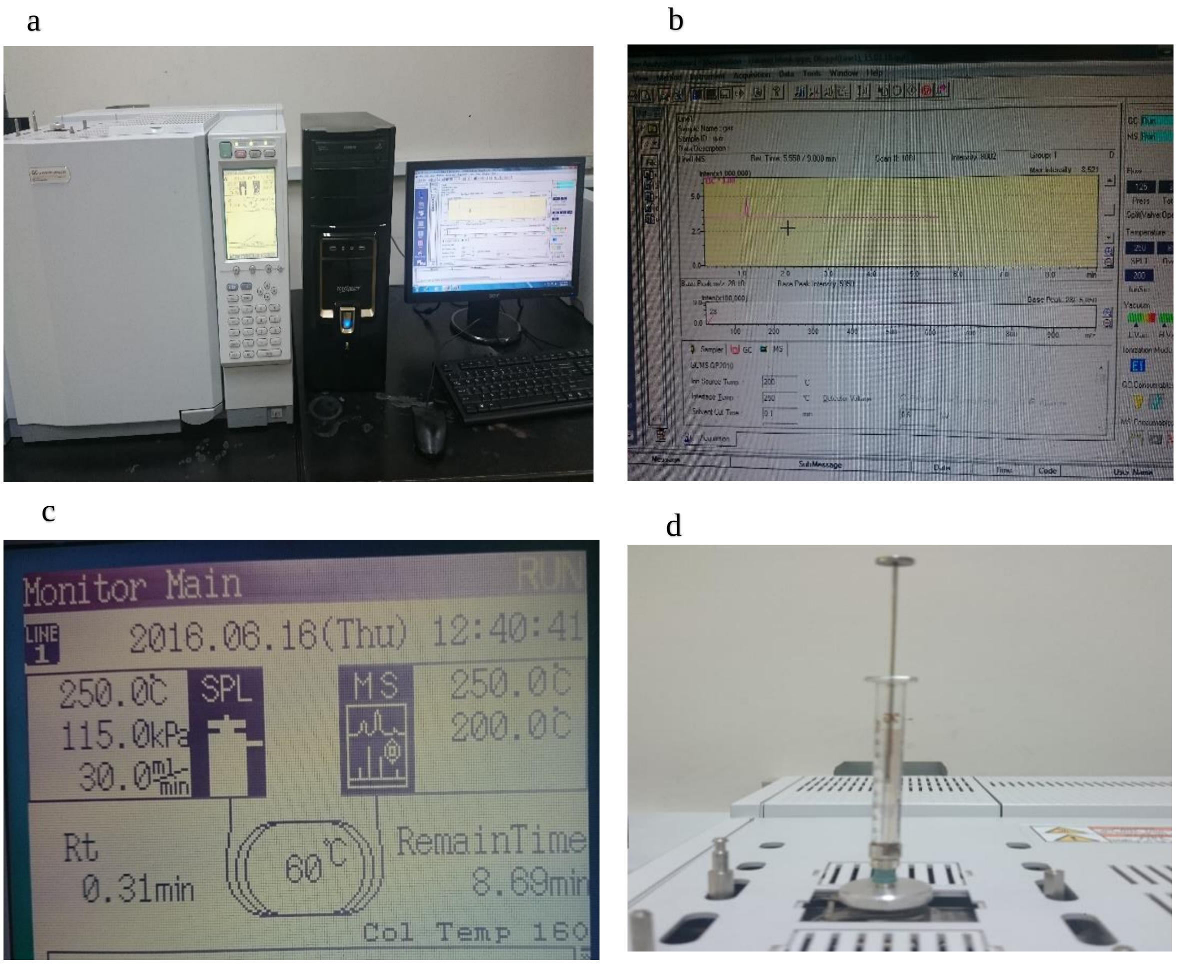

Figure 9: (a), (b), (c) and (d): Gas analysis through GC-MS

Chapter 4

RESULTS AND DISCUSSION

In this chapter the results obtained by the methodology adopted in the previous chapter to produce biogas with the addition of co-substrates; cotton gin and leachate and their correlation with respect to production rate will be discussed.

4.1 Analysis of Substrate

The results below (in table 4.1) show the characteristics of digestion substrate. These analyses signify the highest total solid content present in cotton gin. volatile solids content and COD was highest in cow dung i.e., 82% and 48890 mg/l respectively, this was due to the presence of microorganism within the inoculum. The highest amount of moisture content was found in Leachate had the percentage of Volatile solids was highest in cow dung followed by leachate and then cotton gin at 82% 80% and 78% respectively, following a same trend as that of in the study carried out by Corral et al (2008) where 81.5%, 81% and 81% of solid waste was present in cow dung, leachate and cotton gin respectively.

Table 9: Initial analysis of the substrates

Parameters	**Cow dung**	**Cotton Gin**	**Leachate**
Moisture content (%)	74.8	6.5	98.6
Total Solids (%)	25.2	93.5	1.4
Volatile Solids (%)	82	78	94
COD mg/l	48890	10480	12630

The study of digesters showed a that slurry was thick and blackish. Initially it was smelly but with the effect of degradation the smell was reduced.

4.1.1 Moisture content

The substrate analysis results indicated that the moisture content present in cotton gin was 7.6% and 5.33% and that of cow dung was measured to be as high as 75.1 and 74.37% for pilot run the consecutive run respectively. In a similar study conducted by Abubakar and Ismail (2012), the moisture content in cow dung was 41.2%.

4.1.2 Ash content

All three of the substrates were weighed before as well as after the oven drying to measure the concentration of ash content. After getting the dry solid content, the dried samples were placed in the muffle furnace Model NEY M 525 Series III at 550°C for 5 hours, in this way all the volatile organic matter was removed from the sample and only the ash content was left. The ash content in cow dung was found to be 6% whereas that in cotton gin was as low as 3%. Almost a similar finding was achieved by Khan (2010) in which the ash present in the waste was 15%.

4.1.3 pH

After the preparation of the substrate for anaerobic digestion, pH of the substrates was adjusted to the required optimum level. According to Williams (1998), the preferred range of pH for the efficient functioning of the methanogenic micro-organisms is around 6.8 – 7.5. Metcalf and Eddy also reported that the most appropriate range of pH for the growth of bacteria is around 6.5 – 7.5.

Initially the pH of the substrates was low. The pH of digester ranged from 3.9- to 4.8 thus the pH needed to be adjusted so the required amount (2 grams) of Na_2CO_3 was used to raise the pH to the required level adequate for digestion as shown in Table 10 below.

Table 10: Average initial and final pH

Sample	Initial pH	Final Ph
W1	4.09	7.05
W2	4.87	7.22
W3	4.29	7.18
L1	4.08	7.13
L2	3.94	7.11
L3	4.28	7.18
Average	4.26	7.15

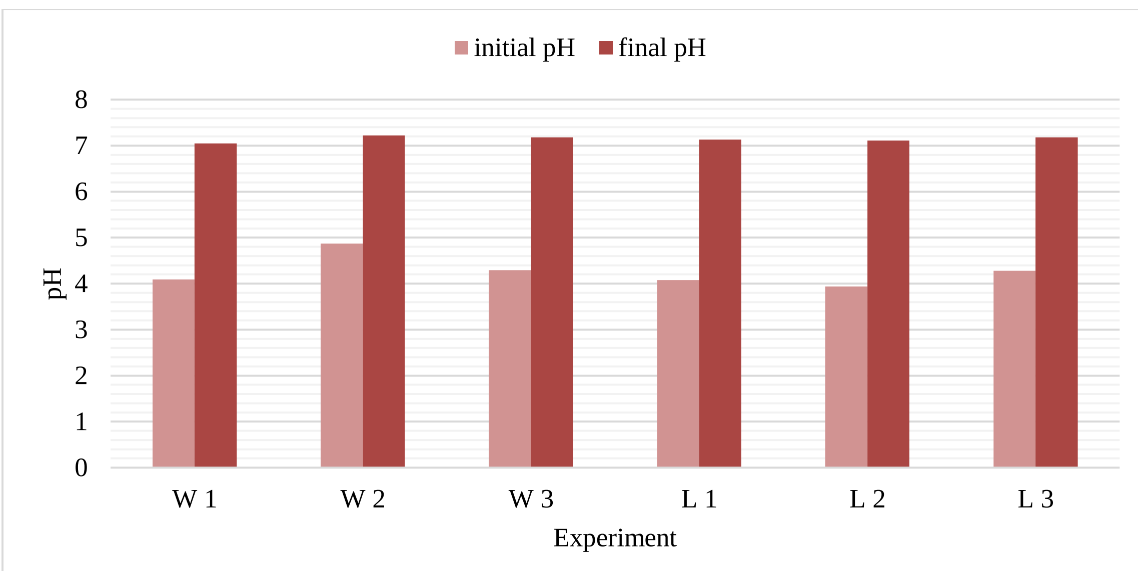

Figure 10: Average initial and final pH of the samples

4.1.4 Total solids

Solid analysis was important in the control of biological treatment process. The initial and final TS for all six of the digesters is given in the table below. Per NRCS Report (2005) the TS of feed material can be upto 14%. Igoni et al. (2008) conducted a research on anaerobic breakdown of MSW for biogas production and found that the TS reduced from 10% to 4%. Zhu et al. (2008) conducted a similar study and established that TS reductions ranged from 50.2% to 65.0%. the percentage reduction in volume for the digesters is given in the graph below.

Table 11: Percentage reduction in total solids

Sample	Initial TS (%)	Final TS (%)	% Reduction in TS
W1	10.27	4.29	58.2
W2	10.02	5.07	49.4
W3	10.53	3.66	65.2
L1	10.86	3..23	70.25
L2	10.05	4.86	54.62
L3	10.40	4.30	58.65

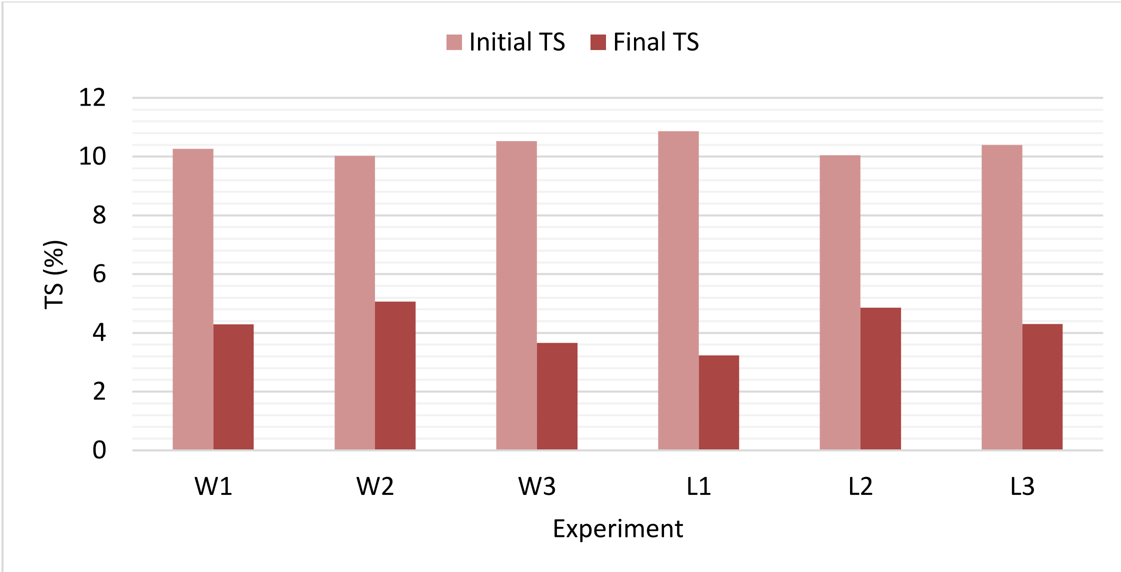

Figure 11: Total solids in variou digesters

4.1.5 Volatile solids

The methane producing potential depends on the amount and nature of the accessible organic material, which is sometimes referred to as the volatile solids (Omar et al., 2008). The breakdown of organic solids depends upon the temperature (Metcalf and Eddy 2003). In the present study the VS present in each sample was analyzed and the percentage of volume reduction for each sample was calculated.

The higher the amount of volatile solid the higher is the percentage volume reduction. The greater amount of volatile solid resulted from the breakdown of organic matter present in the substrates. The percentage volume reduction for all six of the samples in shown in the graph below. For the samples having synergy of cotton gin and cow dung in water have given away lesser volume reduction results ranging from 36.85% to 43.24% whereas those in leachate have shown a higher reduction rate ranging from 75.3% to 80.3% and the overall reduction by 58.6%. These results owe to the fact that the volume reduction in the sample containing cotton gin and cow dung was 58% whereas in the one containing leachate and cow dung was 98% and the overall volume reduction was measured to be 57.6% in a similar study conducted by Collar et al (2008).

Table 12: Percentage reduction in volatile solids

Sample	Initial VS (mg)	Final VS (mg)	% Reduction in VS
W1	1070.91	607.76	43.24
W2	972.31	580.45	40.3
W3	891.35	562.81	36.85
L1	1211.58	238.46	80.3
L2	1105.71	252.08	77.2
L3	1100.40	270.99	75.3
Average Volatile Solids	1058.71	418.74	58.6

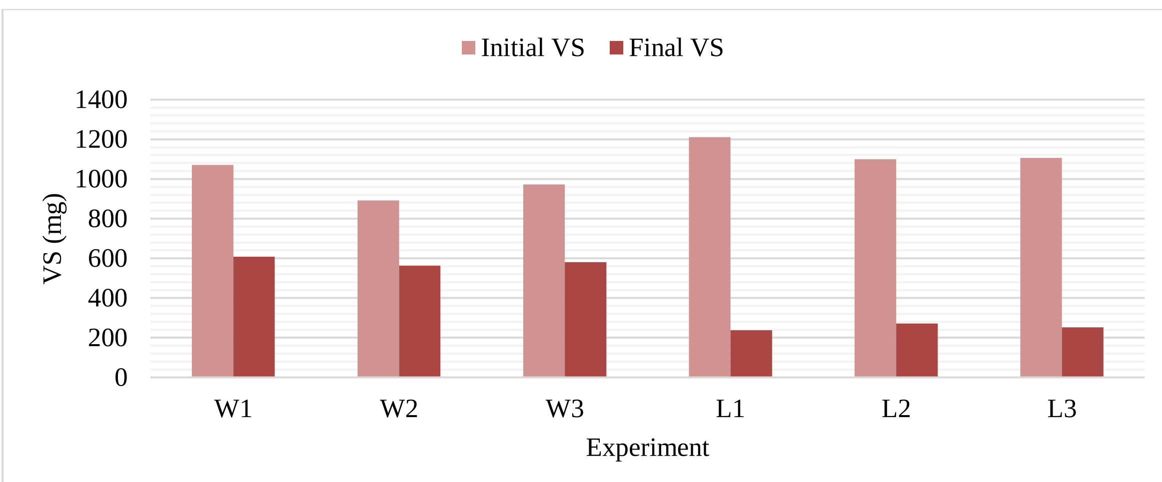

Figure 12: Volatile solids in various digesters

4.1.6 COD

Chemical oxygen demand is the amount of the oxygen used up when during the chemical breakdown of organic material (Metcalf and Eddy 2003). The average percentage removal of COD for the first batch was 58.5% and for the second batch was 59.1%. Sakar et al. (2009) carried out anaerobic digestion of livestock waste treatment, where the chemical oxygen demand (COD) removals ranged from 57 and 78%. Dawood et al. (2011) found that the % COD removal was 50.0% at the retention time of 72 days whereas Abubakar and Ismail (2012) recorded 48.5% COD removal. The % COD reduction is directly proportional to the increasing organic loading rates but not monotonically for different durations. The comparison of final and initial COD for all the samples is given in the table below.

Table 13: Average percentage removal of COD

Sample	Initial COD (mg/l)	Final COD (mg/l)	Removal (%)
W1	15708	7376	53.0
W2	13040	7060	45.9
W3	10790	5885	45.4
L1	20935	5880	71.9
L2	21980	6890	68.6
L3	19720	7900	59.9
Average percentage removal			57.45

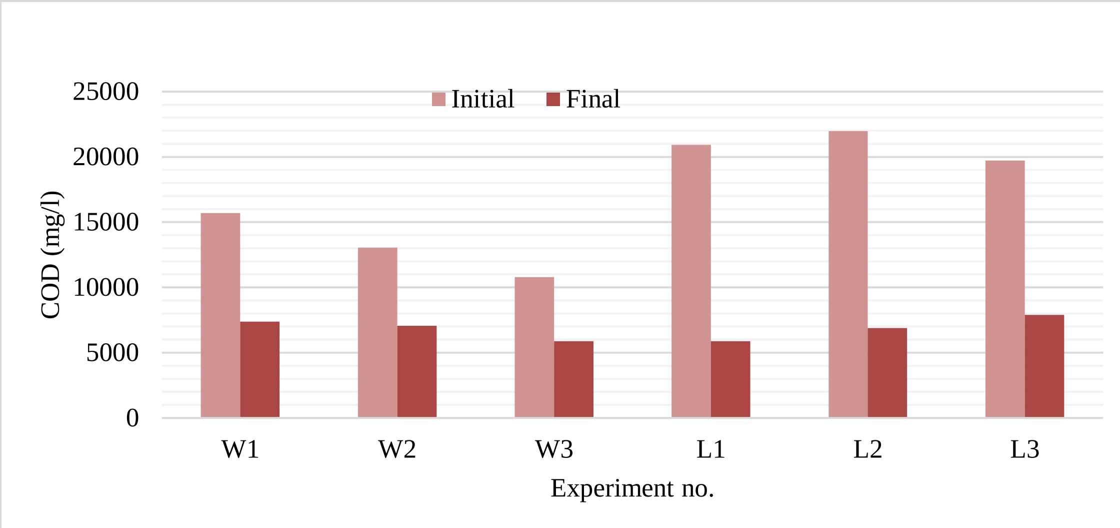

Figure 13: COD removal in various digesters

4.2 Biogas Production from Raw Samples

Anaerobic digestion from single substrates and co-digestion of cow dung with leachate and cotton gin were studied through a single-phase pilot scale anaerobic digestion reactor. The production rates from single substrate such as cow dung, cotton gin and leachate were 110 ml/vs.g, 165 ml/vs.g and 80 ml/vs.g respectively. The digestion of leachate and cotton gin with cow dung utilized the inherent cellulose degrading microbes and the supplementary nutrients in the dung for efficient digestion of fiber in present in cotton gin, Corral et al (2008). The co-digestion of leachate and cow dung has an evident synergistic influence which compensates the missing nutrients thus improving the biodegradation process. This outcome was increased methane yield compared to anaerobic digestion of cow dung alone.

The production of biogas from raw leachate recorded for 45 days however the production lasted for 39 days. The results showed in the figure 10 have an ideal bell shaped graph because of the controlled pH 7.2, 6.9 and 7.4 for sample 1,2 and 3 respectively. The volume produced from all three samples of raw leachate was similar throughout since the samples were from the same source and had same chemical and biological characteristics. gas started to produce from the first day which was due to the presence of trapped air the digester. The maximum volume of biogas recorded was 170ml. the exponential production of biogas. as achieved between days 15 to 21 followed by a lag phase began and there was a drastic decrease in the gas volume. The production from all three of the samples ceased on day 39.

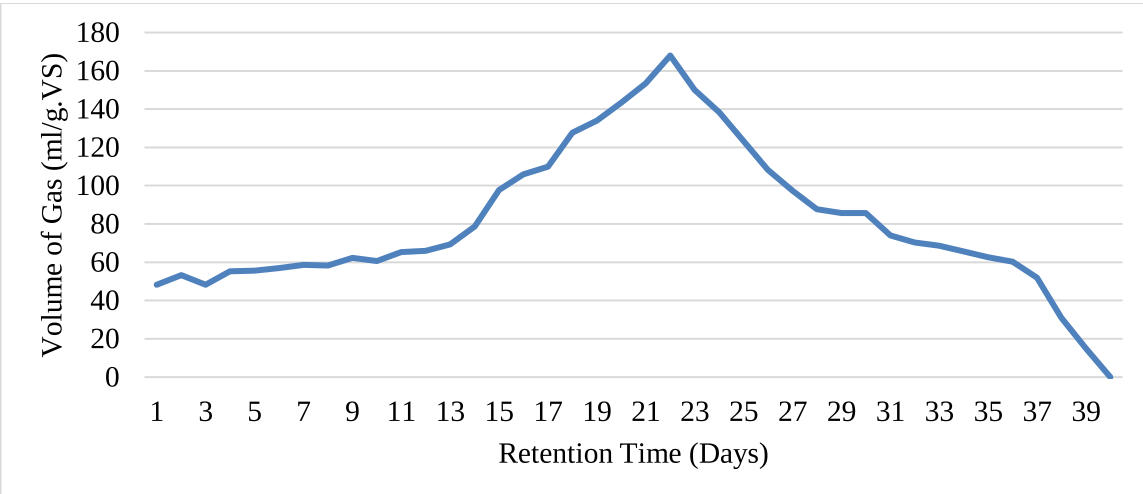

Figure 14: Average biogas production from leachate alone

The production of biogas from cotton gin was recorded after the first 24 hours. The volume of gas produced was 43, 42 and 46 ml/g-VS, 42ml/g-VS and 46ml/g-VS for sample 1,2 and 3 respectively; this was due to the presence of trapped air the digesters thus the values from the first 24 hours were not added in the table. There was a gradual increase in the production of biogas with time. Graph below shows the gas production for 31 days. The lag phase was not achieved and the production continued to increase with increasing retention time. The potential of the production of biogas from cow dung is thus high and the maximum amount of gas produced from raw sample was 170ml/g-VS (the value indicated by the arrow in the figure above) having controlled conditions (temperature and pH). There was no production of gas for the first 4 to 5 days from the cotton gin samples. The maximum amount of gas produced from sample 1, 2 and 3 were 110ml/g-VS, 105ml/g-VS and 110 ml/g-VS respectively as displayed in the figure 4.6 below. However, the production lasted for 25, 28 and 29 days from sample 1,2 and 3 respectively.

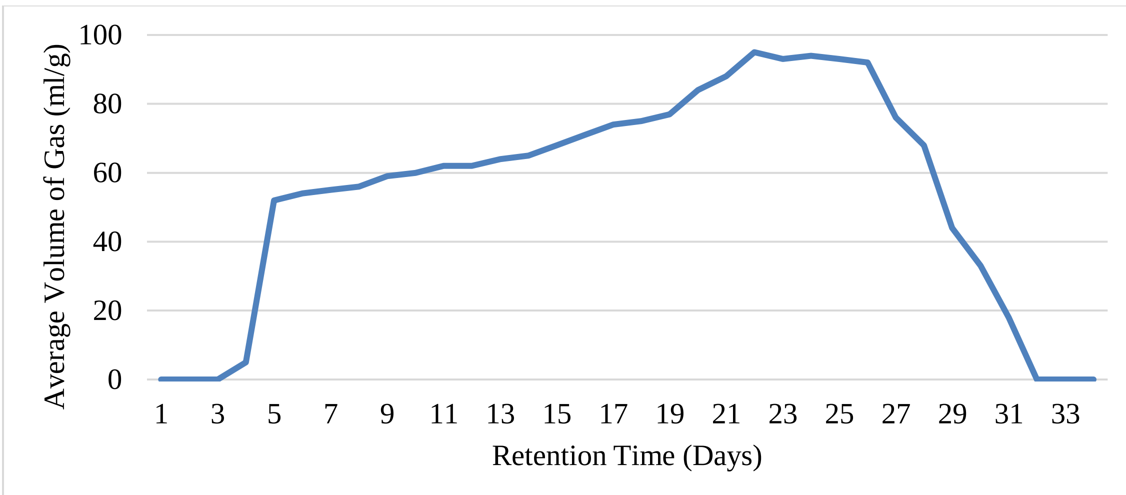

Figure 15: Average biogas production from cotton gin alone

4.3 Cumulative Biogas Production

The result obtained during the cumulative biogas production for the test run and main run along with their correlation between different parameters are discussed in this section.

4.3.1 Cumulative biogas production from test run

The experiment was carried out in ambient temperature range of 26°C-36°C and influent constant temperature of 34°C during a retention period of 45 days.

As mentioned previously, in the test run a total of 484.4g of cow dung, 135.6g of cotton gin and 1800 ml of leachate were added into the digesters. The biogas production started on the second and third day of the experiment in all the six digesters. After reaching the peak, gas production started decreasing. This increase and decrease of gas showed a decent bell shaped trend which normally happens in batch digestion.

The experiment concluded after 45 days. Gas levels over the first 24 hours were not recorded. This gas is also typically not necessary as most of the gas is composed of sulfides in the air trapped in the digester at the beginning of the digestion. Therefore, the data collected shows 43 days of the digestion. Gas analysis after the first 2-3 days period showed a steady increase in methane content. Cumulative gas production rates are shown below.

Average daily production rates are higher in the digesters containing leachate as compared to the samples containing water. Highest production of biogas was observed in the sample containing 1:7 cow dung dissolved in leachate followed by 1:2.5 leachate sample which had the highest proportion of cotton gin. The third highest production was again in the leachate sample having cotton gin and cow dung in 1:5 proportion. A similar trend of gas production is found in all three of the water samples where the digester containing highest proportion of cow dung i.e. 1:7 produced maximum biogas followed 1:2.5 and 1:5 at the lowest for the first 10 days however there was a slight shift in the trend afterwards when the maximum production was that of 1:7 sample.

The maximum amount of gas production recorded was 600ml/g-VS in the 1:7 leachate sample followed by 525ml/g-VS in 1:2.5 leachate owing to a great potential of cotton gin to produce biogas. This sample contained the highest amount of total solid that was 110.5 grams of solid content where the amount of cotton gin was 10.5 grams. Again, in the water samples the digester containing the highest proportion of cotton gin showed the highest startup volume (100ml) collected the second day. In a similar study conducted by Collar (2008) the production of biogas from cow dung was increased with the co-digestion of cotton gin waste and municipal solid waste where Cow dung alone produced $64.8m^3$ and $17.1\ m^3$ during run 1 and 2 respectively however the synergy of municipal solid waste the production raised up to $96.6\ m^3$

The amount of liquid was raised from 600ml to 900ml and that of cow dung and cotton gin was made up to 150 grams in each digester. The overall increase in the amount of substrate was 25% for all three of the digester to ensure the uniform ratios for the main run. The new quantity of material present in the digester filled the digester above 75% of its capacity. This was done to prevent the air from entering the digesters.

4.3.2 Cumulative biogas production from main run

The production of biogas for the retention time of 45 days was measured for the main batch. Maximum amount of gas was produced from a leachate sample in 1:2.5, followed by 1:7 leachates and lowest in 1:5 leachate sample the volume of gas produced was 1350ml, 1100 and 110 ml respectively. The same trend was observed in all three of the water samples where the highest volume produced was 1100 ml from 2:5, 900ml from 1:7 and 90ml from sample having cotton gin and cow dung in the ratio 1:5.

The production of biogas in leachate samples was higher than that of water samples. The samples containing cotton gin and cow dung in ration 1:2.5 from both groups; water and leachate produced the highest amount of biogas followed by 1:7 and 1:5 being at the lowest production rate.

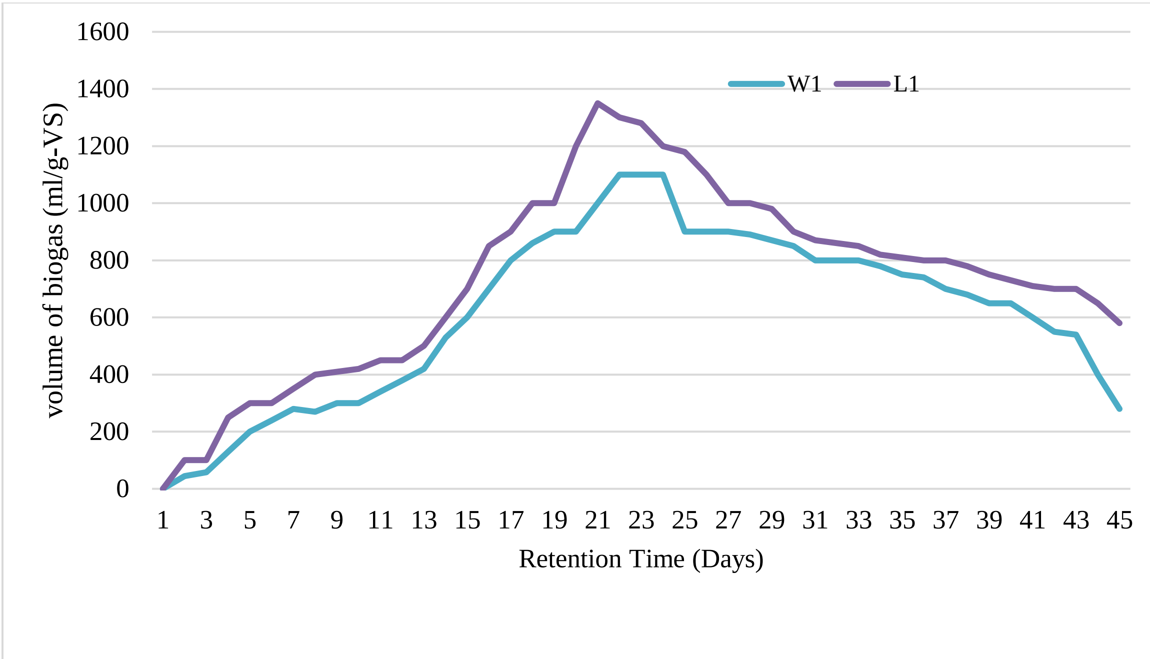

Figure 16: Biogas production from combination 1 (1:2.5)

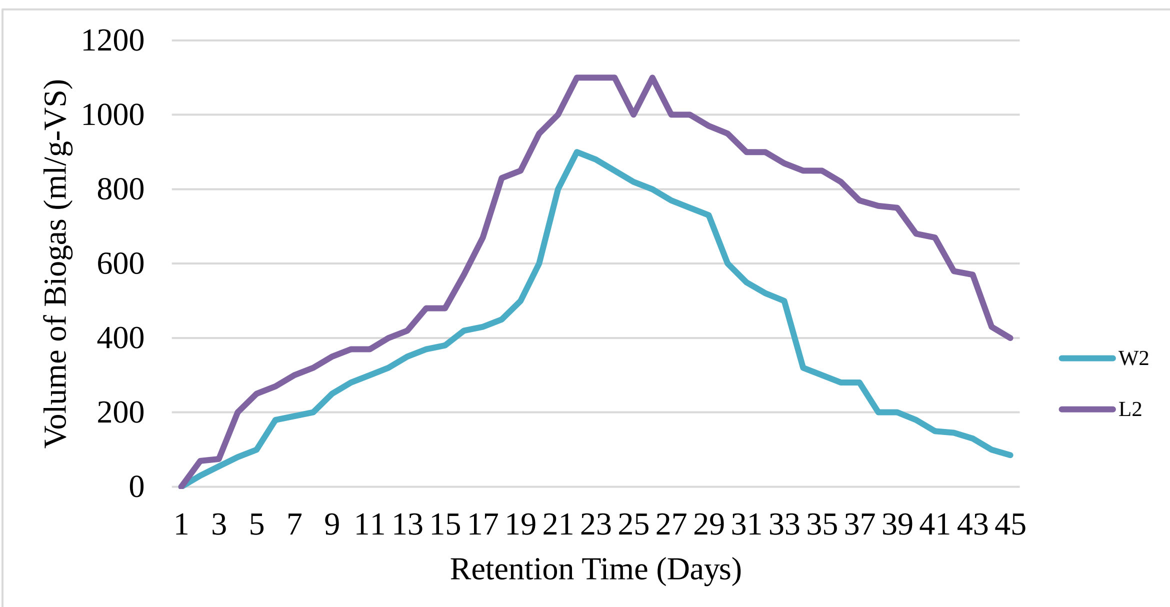

Figure 17: Biogas production from combination 2 (1:5)

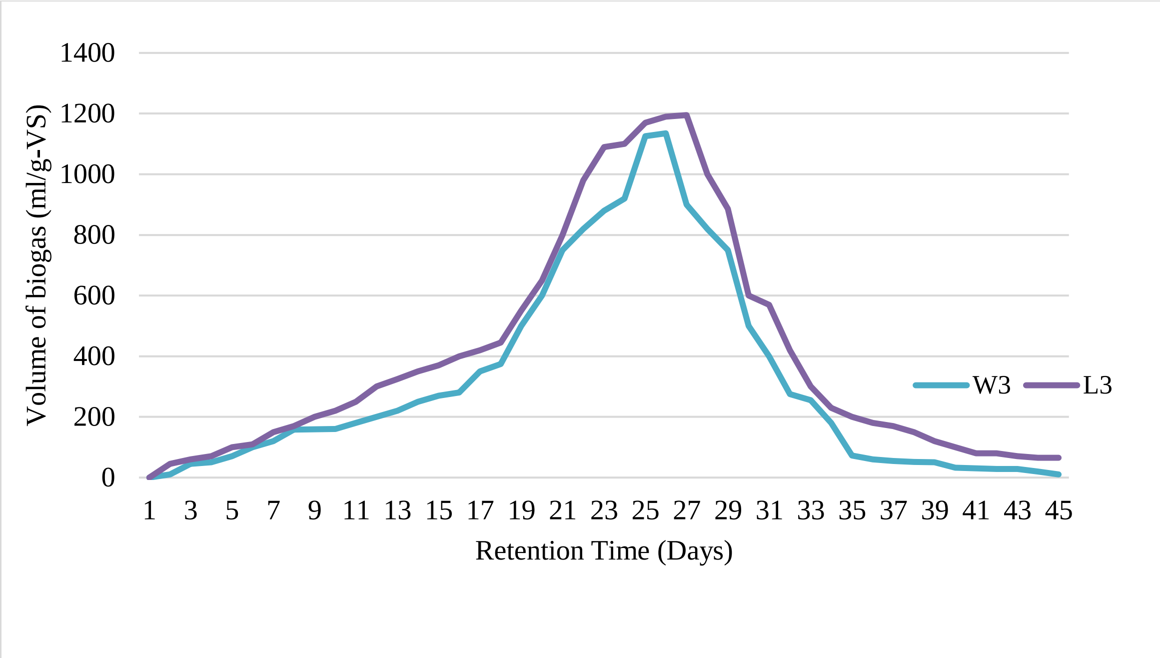

Figure 18: Biogas production from combination 3 (1:7)

4.3.3 Methane gas analysis through flame test

Flame test was carried out simply by lighting the lighter at the neck opening of the bag. Blue flame was observed. As indicated by Mandal et al. (1999) the change in the methane content present in biogas effects the burning of the flame. The flame was obtained due to increased production of biogas. The experiment displayed that this method of biogas generation from the combined digestion of cotton gin and leachate was effective and that the biogas produced can help in domestic energy supply for cooking and heating purpose. The figure 14 is showing the flame being burnt to analyze the biogas.

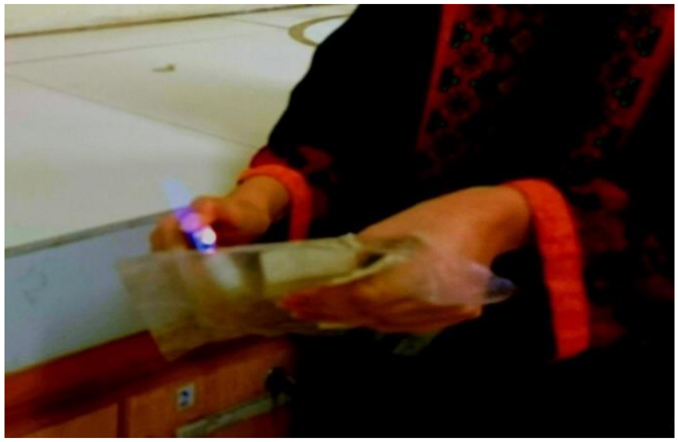

Figure 19: Methane gas analysis through flame test

4.4 Quantitative analysis of Biogas production through GC-MS

The following results were interpreted using this similar study which shows chemical ionization of reagent gases, methane and ammonia. The graphs from this study show and compare both the chemical and electron ionization of these gases. Methane gives 5 different peaks; at 15 and 16, at 17, 29 and 41. The peaks at 15 and 16 show electron ionization of methane while the other three peaks represent chemical ionization i.e. CH^{5+} with a peak at 17, $C2H^{5+}$ with a peak at 29 and C_3H_5 with a peak at 41.

Similarly, the graphs of ammonia give six peaks; at 16 and 17, and at 18,35, 52 and 69. The peaks at 16 and 17 show electron ionization of ammonia while the rest of the peaks represent chemical ionization i.e. NH^{4+} with a peak at 18, $[(NH_3)^{2+}H]^+$ with a peak at 35, $[(NH_3)^{3+}H]^+$ with a peak at 52 and $[(NH_3)^{4+}H]^+$ with a peak at 69.

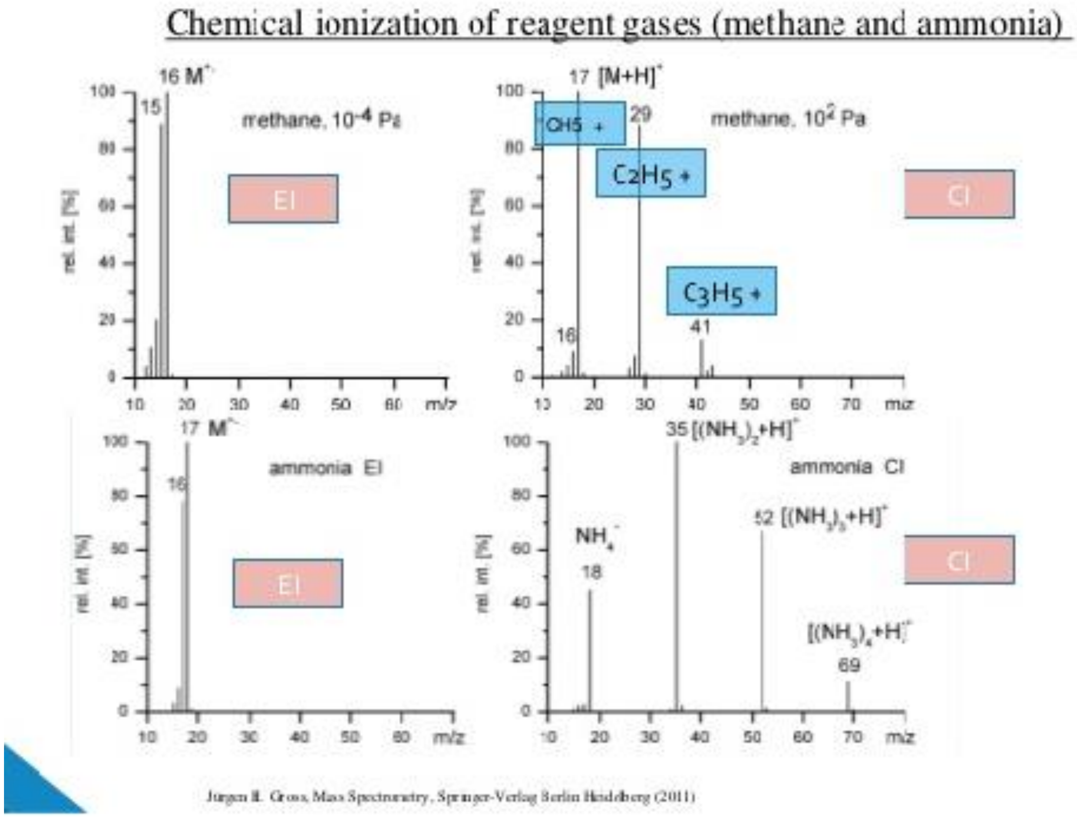

Figure 20: Chemical ionization of reagent gases (methane and ammonia)

The identification of Methane, Carbon dioxide and other trace gases was analyzed through GC-MS. All the gases were identified through TIC (total Ion current). The peaks for Methane and Carbon dioxide were then further identified by SIM (specific ion measurement) technique using the same GC-MS equipment. Figure below shows the Methane Peak at 0.31 minutes.

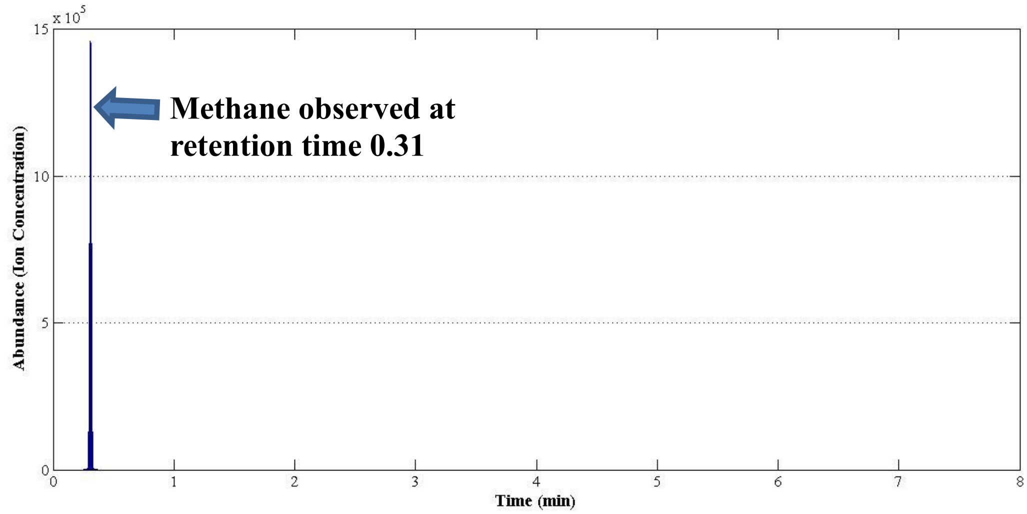

Figure 21: Gas analysis through GC-MC: methane peak as observed

Figure 18 below shows peaks for Methane and Carbon Dioxide at retention time 0.31 and 2.34 respectively. The graph represents the results for sample having cotton gin and cow dung in 1:2.5 ratio in leachate. The sample resulted in the highest percentage of CH_4 production i.e., 67.8% and that of CO_2.

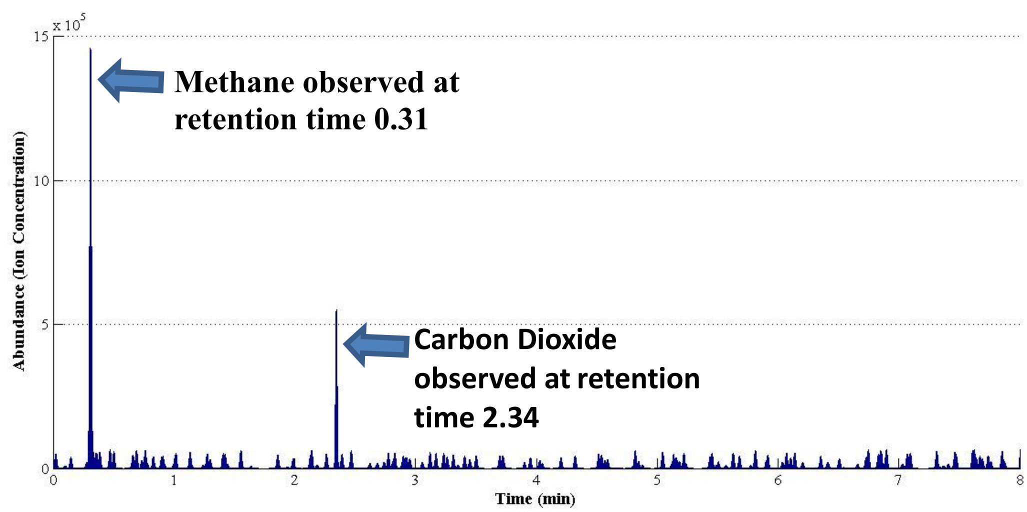

Figure 22: Gas analysis through GC-MS: methane and carbon dioxide peaks

Table 14: Molecular weight and retention time of gases

Compound	Molecular Weight (gm/mol)	Retention Time (min)
Methane CH_4	16	0.31
Carbon Dioxide CO_2	44	2.34

Percentage of CH_4, CO_2 and trace gases for all six samples is presented in the table below.

Table 15: Percentage of methane, carbon dioxide and trace gases

Experiment no.	CH_4	CO_2	Trace Gases
L1	67.8	24.3	7.9
L2	50.2	42.3	7.5
L3	54.7	38.5	6.8
W1	32.8	57.6	9.6
W2	28.3	61.9	9.8
W3	34.5	56.4	9.1

We can draw a solid conclusion from the analyses of the gas samples obtained because of co-digestion that; an overall greater increase in gas volume produced and decrease in volatile solid mass was observed in the substrates prepared in leachate as compared to those prepared in water. Which affirms both of our initial hypotheses.

Chapter 5

CONCLUSION RECOMMENDATIONS

5.1 Conclusion

Co-digestion resulted in 9 times higher biogas production than that of cow dung along. The highest percentage reduction in VS was found in the sample containing cotton gin and cow dung in 1:2.5 ratio in leachate (80.3 % VS reduction) thus giving out the maximum amount of gas i.e., 67.8 %. sample 1:2.5L contained maximum percentage of methane 67.8 %. 1:7 L produced 54.7 % methane followed by sample 1:5 L with 50.2 % production rate. Samples containing water resulted in lower percentage reduction in VS that signifies that relatively a lesser amount of digestible material was converted into gas. Percentage reduction in COD was highest in leachate sample. All three leachate samples ranged from 61.4 % to 71.2 % reduction. Highest COD reduction among the leachate samples was found in the one having cotton gin and cow dung in 1:7 ratio followed by 1:2.5L and 1:5L. For the same ratios in water the percentage reduction ranged from 44.5 % to 57.3 %.

5.2 Recommendations for Future Studies

Noted in this experiment is the fact that while the procedure concluded after 45 days of testing, gas production could have continued. For a full view of the methane production potential of these mixtures, the experiment could be run similarly for a longer period. This can also be done through a reduction of the amount of solids introduced into the reactors at the start of the experiment.

Future studies in the anaerobic digestion and co-digestion of these materials can focus on further optimizing the digestion environment, through nutrient addition or additional pre-treatment strategies. Nutrient balance could have an impact on the actions of the waste within the system as well as the process of microbial digestion. Different studies can also be conducted under different temperature ranges. This study was conducted under mesophilic conditions.

The experiment conducted focused on lab-scale batch operations. If this procedure is to be expanded, a pilot-scale experiment would show the viability of producing methane efficiently from co-digestion. Obtaining a steady supply of digestion material, producing a stream of methane, and potentially switching to a continuous system can all provide studies into the possibility of adapting this data to a larger scale. In this way, a potential efficient methane source could be derived.

Chapter 6

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