PRODUCTION OF BIOGAS FROM CO-DIGESTION OF GOAT MANURE AND WATERMELON (*CITRULLUS LANATUS*) FRUIT PEELS

M. SC. THESIS

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Biogas Production from Co-Digestion of Goat Manure and Watermelon (*Citrullus Lanatus*) Fruit Peels Under Anaerobic Condition

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APPROVAL SHEET

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DEDICATION

This thesis manuscript is dedicated to my dear father Mitiku Yeshanew, my dear Mother Azawint Fenta and my dear friend Addisu Tilahun, who made me the man of my dreams.

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STATEMENT OF THE AUTHOR

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BIOGRAPHICAL SKETCH

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LIST OF ACRONYMS AND ABBREVIATIONS

AD	Anaerobic digestion
ANOVA	Analysis of variance
APHA	American Public Health Association
FAO	Food and Agricultural Organization
GM	Goat manure
HRT	Hydraulic retention time
LDS	least significant difference
M.a.s.l	Meter above sea level
MC	Moisture content
NBP	National Biogas Program
OC	Organic carbon
OLR	Organic loading rate
TS	Total solids
USDA	United State Department of Agriculture
VS	Volatile solids
WMP	Watermelon peel

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Biogas Production from Co-Digestion of Goat Manure and Watermelon (*Citrullus lanatus*) Fruit Peels under Anaerobic condition

ABSTRACT

Biogas is an alternative source of energy that is produced by methanogenic bacteria through the bio-degradation of organic material under anaerobic conditions. In this study, biogas production from co-digestion of the goat manure (GM) and watermelon peels(WMP) in five mix ratios were evaluated under 38ºC using batch fermentation. In all treatments, physico-chemical parameters such as total solid (TS) and volatile solid (VS), organic carbon, and pH were measure before and after digestion. The daily biogas production was subsequently measured by water displacement method for 27 days. All measured of physico-chemical parameters of each substrate significantly varied ($p<0.05$) between before and after anaerobic digestion. Gas production was noticed in all of the digesters from the third day of digestion and went down to zero at about the 23rd days in all substrates. Assessment of cumulative biogas production revealed that the substrate mix ratio of 30% GM and 70% WMP showed the highest production, suggesting this mix ratio of the two substrates is an optimal mix to yield 242.0ml biogas. Overall the results indicated that the increment of biogas yield can be significantly enhanced when GM and WMP are co-digested.

***Keyword**: Biogas, Co-digestion, Manure, Slurry*

1. INTRODUCTION

Globally, the demands of energy consumption are continuously increasing from time to time with increasing of human populations. To overcome these problems, an alternative energy sources have recently become more and more attractive due to the increasing energy demands, the limited resource for buying fossil fuel, the environmental concerns and the strategy to survive post-fossil fuel economy era (Wilkie, 2008). So there is an urgent need for the research of new renewable energy resources such as biogas, solar energy, wind energy, thermal and hydropower sources of energy. Among the alternative energy sources, biogas production from green energy crops and organic wastes has world wide application as it yields a good quality fuel and fermented slurry. It has a very positive impact for the environment since less carbon dioxide is released during its combustion. The use of renewable energy sources can contribute to solve the present and future energy problems (Rilling, 2005).

The alternative energy sources are becoming increasingly important to replace the potentially expensive fossil fuels. Nowadays, they are getting great focus worldwide because of concerns to sustain energy security, climate change and the need to support local agriculture (Maria, 2008). Biogas is a well-established fuel that can replace firewood as an energy source for cooking, lighting, heating and for other purposes in developing countries. Currently, as the fossil-based fuels become scarce and more expensive the economics of biogas production is turning out to be more favorable. Biogas significantly reduces the greenhouse-gas emission compared to the emission of landfill gas to the atmosphere (Aremu and Agarry, 2013).

Biogas is a colorless, flammable gas produced from anaerobically digested animal, plant, human and industrial wastes to give mainly methane (50-70%), carbon dioxide (20-40%) and other trace gases such as nitrogen, hydrogen, ammonia, hydrogen sulphide, water vapors etc. It is an alternative and renewable energy source produced by the decomposition of organic matter through anaerobic digestion process, in which an interlaced community of bacteria cooperate to obtain fermentation through assimilation, transformation and decomposition of organic matters into biogas (Ofoefule *et al*., 2010).

Biogas consists primarily of utilizable methane (CH_4) and inert carbon dioxide (CO_2), which are both colorless and odorless but if H_2S amount increases, it causes danger for users. Methane has 20 times more greenhouse gas potential than carbon dioxide, so the capture and burning of methane significantly reduces the greenhouse gas effect (Atkins *et al.,* 2008). Biogas burns very well when the CH_4 content is more than 50 %, and 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 from cutting trees to get firewood (Ayoub Mohamed Eshraideh *et al.,* 2002).

Energy is one of the most important factors to global prosperity. The global mix of fuel comes from fossil (78%), renewable (18%) and nuclear (4%) energy sources. The dependence on fossil fuel as primary energy source has led to global climate change, environmental degradation and human health problems. About 80% of the world's energy consumption still originates from combusting fossil fuels (Goldenberg and Johansson, 2004). Yet the reserves do not match with the fast population growth and their burning substantially increases the greenhouse gas concentration and contributes to global warming and climate change. So, energy production from biomass, which is called bio-energy, can be seen as one of the key options (Schamphelaire and Verstraete, 2009).

Many bio-energy related processes are being developed; those processes involving microorganisms are especially promising as they have the potential to produce renewable energy on a large scale without disrupting strongly the environment or human activities (Rittman, 2008). Organic substances exist in a wide variety from living things to dead organisms and it is composed of carbon combined with elements such as hydrogen, oxygen, nitrogen and sulpher to form variety of organic compounds such as carbohydrate, protein and lipid. It usually contains 50% and above methane and other gases in a relatively low proportion namely CO_2, H_2, N_2 and O_2 (Kalia *et al*., 2000).

Deforestation is a very big problem in developing countries like Ethiopia where, people cut trees for wood fire, construction and expansion of arable land. As more and more trees are cut down in unsustainable manner, the impact on the environment including soil erosion, decreasing soil fertility, land slide, etc. will become significantly visible. Furthermore, use of crop residues and dung cakes as a substitute for fuel wood reduces soil fertility and agricultural productivity (Asnake *et al.,* 2006). Use of manure as fire wood for source of energy is also harmful for human health since the smoke arising from the burning cause air pollution. Thus, the situation calls for the use of an eco-friendly substitute such as biogas for energy source (Dawit, 2011).

Any kind of Biomass that contains organic matter like carbohydrates, fats, proteins …etc can be used as a substrate for biogas production. In higher institutions the production of wastes like kitchen wastes and animal wastes are highly available for the production of biogas. In Haramaya University, for example, there is a huge amount of solid animal excreta from cattle, pig, goats, sheep, and the poultry farm. In addition, a large quantity of different fruit peels are disposed off daily from households and/or fruit market areas of Bate, Harar, Jigjiga, Dire Dawa towns etc. Indeed, biogas production from different organic materials such as *Moringa stenopetala* seed cake powder (Eyasu, 2008), poultry litter (Ebrahim, 2006), Khat (*Catha edulis*) waste (Tesfaye, 2007), banana peels (Belayhun, 2014) and others was performed in Ethiopia. However, no research has been done so far on the effect of the co-digestion of goat manure and watermelon fruit peels on biogas production. Watermelon fruit peel is one of the agricultural wastes that may be used as good potential source of biogas production. This research was, therefore, meant to evaluate watermelon peel for biogas production in sole and combined with goat manure in different proportions with the following objectives.

General objective

- ❖ The general objective of this study was to produce biogas from watermelon fruit peels waste and goat manure combined or separately under anaerobic condition.

Specific objectives

- ❖ To characterize goat manure and watermelon waste peels in terms of total solids, volatile solids, pH, and organic carbon content before and after anaerobic digestion.
- ❖ To find out the optimum mix ratio of goat manure and watermelon waste fruit peels yielding high biogas.

2. LITERATURE REVIEW

2.1 Biogas Production

Biogas is a methane rich gas produced by anaerobic breakdown of organic wastes with the help of archaebacteria under oxygen free environment and it comprises 60% of methane, 40% of carbon dioxide and 0.2 - 0.4% of hydrogen sulfide (Molina, 2007). The natural generation of biogas is an important part of the biogeochemical carbon cycle. Archaebacteria are the last link in a chain of micro-organisms which degrade organic material and return the decomposition products to the environment. In this process biogas is generated, as a source of renewable energy (Werner Kossmann, 2000).

It is a flammable gas made of a mixture of gases produced by methanogenic bacteria while acting upon biodegradable materials in an anaerobic condition. Biogas is a colorless gas that burns with clear blue flame. It is about 20% lighter than air and has an ignition temperature in the range of 650° to 750° C (Claude, 2009).

Biogas consists of mainly methane, carbon dioxide, hydrogen sulfide and traces of other gases (Table 1). Methane is produced by the anaerobic breakdown of organic materials including agricultural wastes, organic kitchen wastes etc (Werner Kossmann, 2000). Biogas producing microorganisms include organic material splitting bacteria and archaebacteria that degrade complex organic materials to produce methane under anaerobic conditions (Claude *et al*., 2009). The resulting bio-slurry, which is used as fertilizer has a reduced load of parasitic diseases and pathogenic bacteria for crop production (Forum for Environment, 2010).

Biogas production is a simple technology that helps to reduce the use of forest resource for hold house energy consumption, and hence prevents deforestation (Dagnachew *et al*., 2003).

Table 2.1 Chemical compositions of biogas (Source: Madu and Sodeinde (2001).

Constituents	% Composition
Methane	55-75
Carbon dioxide	30-45
Hydrogen sulphide	1-2
Nitrogen	0-1
Hydrogen	0-1
Carbon monoxide	Traces
Oxygen	Traces

2.2 Biogas production for sustainable environments

Replacing biomass energy with biogas could help to solve a lot of problems in the environment that are typically associated with using biomass fuels. The indoor air quality of homes will be dramatically improved as a result of employing biogas instead of burning biomass directly (Li *et al*., 2005). Substituting biogas for firewood also helps to reduce the pressure on forests for energy demand. This in turn has important implications for watershed management, slowing down deforestation and soil erosion. This in turn maintains water cycle and avoids recurrent drought (Dagnachew *et al*., 2003).

The use of slurry improves nutrient recycling in agriculture and can substitute chemical fertilizers, thus reducing the related environmental problems (Shrestha and Huong *et al.,* 2010).

2.3 Biogas technology in Ethiopia

Biogas technology is a promising option for the most efficient utilization of organic waste in a fermentation tank. It involves organic matter, micro organisms, an environment that lacks air (oxygen) and optimum temperature to produce biogas. Biogas technology also has various benefits for human beings, such as socio-economic and environmental benefits. The environmental benefits of biogas technology include improvement of indoor air quality, better management of animal manure and human excreta, thus improving sanitary conditions in the immediate vicinity of the rural homes. Through reduction of deforestation, the technology also helps in a better watershed and soil management. The use of slurry helps in improving the soil nutrient and use of biogas for energy also helps in reserving the expenditure of imported petroleum products. At a global perspective, reduction in the use of fuel wood, dung cakes and kerosene reduces greenhouse gas emission (Shakya and Charushree, 2009).

In Ethiopia, biogas technology was introduced in 1979 and the first batch type of biogas digester was constructed at Ambo Agricultural College. However, the technology was less disseminated until the National Biogas Program (NBP) was launched in 2008 (Eshete *et al*., 2006). Much of the energy derived from biogas technology had been allocated for household energy consumption since its introduction to Ethiopia in 1979 (Siltan, 1989). Currently around 40 % of these biogas plants are not operational due to lack of effective management, technical problems, loss of interest, reduced animal holdings, evacuation of ownership and water problems (NBP, 2007).

After the establishment of national biogas program, close to 859 biogas plants have been constructed and in a regular use. Out of these 206 were in Tigray region, 143 in Amhara region, 330 in Oromiya region and 180 are found in SNNP regional state (Claudia and Yitayal 2011). Getachew *et al.* (2006) pointed out that at least over one million households in Amhara, Oromiya, Tigray and Southern Nations, Nationality and People's Regional States have the potential for the installation of a domestic biogas plant. The domestic biogas technology attracted interest mainly due to consideration of animal dung, the raw material that is plenty in many rural households of the country.

2.4 Feedstock for biogas production

Though there can be variation in biogas production potential among feedstock, all organic materials can serve as substrate for biogas production in sole or in combination. Plant materials and animal manure have recently been used for production of biogas by co-digestion under anaerobic condition. The co-digestion of plant material and animal manure increases the rate of biogas production as compared to the sole digestion of feedstock. Because mixing substrates was found to balance between carbon and nitrogen ratio (Elmashad *et al*., 2004).

In this study, co-digestion of goat manure and watermelon waste peels have been considered for biogas production under anaerobic condition in sole and mixed in different ratio. Watermelon (*Citrullus lanatus,* Cucurbitaceae) was originated in southern African countries and spread to all over the tropical and subtropical regions. In hot parts of Ethiopia such as in Somali regional state, it is highly cultivated.

2.5 Anaerobic Digestion for biogas production

Anaerobic digestion is a process of controlled decomposition of biodegradable materials under managed conditions where oxygen is absent. It is a complex process that requires specific environmental conditions and different bacterial populations. The bacterial populations degrade organic compounds so as to produce a valuable high energy mixture of gases (mainly CH_4 and CO_2) and a nutrient rich fertilizer (Sandars *et al*., 2003). The process usually prefers mesophilic or thermophilic condition for anaerobic archaebacteria to convert the inputs into biogas (Steinmetz *et al*., 2013).

According to Li *et al*. (2009) anaerobic digestion is considered as waste-to-energy technology and it is widely used in the treatment of different organic wastes. It is consists of mixed biological systems in which organic materials such as carbohydrate, lipids and proteins are utilized by microorganisms to produce methane and carbon dioxide-rich in their normal metabolic activities. The anaerobic digestion involves a large number of microorganisms including hydrolytic bacteria, acetic acid-forming bacteria and methangoenic bacteria, which convert the feedstock to the methane and carbon dioxide rich biogas through a number of different processes (Cibowski, 2004).

2.5.1 Hydrolysis

Hydrolysis is the first step of anaerobic digestion in the degradation of large organic matter like polysaccharide, protein and fat into their monomers, such as sugars, amino acids and fatty acids using water as a medium of reaction (Parawira, 2008). This is formed by extra hydrolytic enzymes. The hydrolytic enzymes include cellulase, hemi cellulose amylase, lipase and protease (Parawira, 2008). Many cellulose degrading organism have their enzymes attached to the cell wall and simultaneously they attach to the substrate for more effective degradation. The hydrolysis of complicated structure such as lingo-cellulose requires weeks (Gerardi *et al*., 2003). As such hydrolysis is time limiting step, while the methanogenesis is considered rate-limiting step for already available substrate (Vavilin *et al*., 2008).

In hydrolysis, complex organic substances are converted to simple ones. For example, carbohydrate to sugar, fats to fatty acids and protein to amino acids by hydrolytic bacteria. This step takes longer time due to limiting accessibility of the extra cellular enzymes to intra cellular polymeric materials which are protected by cell covering (Nava *et al.,* 2002; Kim, 2003).

2.5.2 Acidogenesis

During acidogenesis, the monomers formed in the hydrolysis stage are taken up by anaerobic bacteria and degraded in the acidogenic stage. The aim of this stage is that to degrade the results in hydrolysis stage into shorter chain and convert into alcohol, hydrogen, ammonia, carbon dioxide and organic acid such as butyric acid, propionic acid, acetic acid. An organic acid produced in this stage is called intermediate products (Gerardi, 2003).

2.5.3 Acetogenesis

Products from acidogenesis are converted to CH_3COOH, H_2 and CO_2 by acetogens. These products are formed from organic acids. In the acetogenesis process simple molecules are created through the acidogenesis phase are further digested by acetogens to produce largely acetic acid as well as carbon dioxide and hydrogen. Acetogenic organisms are the vital link between hydrolysis-acidogenesis and the methanogenesis in anaerobic digestion. Acetogenesis provides the two main substrates for the last step in the methanogenic conversion of organic material, namely hydrogen and acetate (Buswell and Sollo, 1948).

2.5.4 Methanogenesis

The production of methane and carbon dioxide from intermediate products is carried out by methanogenic archaebacteria. Formic acid, acetic acid, methanol and hydrogen can be used as energy sources by the various methanogens (Dhadse *et al*., 2012). The formation of methane is the ultimate product of anaerobic treatment. The acetoclastic group comprises two main methanogenic bacteria (Zaher *et al.,* 2007).

High concentration of hydrogen disables acetogens to act and that is why it is of importance that methanogenic bacteria use the hydrogen (Bie, 2002). . The vital functions of these archaea bacteria are that to consume hydrogen in stable temperature modes (Zorg Ukraine Biogas Plants, 2009).

The methane former works slower than that acid former, therefore, the pH has to stay constant consistently, slightly basic, to optimize the creation of methane. One needs to constantly feed in sodium bicarbonate to keep pH slightly basic (Prebeheim *et al.,* 2010). This compound involves in the conversion of simple compounds (acids) into methane and CO_2 by CO_2 utilization anaerobic methanogenic bacteria (Ito, 2001).

2.6 Factors that affect the rate of Anaerobic Digestion

Environmental factors which influence the process of biological reaction amenable to the external control in the anaerobic process. Any drastic change in these factors can adversely affect the biogas production (Chatterjee, 2007). The performance of anaerobic digestion plants can be controlled by studying and monitoring the various parameters (Yadvika, 2004).

2.6.1 Temperature

Anaerobic digestion may be carried out under psychrophilic, mesophilic or thermophilic conditions. In the sewage sludge mesophilic anaerobic digestion is more widely used compared to thermophilic digestion, because of the lower energy requirements and higher stability of the process. However, thermophilic digestion is more efficient in terms of organic matter removal and methane production (Ahring *et al*., 2001).

Methane production has been documented under a wide range of temperatures but archaebacteria are most productive in either mesophilic conditions 25-45°C, or in the thermophilic conditions at 50-65°C (Ostrem *et al*., 2004). A thermophilic temperature reduces the required retention time. The microbial growth, digestion capacity and biogas production could be enhanced by thermophilic digestion, since the specific growth rate of thermophilic bacteria is higher than that of mesophilic bacteria (Kim and Speece, 2002).

2.6.2 pH value

The acidity of substrate is measured by pH meter, which is an important parameter affecting the growth of microbes during anaerobic digestion. The acid concentration in aqueous systems is expressed by the pH value, i.e., the concentration of hydrogen ions (Yadvika *et al*., 2004). pH values below 6.8 inhibit the archaebacteria activity. To avoid drops in pH chemicals are added to the organic substrate to producing a buffer capacity. Sodium bicarbonate, sodium hydroxide, sodium carbonate and sodium sulphide are the most used chemicals (Esposito *et al*., 2012).

pH value of 7 is neutral, less than 7 is acidic and more than 7 is alkaline. During anaerobic fermentation, microorganisms require a neutral or mildly alkaline environment for efficient gas production. Biogas production needs an optimum pH value of 6.8 to 7.2 (Mahanta *et al*., 2004). The rate of methanogenesis may decrease if the pH is lower than 6.8 or higher than 7.8. The pH of the digester is created by concentration of volatile fatty acids produced, bicarbonate alkalinity of the system and the amount of carbon dioxide produced (Gomec, and Speece, 2003).

2.6.3 Nutrient

All biological processes require sufficient supply of nutrients particularly carbon and nitrogen as well as other elements are also required in trace quantities. The lack of specific elements can limit microbial growth required for the production of biogas (Anunputtikul and Rodtong, 2004). If the manure is in dry form, the quantity of water to be add the digester has increased accordingly to arrive at the desired consistency of the substrate. When organic substance added to a digester that has large amount of water content, then relatively low amount of water is needed to get total solids content (Jan *et al.,* 2010).

2.6.4 Particle size

The production of biogas is also affected by particle size of the substrate. Too big particle size is problematic for microbes to digest and it can also result in blockage in the biodigester. However, small particle size gives a large surface area for substrate adsorption and thus allows the increased microbial activity followed by increase in the production of gas (Yadvika *et al*., 2004).

According to the reports of Tamirat Asnake (2008) out of the five particle sizes (0.088, 0.40, 1.0, 6.0 and 30.0 mm), maximum quantity of biogas was produced from raw materials of 0.088 and 0.40 mm particle size. Large particles could be used for succulent materials such as leaves. However, for other materials such as straw, large particles could decrease the gas production. The results suggested that a physical pretreatment such as grinding could significantly reduce the volume of digester required, without decreasing biogas production (Gollakota, 2004).

2.6.5 Hydraulic retention time (HRT)

Retention time is an incubation period of time that an organic substance stays in biodigester starting from the first day up to the last day of biogas formation. it is average time spent by the biomass inside a continuous biogas plant before it comes out from the digester is known as the hydraulic retention time or it is the time needed to achieve the complete degradation of the organic matter. The required retention time for completion of the anaerobic digestion varies with differing technologies, process temperature and waste composition. The retention time for wastes treated in mesophilic digester range from 10 to 40 days (Demetrides, 2008).

The retention times of mesophilic and thermophilic digesters range are between 25 and 35days (Bouallagui *et al*., 2003). The disadvantage of a longer retention time is that a large reactor size is needed for a given amount of substrate to be treated (Hassan 2003). Although a short retention time is desired for reducing the digester volume, a balance must be made to achieve the desired operational conditions, for example, maximizing either methane production or organic matter removal (Zamudio and Esteban Manuel 2010).

2.6.6 Pre-treatment

The main purpose of pre-treatment is to remove structural impediment for hydrolysis and subsequent degradation process in order to enhance digestibility, improve the rate of enzyme hydrolysis and increase yields of intended products (Hendriks and Zeeman 2009). Physical pretreatment is the most common methods disintegration by grinding and mincing. The plantain peels was grinded in a day to aid faster digestion. It was then soaked in a plastic water bath overnight for partial decomposition of the peels by anaerobic microbes which have been reported to aid faster digestion especially for plant wastes (Fulford 1998).

Biological processes also used as pre treatment for disintegration takes place by means of lactic acid which decomposes complex components of certain substrates. Recently also disintegration with enzymes has been quite successful, especially using cellulase, protease or carbohydrases at a pH of 4.5 to 6.5 and a retention time of at least 12 days, preferably more (Hendriks and Zeeman, 2009).

2.6.7 Organic loading rate (OLR)

The rate at which substrate was supplied to the biodigester is referred to as organic loading rate. The gas production rate in the biodigester is highly dependent on the organic loading rate (Yadvika *et al*., 2004). Volatile Solids contains largely carbon, oxygen and nitrogen which burn off from an already dry sample in a laboratory furnace at 500-600°C, leaving only the ash which contains largely calcium, magnesium, phosphorus, potassium and other mineral elements that do not oxidize. Organic loading rat is usually expressed in terms of kilogram volatile solids per m^3 and day (Arogo *et al.*, 2009).

2.6.8 Seeding

To start up a new anaerobic process, it is critical to use inoculums of microorganisms to commence the fermentation process. The common seeding materials include digested sludge from a running biogas plant or material from well-rotted manure pit or cow manure slurry (Yadvika *et al*., 2004). Forster-Carneiro *et al.,* (2007) pointed out that digested sludge is best inoculums source for anaerobic thermophilic digestion of the treatment of organic fraction of municipal solid waste at dry condition. The rumen fluid inoculums caused biogas production rate efficiency increase more than two times in compare to manure substrate without rumen fluid inoculums (Sunarso *et al.,* 2012). Rojas *et al* (2010) states that the addition of manure slurry to the batch reactor as part of the starter improved the biogas production.

2.6.9 Mixing condition (Agitation)

The close contact between micro-organisms and the substrate material is an important for efficient digestion process. The mixing of the digester contents has a number of benefits, one of the most obvious being that it helps to mix up material, evening out any localized concentrations, thus also helping to stop the formation of dead zones. In addition, it increases the waste's availability to the bacteria, helps remove and disperse metabolic products and also acts to ensure a more uniform temperature within the digester. There have been some suggestions that efficient mixing enhances methane production, but the evidence is inconclusive, so it seems likely that this may only be of noticeable benefit for some systems or operational regimes (Gareth and Judith, 2003).

Mixing also promotes heat transfer, particle size reduction as digestion progresses and release of produced gas from the digester contents (Prasad *et al*., 2008). Roa *et al* (2010) showed that there is significant stirring effect on the anaerobic digestion only when seed sludge from a biogas plant was used as a starter. In this case, the experiments without stirring yielded, without starter, only about 50% of the expected biogas for the investigated substrates. On the other hand, vigorous continuous mixing was shown to disrupt the structure of microbial flocks, which in turn disturbs the syntrophic relationships between organisms thereby adversely affecting the reactor performance (Kim *et al*., 2002).

2.6.10 Moisture Content

High moisture contents usually facilitate the anaerobic digestion; however, it is difficult to maintain the same availability of water throughout the digestion cycle. Initially water added at a high rate is dropped to a certain lower level as the process of anaerobic digestion proceeds. High water contents are likely to affect the process performance by dissolving readily degradable organic matter. It has been reported that the highest methane production rates occur at 60–80% of humidity. Methanogenesis processes during anaerobic digestion at different moisture levels i.e., 70% and 80% (Khalid *et al*., 2011).

According to Sadaka and Dan Engler (2003), water content is one of the very important parameters affecting anaerobic digestion of solid wastes. There are two reasons viz.; (a) water make possible the movement and growth of archaea facilitating the dissolution and transport of nutrient and (b) water reduces the limitation of mass transfer of non-homogenous or particulate substrate.

2.6.11 Carbon: Nitrogen (C/N) ratio

Optimum C/N ratios in anaerobic digesters are between 20 – 30 in order to ensure sufficient nitrogen supply for cell production and the degradation of the carbon present in the wastes (Fricke *et al.,* 2005). Hartmann and Ahring (2006) stated that a solid waste substrate with a high C/N ratio is an indication of rapid consumption of nitrogen by archaebacteria and results in lower gas production. On the other hand, a lower C/N ratio causes ammonia accumulation and pH values exceeding 8.5, which is toxic to methanogenic bacteria.

Optimum C/N ratios of the digester materials can be achieved by mixing materials of high and low C/N ratios, such as organic solid waste mixed with sewage or animal manure (Zaher *et al.,*

2007).

For the optimal growth activity of archaebacteria, it is essential that many nutrients are available at the correct chemical form and concentration. Carbon in carbohydrate and nitrogen in protein, nitrates, etc are the main nutrients for anaerobic bacteria. While carbon supplies energy, nitrogen is needed for building up the cell structure. In order to provide an index for the presence of nitrogen in the correct concentrations, the concept of carbon to nitrogen ratio was extended to biogas substrates. Earlier this concept was greatly emphasized and C/N ratio of 15:1 to 25:1 was found to be optimum. This was then considered universal for all substrates. However, with greater understanding in the field, it is clear that the ratio considered should be about the bio-degradability index. Bio-degradable carbon to available nitrogen ratio of 25:1 to 30:1 has been found to be ideal for biogas production (Marchaim 1992). However, the various organic wastes used for biogas production differ unduly in their C/N ratio and hence an optimum mix of the input materials is necessary to get the optimum ratio.

Poultry litter contains a high level of organic nitrogen due to the high content of protein and amino acids. While ammonium ions can be utilized by some members of an anaerobic population, the excess of ammonium can inhibit the destruction of organic compounds, the production of volatile fatty acids (VFA), and methanogenesis (Nailia *et al*., 1979).

2.6.12 Co-digestion process

Co-digestion is the simultaneous digestion of a homogenous mixture of two or more substrates. The most common situation is when a major amount of a main basic substrate is mixed and digested together with minor amount of single or a variety of additional substrate (Mata *et al*., 2000).

Co-digestion is a waste treatment method where different types of wastes are treated together. Co-digestion offers several ecological, technological and economical advantages, so it can improve organic waste treatment through anaerobic digestion. Co digestion with other wastes, whether industrial, agricultural or domestic, has been a successful option for improving biogas production. The process provides improved nutrient balance from a variety of substrates which helps to maintain a stable and reliable digestion performance and produce a good fertilizer quality of the digestion (Feng and Rundong, 2010).

The advantage of co-digestion includes better degradability, enhanced biogas production methane yield arising from availability of additional nutrients, as well as more efficient utilization (Elijah *et al*., 2009).

3 MATERIALS AND METHODS

3.1 Study Area

The study was conducted in Microbiology and Central Laboratory of Haramaya University main campus. The university is found around 510 km away from Addis Ababa in Haramaya district eastern Hararge Oromiya Ethiopia. The University is geographically located at latitude of 9°26’ N, longitude of 42°03’E and an altitude of 1980 m.a.s.l (FAO, 1990).

3.2 Substrate Preparation

Fresh goat manure and watermelon peels were used as substrates for anaerobic digestion. Fresh goat manure was collected from Haramaya university goat farms while WMP was collected from Jigjiga town in Somali Regional State of Ethiopia. The rumen fluid that was used as inoculums to start anaerobic digestion was obtained from slaughter house of Haramaya University. The GM was sun-dried for two days while WMP was chopped and sun-dried for a week. The substrates were then separately grind using mortar and pestle to ensure homogeneity during AD.

3.3 Design of the Experiment

The experimental design was completely randomized design using batch mode in 0.5 L plastic bottle digester. Each digester contained fixed amount of goat manure (GM) and watermelon peel**.** The digester was filled with fresh GM and WMP combined in different proportion or in solo to know the content of biogas production. The combinations were done in five treatment proportions (table 2). To each of the treatment, appropriate amount of distilled water and inoculums’ were added to achieve the recommended (8%) total solid content of digester (Tchobanoglous *et al.,* 1993). Each treatment had three replications. The temperature of biodigester was maintained at 38°C by keeping in oven, which represents mesophilic condition (Knottier (2003). Initial pH values for samples were also maintained 6.8-7.2 the optimum pH range to archaebacteria for biogas production (Thy *et al*., 2003; Yadvika *et al*., 2004).

Table 3.1:-Mixing ratio of the substrates

Digesters	% GM	%WMP
A	100	0
B	70	30
C	50	50
D	30	70
E	0	100

3.4 Digester Configuration and Setup for Biogas Production

Three plastic bottles were arranged in order in such a way that the first bottle contained slurry, the middle bottle contained acidified brine solution and the last bottle was empty for collecting the brine solution that was expelled out from the second container. The acidified brine solution was prepared by addition of 500g of sodium chloride to distilled water and mixing until a supersaturated solution was formed and then 3 drops of sulphuric acid was added to prevent the dissociation of biogas in the water.

All the three containers were interconnected using a plastic tube. The digesters were equipped with tightly sealed accessories in order to control the entry of oxygen and prevent loss of gas outside the bottle (Taeme *et al.*, 2014). The tube connecting the first bottle to the second bottle was fitted just above the slurry in the first bottle to help gas collection. Thus, the biogas produced by fermentation of the slurry was driven from the first bottle to the second bottle that contained a brine solution so as to displace a volume of the brine solution equivalent to the volume of biogas produced. From this setup, daily biogas production was measured by water displacement method using brine solution (Itodo *et al*., 1992).

3.5 Measurement of Biogas

The amount of gas produced was measured by water displacement method using 90% NaCl solution (brine) (Yetilmezsoy and Sakar, 2008). The daily gas production was recorded for the different treatments for 27 days until the gas production ceases.

3.6 Analysis of the Physico-chemical Characteristics of the Substrates

3.6.1 Total solids

Total solids are the amount of solids present in the sample after the water present in it is evaporated at about 105°C in oven. For the determination of TS, a clean evaporating dish was first dried in (105^{0}C for 1 hour), cooled in desiccators and weighed immediately before use. Then, 10 g of freshly collected samples of each substrate was weighed using a digital balance, and placed onto a pre-dried and weighed evaporating dish. Then, the dish was put inside an oven at 105^{0}C using a crucible. The crucible was allowed to stay in the oven for 24 hours, and then was taken out, cooled in desiccators and weighed (APHA 2540 B, 1999). Then, the percentage of the TS was calculated as follows:

$$\% \text{ Total solid} = \frac{(A-B)}{(D-B)} \times 100$$

Where, A = weight of dish + dry sample (in grams)

B = weight of dish (in grams)

D = weight of wet sample and dish (in grams)

3.6.2 Volatile solids (VS)

The volatile solids content of the samples were determined by transferring the dried samples into muffle furnace, heating at 550ºC for 3 hours and weighing after cooling at room temperature. The percentage of volatile solids for all samples was calculated by using the following equation (APHA 2540 E, 1999).

$$\%VS = \frac{MDS-M(ash)}{MDS} \times 100$$

Where, %VS= volatile solid

MDS = mass of dried sample

M (ash) =mass of ash

3.6.3 Determination of pH

The pH values were determined using digital pH meter before and after anaerobic digestion. Before anaerobic digestion, the samples were diluted using distilled water before inoculation with rumen fluid and an electrode was inserted into samples of substrate to measure the pH values. However, pH measurements after anaerobic digestion were done using pH electrode which was inserted into samples of substrate that is digested at the end of the experiment.

3.6.4 Organic carbon

The carbon content of the substrates was obtained from volatile solids data using an empirical equation as reported by Badger *et al*.,(1979) and Barrington *et al*.,(2002).

$$\% \text{ carbon} = \frac{\%VS}{1.8}$$

Where, %VS volatile solid

3.7 Moisture content

The most commonly used method to determine moisture content is the oven-dying method. In this study moisture content was determined as the percentage of initial weight of the material lost through heating. To achieve this, 10g of each sample was dried separately in an oven at 105^{o}C for 24 hours and weighed. Moisture content was then calculated using the following formula (Elias, 2010).

$$\% \text{ MC} = \frac{W-D}{W} \times 100$$

Where, MC =moisture content

W =initial weight of sample (g)

D =weight of sample after drying at 105^{o}C for 24 hrs

3.8 Liquid content

The substrate of each biodigester was mixed with appropriate amount of distilled water and inoculums to achieve the recommended 8% total solids content. The total amount of liquid (distilled water and rumen fluid) that was added to the biodigester was determined by the following formula (Ituen *et al*., 2007).

$$8\% = \frac{(\text{MFTS})}{A + B}$$

Where, MFTS= mass of fixed total solid,

A= mass of fresh sample added

B= mass of water and inoculums to be added to get 8% TS suspension in the digester

Fixing amount of inoculum (100 ml) and distilled water were added in the digester to facilitate the rate of anaerobic digestion.

Table 3.2 The proportion of substrates added in the five digesters in three replicates (Tchobanoglous *et al*., 1993).

Digester	TS GM(g)	TS WP(g	%VS GM	%VS WP	Volume water added(ml	Volume Inoculums added (ml)	Fresh GM added (g)	Fresh WMP added (g)
A	23.5	0	60	0	168.8	100	25	0
B	16.45	6.3	46.7	20	159.4	100	17.5	7.5
C	11.75	10.5	33	33	153.12	100	12.5	12.5
D	7.05	14.7	21.9	51.1	146.9	100	7.5	17.5
E	0	21	0	77.6	137.5	100	0	25

A = (100% GM and 0 %WMP), B= (70%GM and 30%WMP), C= (50% GM and 50%WMP) D = (30% GM and 70%WMP) and E = (100%WMP and 0% GM).

3.9 Data Analysis

After the completion of the whole laboratory process the data were subjected to analysis of variance (one-way ANOVA) using SPSS version 20 to investigate statistical significance between the different digesters, whereas paired samples T-test was used to investigate statistical significance within a digester. Differences between means were considered statistically significant at $P<0.05$.

4 RESULTS AND DISCUSSION

4.1 The physico-chemical parameters of the substrates

4.1.1 The pH Value

Analysis of pH shows that the pH of 100% GM slurry before anaerobic digestion was about 6.94±0.03, whereas that of 100% WMP was 6.09±0.03. For anaerobic digestion, methanogenic bacteria prefer pH between 6.8 and 7.2. Thus, the pH value of 100%GM was optimum for biogas production, whereas that of 100%WMP was less optimal (Thy *et al*., 2003; Yadvika *et al*., 2004) (Table 4.1). From this result, it clearly stated that mixing the substrates resulted in the rise of pH compared to that of WP alone, but decreased pH from that of WMP alone. Before anaerobic digestion the pH was found to increase with increasing of GM proportion in the mix, suggesting that GM helps to maintain the pH to meet the optimum required. Therefore, mixing substrates is a good way of adjusting the pH value to the optimum rang (Hills and Roberts, 1981).

After AD, pH values of all treatments were found to increase when compared to before AD, though there was significant difference between treatments. This may due to the production of alkaline compounds such as ammonium ion. The highest pH value after AD was recorded in digester with 100% WMP (8.14). This may due to high accumulation of methanogenic bacteria and their waste products that may contribute to the rise of pH (Gerardi, 2003).

Table 4.1: Comparison of pH values between before and after anaerobic digestion of the substrates (values are mean ± SE, n=3)

Digester	Initial PH	Final PH
A	6.94 ± 0.03^{Ab}	7.41 ± 0.05^{Ea}
B	6.53 ± 0.04^{Cb}	7.75 ± 0.02^{Da}
C	6.78 ± 0.01^{Bb}	7.93 ± 0.02^{Ca}
D	6.23 ± 0.04^{Db}	8.06 ± 0.02^{Ba}
E	6.09 ± 0.03^{Eb}	8.14 ± 0.04^{Aa}

Means followed by different small letters in a row are significant different at $p<0.05$ for paired samples T-test within biodigester. Means followed by different capital letter in column are significantly different at $p<0.05$ between biodigester. ***GM= goat manure and WMP=watermelon peels.***

4.1.2 Total and Volatile Solid values before and After AD

There were statistically significant differences in TS and VS between treatments both in before and after AD (Table 4.2). The values of both TS and VS were significantly decreased after AD in all treatments (Table 5). Reductions of TS and VS may due to conversion of the substrates into biogas through AD (Rafique *et al*., 2010).

However, the maximum decrement of volatile solids was observed under the biodigester D that contain 30% GM + 70%WMP (73.0±0.12 to 56.8±0.09), suggesting more digestion of substrates by bacteria for either production of biogas or their own metabolic use. The maximum value of TS reduction was also recode from the biodigester D that contain 30% GM and 70% WMP 86.8±0.12 to 76.16±0.09, the reason may be highly digested by methanogenic bacteria and converted into biogas and other products.

In this studied the maximum decrement of TS and VS after digestion has been directly related to maximum biogas productions, i.e in biodigester D (30% GM and 70% WMP) maximum TS and VS was decreased as shown (table.4.2) and in this biodigester also the maximum biogas production was record. Thus total solids and volatile solids destruction is a good parameter for evaluating the efficiency of anaerobic digestion (Abubaker and Ismail, 2012) and it is a good indicator of biogas production (Anonymous, 1981)

Since, the minimum decrement of TS and VS after anaerobic digestion was record but the biogas yield was not much. It can be attributed to its relatively lower organic content or conversion of the substrates to cellular materials for growth and reproduction of bacteria (Gerardi, 2003 and Tamrat *et al.,* 2013).

Table 4.2: Comparison of % total solid and volatile solid between before and after anaerobic digestion of each digester (values are mean ± SE, n=3).

Biodigester	% Total Solid		Volatile Solid %	
	Initial	Final	Initial	Final
A	94.1±0.09^{Aa}	86.6±0.32^{Ab}	60.0±0.23^{Ea}	54.9±0.32^{Db}
B	91.3±0.2^{Ba}	82.0±0.27^{Cb}	66.7±0.13^{Ca}	55±0.11^{Cb}
C	88.8±0.12^{Ca}	78.6±0.21^{Db}	66.0±0.4^{Da}	54.3±0.13^{Eb}
D	86.8±0.12^{Da}	76.16±0.09^{Bb}	73.0±0.12^{Ba}	56.8±0.09^{Bb}
E	83.8±0.17^{Ea}	75.16±0.04^{Eb}	77.6±0.23^{Aa}	69.7±0.3^{Ab}

Means followed by different small letters in row are significantly reduced at p<0.05 for paired samples T-test within biodigester. Means followed by different capital letter in column are significantly different at p<0.05 between biodigester. ***A contains 100%GM, B contains 70%GM and 30%WMP, C contains 50%GM and 50%WMP, D contains 30%GM and 70%WMP, and E contains 100% WMP. GM= goat manure and WMP=watermelon peels.***

4.1.3 Organic Carbon

The carbon content of the feed stock was obtained from volatile solids data (% carbon) as reported by Haug (1993). According to the experimental result, the percent of organic carbon was significantly reduced after anaerobic digestion in all biodigester. This indicates that organic carbon might have been consumed by bacteria to support their metabolic activities or converted in to biogas (Gerardi, 2003; Devlin *et al*, 2011).

There was significant difference in % organic carbon between treatments both before and after AD (Table 4.3). The highest organic carbon was recorded from 100%WMP alone, suggesting the high content of biodegradable materials. Percent organic carbon was significantly reduced in all treatments after AD. This may show that there was degradation of substrates in all treatments (Abdel-Hadi and El-Azeem, 2008).

However, the highest carbon reduction was record in a mix ratio of 30% GM+70% WMP (40.5±0.06 to 31.6±0.05). The greater organic carbon decrement after AD implies the more degradation process was takes place in the biodigester. Organic carbon can be removed during

AD either by being converted to cellular materials for growth and reproduction of bacteria or biogas production (Gerardi, 2003).

Table 4.3: Comparison of % organic carbon before and after anaerobic digestion of the various substrates (values are mean ± SE, n=3)

Digester	Initial carbon	Final carbon
A	33.3 ± 0.13^{Ea}	30.5 ± 0.17^{Db}
B	37 ± 0.07^{Ca}	30.6 ± 0.08^{Cb}
C	36.6 ± 0.22^{Da}	30.2 ± 0.07^{Eb}
D	40.5 ± 0.06^{Ba}	31.6 ± 0.05^{Bb}
E	43.0 ± 0.07^{Aa}	38.7 ± 0.15^{Ab}

Means followed by different small letters in row are significant reduced at $p<0.05$ for paired samples t-test within biodigester. Means followed by different capital letter in column are significantly different at $p<0.05$ between biodigester. ***A contains 100%GM, B contains 70%GM and 30%WMP, C contains 50%GM and 50%WMP, D contains 30%GM and 70%WMP, and E contains 100% WMP. GM= goat manure and WMP=watermelon peels.***

4.2 Average Daily and Cumulative Biogas Productions

Biogas production from co-digestion of goat manure and watermelon waste peels under mesophilic (38°C) condition was measured through the experimental periods by using water displacement method from each biodigester until there was stops gas productions any more. In the first two days of the experimental observation, there was no any biogas production recorded. However, in between 4th and 11th days of the experimental observation biogas production was found to increase in all treatments (Fig 4.1).

The biogas production was peaked on the 8^{th} day of experimental period in biodigester A with 54.2 ml of gas measured. This biodigester was containing only 100% GM which may be rich in the availability of readily biodegradable organic matter in the substrate. This might be also due to partial fermentation takes place in digestive tract of the ruminant animal (Deubein and Steinhauser, 2008).

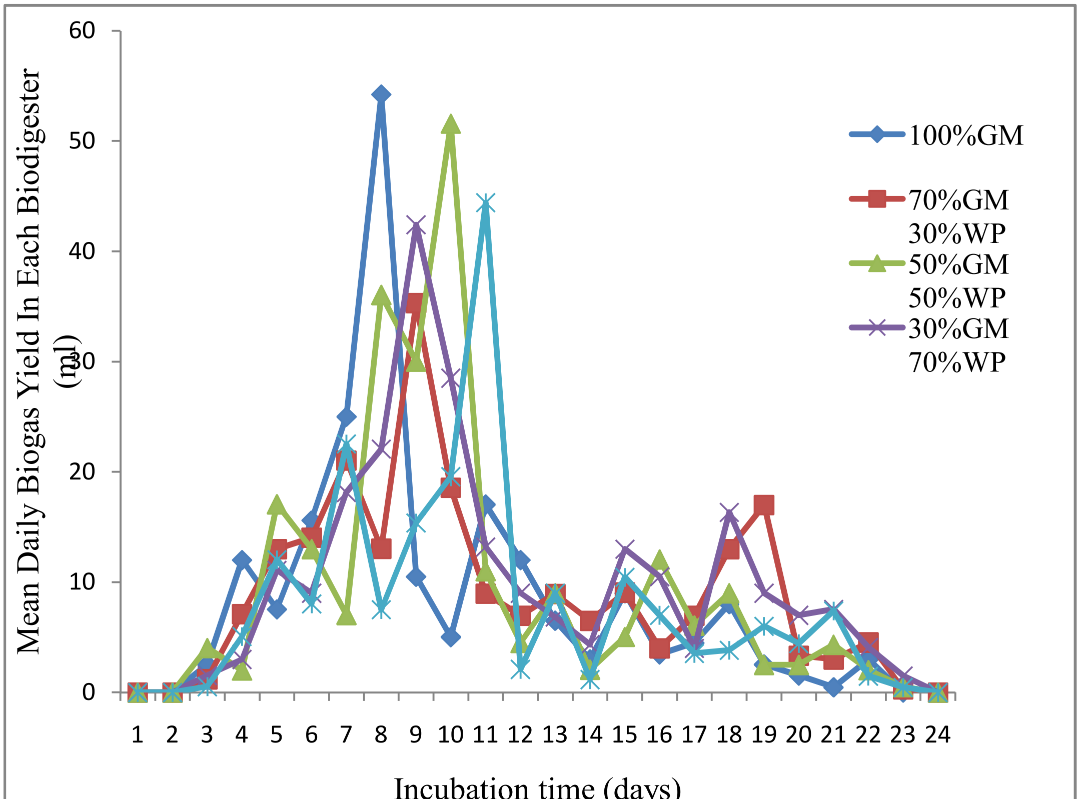

Figure 4.1 Daily biogas production of each biodigester

After 13th day of fermentation, biogas production found to decrease and eventually reached 0 ml on the 24th day of fermentation in all digesters. This decline in biogas production could be due to the increased bacterial population and the depletion of readily decomposable substrate (Ahn *et al*., 2009). It may also be due to the increased pH value after AD which may result from the buildup of ammonium ion (Hansen *et al*., 1998). For optimal performance of the microbes, the pH value within the biodigester should be kept in the range of 6.8 - 7.2. The pH value below or above this interval may restrain the process in the reactor since micro-organisms and their enzymes are sensitive to pH deviation (Yadvika *et al*., 2004).

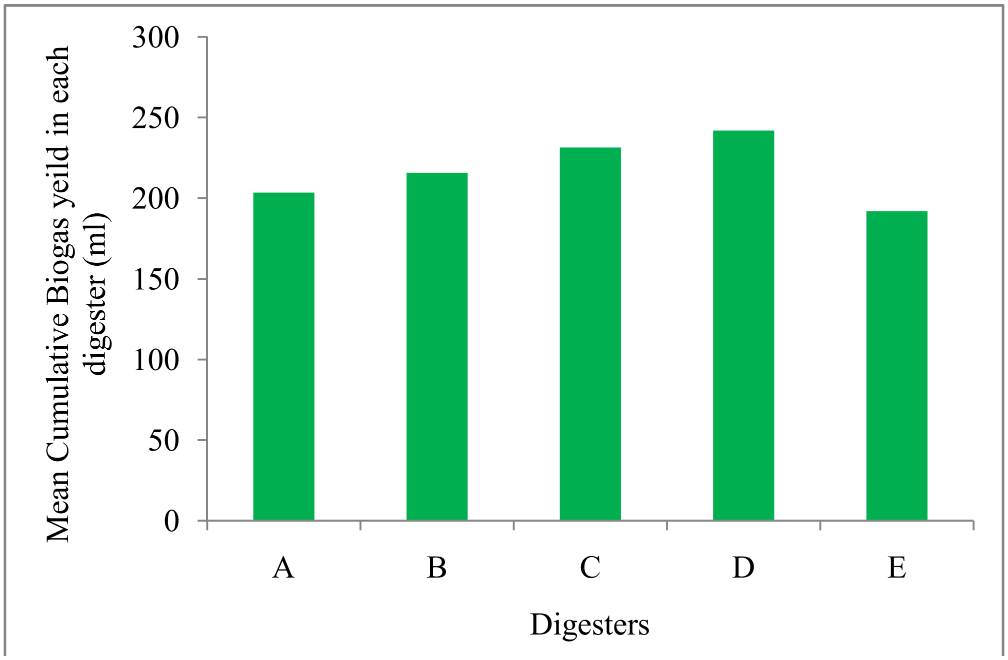

Figure 4.2 The mean cumulative biogas productions in each digester over 30 days

Figure 4.2 indicates that, the high cumulative biogas production was recorded from biodigester D (242.0 ml) which contained substrate mix of 30%GM and 70%WMP. This mix ratio may have a proper nutrient balance that facilitates the growth of bacteria for biogas production (Macias-Corral *et al*., 2008; Lema, 2013). Previous studies have revealed that the digestion of more than one kind of substrate could establish positive synergism in the digester (Danqi, 2010; Jianzheng *et al*., 2011).

The minimum (191.86 ml) cumulative biogas production was recorded from 100%WP substrate. This may be due to slightly acidic nature of the substrate. Anonymous (1992) reported that microbes require neutral or mild alkaline conditions for optimal biogas production. The cumulative biogas production for 100% GM, 70% GM+30% WMP, 50% GM+50% WMP, 30% GM+70% WMP and 100% WMP were 203.4, 215.8, 231.3, 242.0 and 191.86 ml respectively. As the proportion of goat manure in the mix ratio decrease from 70% to 30%; however, the cumulative biogas yield was increased. Thus, it can be concluded that co-digestion of GM and WMP was more productive with GM proportion not exceeding more than 30% in digester.

5 Summary, Conclusion and Recommendation

5.1 Summary and Conclusion

Biogas production through anaerobic digestion of organic waste materials under optimum pH, temperature and moisture content provides an alternative energy source. Currently, the world energy consumption has increased vigorously from time to time over the last century as a result of world population increase. To alleviate this problem the developing country like Ethiopia could develop biogas technology, because it is a modern and eco-friendly technology that is based on the decomposition of organic materials in anaerobic environment. Different fruit peels are biodegradable by nature and are potential source of the biogas. So the objective of this study was production of biogas through co-digestion of watermelon fruit peel and goat manure in five mixe ratios under anaerobic environment and mesophilic conditions (38ºC) using batch mode fermentation.

In all biodigester, total solid, volatile solid, organic carbon, and pH were analyzed before and after anaerobic digestion. The daily biogas production was measured by using water displacement method for 27 days. Assessment of cumulative biogas production revealed that the substrate mix ratio of 30% GM and 70% WMP was superior in gas production to other mixed substrates in the biodigester. Therefore, the mix ratio of 30% GM and 70% WMP of substrates is an optimal mix ratio to yield 242.0ml cumulative biogas product.

5.2 Recommendations

Based on the finding of this study, scope for future research studies and development activities need to consider the following recommendations:

- This investigation was done only at mesophilic temperature of 38°C, but further similar studies should also be carried out at 40°C or above temperature conditions to increase the rate of digestion.
- Since this experiment was carried out on the co-digestion of goat manure with watermelon peel waste, further similar studies should also be made on the co-digestion of watermelon peel with other substrates to enhance biogas yield.

- ❖ Further study is again necessary to define the composition of organic matter like carbohydrates, proteins, lipids and cellulose to add the appropriate enzymes.
- ❖ Further study is again necessary to check the presence of microbial groups in the inoculums added in the digester.

6 REFERENCES

Abdel-Hadi, M.A. and Abd El-Azeem, S.A.M. 2008.Effect of heating, mixing and digester type on biogas production from buffalo dung. *Misr J. Agric. Eng.* 25(4): 1454-1477

Abuabaker, B. and N. Ismail, 2012.Anaerobic Digestion of Cow Dung for Biogas Production. *ARPN J. Eng. Appl. Sci.* 7:169-172.

Ahn, H., Smith, M., Kondrad, S. and White, J. 2009.Evaluation of biogas production potential by dry anaerobic digestion of switch grass-animal manure mixtures. *Appl. Biochem. Biotechnol*. 160:965–975.

Ahring, B. K., Z. Mladenovska, R.Iranpour, P.Westerman.,2001. State of the art and future perspectives of thermophilic anaerobic digestion, in: Proceedings of the 9thWorld Congress on Anaerobic Digestion, vol. 1, Antwerpen, Belgium, pp. 455–460.

American Public Health Association 1999.Standard Method for the Examination of Water and Wastewater 20th Edition. Washington: APHA.

Anonymous(1981). Biogas technology and utilization. A status Report. Department of Science and Technology. *Journal of coffee research*. 19, 17-18

Anonymous. 1992. *Biogas Technology in China*. Chengdu Biogas Research Institute of the Ministry of Agriculture.

Anunputtikul, W. and Rodtong, S. 2004. Laboratory Scale Experiments for Biogas Production from Cassava Tubers. *The Joint International Conference* on *Sustainable Energy and Environment (SEE)*, Hua Hin, Thailand

Aremu, M .O. and Agarry, S. E. 2013. Enhanced Biogas Production From Poultry Droppings Using Corn-Cob And Waste Paper As Co-Substrate. *International Journal of Engineering Science and Technology (IJEST*), 5(02): 247- 253.

Arogo, J.O, Wen, Z., Ignosh, J., Bendfeldt, E. and Collins, E.R. 2009. *Biomethane Technology.* College of Agriculture and Life Sciences, Virginia Polytechnic Institute and State University, publication 442-881.

Asnake Mekuria.2006. The role of land use on impact of drought Shabele, Berenta woreda, Amhara National *http://cgspace.cg.ar. Org/ bit stream/handle. Available on line Retrieved in September 8, 2013.*

Atkins, M., Fuchs, M., Hoffman, A. and Wilhelm N. 2008. Plastic Tubular Biogas Digesters: A Pilot Project in Uru, Kilimanjaro, Tanzania. ttp://www.watsoninstitute.org/ge/watson Accessed on, June 12, 2014

Ayoub Mohamed Eshraideh. 2002. Biogas purification, compression and bottling. *Build a Biogas Plant*. Published by Global institute for research and education G.J. E.D.T., Vol.2 (6):34-38 (November-December, 2013) ISSN: 2319 – 7293

Badger, C.M., Bogue, M.J. and Stewart, D.J. 1979. Biogas production from crops and organic wastes. *New Zealand Journal of Science*, 22: 11-20.

Barrington, Choiniere, D., Trigui, M., and Knight, W. 2002. Effect of carbon source on compost nitrogen and carbon losses. *Bioresource Technology*, 83:189-194.

Bie. 2002. Increase of biogas production at KÄPPALA WWTP. Disintegration methods and laboratory scale biogas measurement. *Industrial Ecology, Royal Institute of Technology*. http://www.win2pdf.com. Accessed on, November 16, 2014

Bouallagui, H., Ben Cheikh, R., Marouani, L. Hamdi, M., 2003.Mesophilic biogas production from fruit and vegetable waste in a tubular digester. *Bio-resource technology,* 86:85–89.

Buswell, A.M., Sollo, F.W. 1948.The mechanism of Methane Fermentation. *American Chemical Society* 70: 1778-1780.

Chatterjee, A.K., 2007.Introduction *to Biotechnology*, second edition (pp.124-144).

Ciborowski, P. 2004. Anaerobic Digestion in the Dairy Industry, Minnesota Pollution Control Agency Air Innovations Conference, www.epa.gov. *retrieved* 19.08.07

Claude, J. 2009. Making a Link between Biogas and Organic Agriculture in Africa. Kampala, Rwanda. http://www.unep-unctad.org/cbtf/organicagriculture.asp Accessed on, August 6, 2014.

Claudia, B and Yitayal Addis. 2011, survey of biogas plant in four regional state of Ethiopia *SWV Ethiopiahttp:// etd.aau.edu. et/ dspace/bitstream/123456789/3390/1/.*

Dagnachew Legesse, Vallet-Coulomb, C. and Gasse, F. 2003. Hydrological response of a catchment and land use change in tropical Africa: Case study south central Ethiopia. *Journal of Hydrology*275: 67-85.

Dane and Liu, 2007. Diversity and origin of cultivated and citron type watermelon (Citrullus lanatus). *Genetic Resources and Crop Evolution* 54: 327-336.

Danqi, Z 2010. Co-digestion of Different Wastes for Enhanced Methane Production. Msc thesis, The Ohio State University, Ohio, USA.

Dawit Diriba, 2011. Energy demand in rural Ethiopia from household perspective. *The journal of energy and development,* 2:1, 2012.

Demetriades,P. 2008.Thermal *pre-treatment of cellulose rich biomass for biogas production*.

Deublein, D., and A. Steinhauser, 2008.*Biogas from waste and renewable source: An introduction*. Wiley-vch.

Devlin, D., Esteves, C.S.R.R., Dinsdale, R.M. and Guwy, A.J. 2011. The effect of acid pretreatment on the anaerobic digestion and dewatering of waste activated sludge. *Bioresource Technology*. 102: 4076-4082.

Elias Jigra 2010. Renewable biogas energy production from Cladodes of *Opuntia ficus-indicia, Msc thesis Addis Ababa university,* pp26-28, 34*,* unpublished .

Elijah, I. M., Ibifuro and M.Yahaya, 2009. The study of cow dung as co-substrate with rice husk in biogas production, *Scientific Research and Essays,* 4 (9): 861–866. ISSN 1992- 2248.

El-Mashad, H.M., G. Zeeman, K.P. Wilko, P.A. Gerard and G. Lettinga. 2004. Effect of temperature and temperature fluctuation on thermophilic anaerobic digestion of cattle manure. *Bioresour.Technol.* 95:191–201.

Eshete, d. G., sonder, d. K. & heegde, m. F. T. 2006 Report on the feasibility study of a national program for domestic biogas in Ethiopia; *SNV Netherlands Development Organization*: Addis Ababa,Ethiopia,

Esposito, G., Frunzo, L., Liotta, F., Panico, A. and Pirozzi, F. 2012. Bio-Methane Potential Tests to Measure the Biogas Production from the Digestion and Co-Digestion of Complex Organic Substrates. *Journal of Open Environmental Engineering*, 5: 1-8.

FAO.1990. Food and Agricultural Organization of the United Nations Agrometeorology group, Remote Sensing Center *Research and Technology Division*. Rome, Italy.

Feng, L., and L. Rundong., 2010.Anaerobic Co-digestion of Cow-manure and source separation BMW. *Shenyang Institute of Aeronautical Engineering, Liaoning High College New Century Excellent Research Project*.

Forster-Carneiro, T., Pe´rez, M., Romero, L.I. and Sales, D. 2007. Dry-thermophilic anaerobic digestion of organic fraction of the municipal solid waste: Focusing on the inoculums sources. *Bioresource Technology* 98:3195–3203.

Forum for Environment. 2010. Ethiopian Environment Review, No. 1. Addis Ababa, Ethiopia.

Fulford, D.D. 1998. Running a biogas program, a handbook. *Intermediate technology Publications,* London. 123pp.

Fricke, K., Santen, H. and Wallmann, R. 2005. *Comparison of selected aerobic and anaerobic procedures for MSW treatment*. Waste Management Online, Retrieved: July 29, 2005*, http:// www.sciencediret.com.*

Gareth, M., E. and Judith, F. 2003. *Environmental Biotechnology Theory and Application*. John Wiley & Sons Ltd, the Atrium, Southern Gate, Chichester, West Sussex PO19 8SQ, England.

Gerardi, M.H, 2003.*The microbiology of anaerobic digesters*. A John Wiley and Sons. 11:99-103

Getachew Esthete and K. S. F. H. 2006. Report On The Feasibility Study Of A National Programme For Domestic Biogas In Ethiopia, *SNV Ethiopia*

Goldemberg J, Johansson TB 2004. World energy assessment overview 2004 update. New York: UNDP 88.

Gollakota, K.G.and Meher, K.K. 1988. Effect of particle size, temperature, loading rate and stirring on biogas production from castor cake. *Biol. Wastes* 24, 243–249.

Gomec, Y.C., Speece, R.E., 2003. The role of pH in the organic material solubilasation of domestic sludge in anaerobic digestion. Water Sci. technol. 48:143-150.

Hansen, K., I. Angelidaki and B.K. Ahring, 1998. Anaerobic digestion of swine manure: inhibition by ammonia. *Water Res.* 32:5-2.

Hassan, E.A. 2003. Production from forage and sugar beets process control and optimization-Ecology and Economy. *Agrotech*.19:70-72

Haug, R. T, *The practical handbook of compost engineering*, Lewis Publishers, Florida, USA, 1993.

Hartmann, H. and Ahring B.K. 2006. Strategies for the anaerobic digestion of the organic fraction of municipal solid waste: an overview. *Water Sci. Technol*. 53 (8):7-22.

Hendriks, A.T.W.M. and Zeeman, G. 2009. Pretreatments to enhance the digestibility of lignocelluloses biomass. *Bioresource Technology*, 100(1): 10-18.

Hills, D. J. and D.W. Roberts, 1981.Anaerobic digestion of dairy manure and field crop residues. *Agric. Wastes* 3:179–189

Itodo, I.N., and Philips, T.K 2001"determination of suitable material for anaerobic biogas digester". In*: proceeding of the second international conference and* 23 *annual General meeting Vol.* (23) *institution of Agricultural Engineering, pp*.437-441.

Itodo, I.N, E.B. Lucas and E.I. Kucha, 1992.The Effect of Media Materials and its Quality on Biogas Yield.*Niger. J. Renew energ.* 3:45-49.

Ituen EE., John N.M, Bassey B.E 2007. Biogas production from organic wastes in Akwabom state of Nigeria, Appropriate technology of environmental protection in the developing world. Selected paper from ERTEP 2007, July 17-19,

Jan,L, 2010. Domestic biogas compact course. Technology and mass-dissemination experiences from Asia: post graduate program renewable Energy. *http://www. Uni-oldenburgide/ fileadmin/user. Accessed on*, July 22, 2014

Jianzheng L, Ajay K, Junguo H, Qiaoying B, Sheng C, Peng W (2011). Assessment of the effects of dry anaerobic co-digestion of cow dung with waste water sludge on biogas yield and biodegradability. *Int. J. Phys. Sci*. 6(15):3679-3688.

Kalia VC, Sonakya V, Raizada 2000. Anaerobic digestion of banana stems waste. *Bioresour. Technol.* 73: 191-193.

Khalid, A., Arshad, M., Anjum, M., Mahmood, T. and Dawson, L. 2011. The anaerobic digestion of solid organic waste. *Journal of Waste Management*, 31: 1737–1744

Kim, M. & R.E. Speece., 2002. Reactor configuration-Part II comparative process stability and efficiency of thermophilic anaerobic digestion. *EnvironmentalTechnology*, 23:643-654.

Kim J. park C, Kim T. M, kim S, S.W and Lee J. 2003 effects of various pre-treatments for enhanced anaerobic digestion with waste activated sludge*. J. Bio sci. Bioeng. 95:271-275.*

Knottier, M., 2003.*Integration of biogas technology, organic farming and energy crops.* The future of biogas in Europe II, European biogas workshop. University of Southern Denmark, Denmark.

Lemma Abayneh.2013.Biogas production from co-digestion of wet coffee processing Waste (pulp) and cow dung. An MSc Thesis Presented to the School of Graduate Studies of Haramaya University. pp.46.

Li, Z., Tang, R., Xia, C., Luo, H. and Zhong, H. 2005. Towards green rural energy in Yunnan, China. *Renewable Energy* 30: 99 - 108.

Li. R. S.Chen and X. Li, 2009.Anaerobic co-digestion of kitchen waste and cattle manure of methane production. *Energy Sources*. 31: 1848-1856

Macias-Corral, M., Z. Samani, A. Hanson, G. Smith, P. Funk, H. Yu and J. Longworth, 2008. Anaerobic digestion of municipal solid waste and agricultural waste and the effect of co-digestion with dairy cow dung. *Bioresour. Technol.* 99:8288–8293

Madu, C. and Sodeinde, O.A. 2001. Relevance of biomass in the sustainable energy development in Nigeria. In proc. *National Engineering Conference and Annual General Meeting of the Nigerian Society of Engineers*, 220-227.

Mahanta P, Dewan A., Saha, UK., Kalita, P .2004. Influence of Temperature and total solid concentration on the gas production rate of biogas digester. *Journal of Energy Southern Africa*, 15(4):112-117.

Marchaim, U. 1992. *Biogas processes for sustainable development*. FAO Agricultural services Bulletin 95.

Maria, M.M., 2008. Renewable Energy for Development, Special Theme Issue*: Partnerships for Bioenergy in Africa.Stockhholm Environmental Institute.*

Mata-Alvarez, Tilche, A. and Cecchi, F. 2000. *Proceedings of the Second International Symposium on Anaerobic Digestion of Solid Wastes*, Barcelona.1:375-380. methane production. *Energy sources:* 31: 1848-1856.

Molina, F., G Ruiz Filippi, C. Garcia, E. Roca, and J. Lemma. 2007. “Winery effluent treatment at an anaerobic hybrid USBF pilot plant under normal and abnormal operation*, Water Science and Technology.*56. 25-31.

Navia R, Soto M, Vidal G, Bornhard C, and Diez M.C: 2002 alkaline pre-treatment of kraft mill sludge to improve its anaerobic digestion. Bull. Environ .Contam. To xicol. 69: 869-876

NBP, 2008. Biogas for Better Life .Ethiopia, Addis Ababa, Ethiopia Rural Energy Development and Promotion Centre (EREDPC) and SNV/Ethiopia.

NBPE. 2007. *Biogas for Better Life*: Brief Program Profile. Addis Ababa Ethiopia

Nailia, I. K., Roustem, E. K., Rimma, P. N. and Mark, A. N. 1979. The Influence of Ammonium and Methods for Removal during the Anaerobic Treatment of Poultry litter. *J. Chem. T ech. Biotechnol* 70: 99-105.

Ofoefule, Akuzuo, U., Nwankwo, Joseph I., Ibeto and Cynthia, N. 2010. Biogas Production from Paper Waste and its blend with Cow dung. *Advances in Applied Science Research*, 1 (2):1-8.

Ostrem, K.M., K. Millrath, N.J Themelis, 2004. *Combining anaerobic digestion and waste-toenergy*.12th North America Waste to Energy Conference.

Parawira,W, Read J>S; mottiasson< B. and Bjornson. L; energy production from agricultural residues: high methane yields in pilot scale two stage anaerobic digestion. *Bio mass and energy*.2008.32 (1): pp 44-50

Prasad, K., Inmaculada, B., Lars, E. and Irini, A. 2008. Effects of mixing on methane production during thermophilic anaerobic digestion of manure: Lab-scale and pilot-scale studies. *Bioresour. Technol* 99:4919–4928.

Rafique, R., T.P. Gorm, A. Nizami, Z. Asam, D.M. Jerry and G. Kiely, 2010. Effect of thermal, chemical and thermo-chemical pre-treatments to enhance methane production. *Energy*. 35:4556-4561

Rilling, N. 2005.Anaerobic Fermentation of Wet and Semidry Garbage Waste Fractions.*pp*. 355-373*In*: Jördening,H and Winter, J.(eds). *Environmental Biotechnology. Concepts and Applications.* WILEY-VCH Verlag GmbH & Co. KGaA, Weinheim. ISBN: 3-527-30585

Rittmann BE (2008). Opportunities for Renewable Bioenergy Using Microorganisms. *Biotechnol. Bioeng*. 100:203-212.

Rojas, C., Fang, S., Uhlenhut, F., Borchert, A., Stein, I. and Schlaak, M. 2010. Stirring and biomass starter influences the anaerobic digestion of different substrates for biogas production. *Eng. Life Sci*. 10(4): 339–347.

Sadaka, S.S. and Dan Engler, C.R. 2003. Effect of initial total solids on composting of raw manure with biogas recovery. *Journal of Compost Science and Utilization*, 11:361-369.

Sandars, D.L., H.W. Doelle, and Juniper, 2003. Environmental benefits of livestock manure management. Practice and technology by life cycles Assessment.*Biosys. Eng* .84: 267- 281.

Schamphelaire W, Verstraete G 2009. Biological Energy Conversion, Biotechnol. Bioeng. 103(2):1.Science and Technology, Government of India, New Delhi, 20-46.

Shakya. I and N. Charushree, 2009. Benefits and Impacts of Biogas Technology. Training on Biogas for Participants from HoAREC, Ethiopia July 6: BSP Nepal.

Shrestha, A. 2010. Prospects of Biogas in Terms of Socio-Economic and Environmental Benefits to Rural Community of Nepal: A Case of Biogas Project in Gaikhur VDC of Gorkha District. Master Thesis. Tribhuvan University, Kathmandu, Nepal.

Siltan, A. 1989.The development of biogas in Ethiopia, *Ministry of Mines & Energy* Addis Ababa Ethiopia.

Steinmetz, R., Perazzoli, S., Mezzari, P., Nunes, E. and Silva, M. 2013. Biogas production From Microalgae Biomass. *American Journal of Agricultural and Biological Sciences*, 7 (4): 494-502.

Sunarso, S., Johari, Z., Widiasa, I. N. and Budiyono. 2012. The Effect of Feed to Inoculums Ratio on Biogas Production Rate from Cattle Manure Using Rumen Fluid as Inoculums. *Internet J. of Waste Resources* 2(1):1-4.

Tamrat Aragaw, Mebeaselassie Andargie and Amare Gessesse. 2013, from the co-digestion of cattle manure with organic kitchen waste to increase biogas production using rumen fluid as inoculums. *Int.J. of Physical science.* 8(11): 444-450.

Tamrat Asnake. 2008. Potential of floriculture residue for Biogas production. MSc. Thesis, Addis Ababa University.

Tchobanoglous, G., H. Theisen and S.Vigil, 1993.*Integrated Solid Waste Management Engineering: Principle and Management Issues.*Singapore, McGraw-Hill U.S.

Teame Gebrekidan, Meseret C. Egigu and Manikandan Muthuswamy, 2014 Efficiency of biogas production from cactus fruit peel co-digestion with cow dung. *International Journal of Advanced Research 2(7): 916-923*

Thy, S., Preston, T.R. and Ly, J. 2003.Effect of retention time on gas production and fertilizer value of biodigester effluent.*Livest Res Rural Dev.* 15(7):1-24

Vavilin.V.A, Fernandez, Palatsi,J, and Flotats,x; Hydrolysis kinetics in an aerobic degradation of particulate organic materials: *An over view. Waste management* 2008. 28(6):*pp* 939-951.

Werner Kossmann, U.P., 2000. Biogas Digest-Volume-I.Eschborn: Information and Advisory Service on Appropriate Technology (ISAT).

Wilkie, A.C. and J.Well. 2008. Bioenergy: *Biomethane from biomass, Biowaste and Biofuel* Washington DC.195-199.

Yadvika, Santosh, Sreekrishnan, T.R., Kohli, S. and Rana. 2004. Enhancement of biogas production from solid substrates using different technique: *Bioresource Technology* 95:1–10.

Yetilmezsoy, K. and Sakar S. 2008. Development of empirical models for performance evaluation of UASB reactors treating poultry manure wastewater under different operational conditions, *J. Hazardous Materials*, 153: 532–543 .

Zaher, U., Cheong, D., Wu, B. and Chen, S. 2007. *Producing Energy and Fertilizer from Organic Municipal Solid Waste.* Washington State University. Washington.

Zamudio Canas and Esteban Manuel. 2010. Technical feasibility of anaerobic co-digestion of Diary Manure with chicken Litter and other wastes Master's Thesis University of Tennessee.

Zorg,. 2009 Ukraine Biogas Production Plants. http://Biogas%20Production%20process _%20Four%20steps%20of%20fermentation. Ht .Accessed on23/11/2010.

7 Appendix

Appendix table 1.Daily mean biogas yield (ml) from co-digestion of goat manure and watermelon waste peel with their mixtures at different mixed ratio ± SE (ml) (n=3)

100% GM and 0% WP	70% GM and 30% WP	50% GM and 50% WP	30%GM and 70% WP	0% GM and 100% WP
0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00
2.53±0.23	1.2±0.29	4.03±0.33	1.53±0.06	0.53±0.11
12.0±0.33	7.1±0.36	2.0±0.28	3.0±0.75	5.06±0.37
7.53±0.28	13.0±0.57	17.06±1.32	11.06±0.87	12.03±1.29
15.5±2.11	14.03±0.58	13.0±1.51	9.0±0.42	8.03±0.56
25.0±1.01	21.03±1.51	7.03±0.63	18.13±1.61	22.53±1.59
54.2±0.39	13.3±0.36	36.03±0.88	22.06±0.66	7.5±0.36
10.6±2.44	35.3±3.46	30.0±4.35	42.4±2.30	15.4±1.52
5.03±1.33	18.56±1.89	51.56±3.35	28.5±1.73	19.6±0.58
17.03±0.50	8.96±0.75	11±0.00	13.2±1.43	44.4±6.42
12.0±4.5	6.96±3.58	4.5±0.86	9.0±2.88	2.1±1.71
6.5±1.89	8.93±1.55	9.0±3.51	6.83±0.16	9.0±0.57
3.0±0.00	6.5±0.86	2.06±0.99	4.33±1.16	1.16±0.44
9.0±4.58	9.06±1.48	5.03±2.16	13.0±3.78	10.43±3.67
3.46±1.00	4.0±1.73	12.0.6±2.03	10.5±0.28	7.0±2.30
4.5±2.02	6.96±2.1	6.06±1.68	4.0±0.76	3.53±1.47
8.0±1.73	13.0±2.08	9.0±2.64	16.3±2.33	1.66±1.20
2.5±1.63	17.0±2.08	2.5±2.02	9±2.08	6.0±1.73
1.5±1.24	3.33±2.84	2.5±1.32	7.0±1.52	4.43±1.10
0.5±0.46	3.0±1.52	4.33±3.21	7.53±2.72	7.4±3.34

3.0±0.33	4.53±0.29	2.03±0.49	4.06±0.29	1.46±0.31
0.0±0.00	0.3±0.17	0.5±0.24	1.5±0.28	0.5±0.29
0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00	0.0±0.00

Appendix table 2. Total Biogas production in the biodigester

Digester	Mixing ratio	Total biogas (ml)
A	100%GM and 0% WP	203.4
B	70% GM and 30% WP	215.8
C	50% GM and 50% WP	231.3
D	30% GM and 70% WP	242.0
E	0% GM and 100% WP	191.86

Appendix table 3: The values of physico-chemical parameters in three replications

		PH		% C.C		% T.S		% V.S	
Rep	Biod	I	F	I	F	I	F	I	F
1	A	6.99	7.3	33.55	30.8	95	83.5	60.4	55.5
1	B	6.6	7.73	37.11	30.66	93	83	66.8	55.2
1	C	6.81	7.97	36.88	30.22	90	76.5	66.4	54.4
1	D	6.18	8.06	40.44	31.55	86.5	76.5	72.8	56.8
1	E	6.03	8.1	43.11	38.44	82	68.5	77.6	69.2
2	A	6.97	7.48	33.33	30.51	93.5	89	60	54.92
2	B	6.45	7.8	36.88	30.48	89.5	79.5	66.4	54.88
2	C	6.76	7.95	36.88	30.22	88	79.5	66.4	54.4
2	D	6.2	8.03	40.66	31.48	88	76.5	73.2	56.68
2	E	6.15	8.23	43.33	38.88	85	79	78	70
3	A	6.87	7.45	33.11	30.22	94	87.5	59.6	54.4
3	B	6.55	7.74	37.11	30.46	91.5	83.5	66.8	54.84
3	C	6.79	7.88	36.22	30	88.5	80	65.2	54
3	D	6.33	8.1	40.51	31.66	86	75.5	72.92	57
3	E	6.1	8.14	42.88	38.88	84.5	78	77.2	70