

COMPARISON OF METHANE PRODUCTION FROM HOUSEHOLD WASTE
USING POND SOIL AND HORSE DUNG AS MICROBIAL INOCULA

by

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Abstract

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Anaerobic digestion is a biochemical process in which various microorganisms transform complex organic compounds to methane, along with carbon dioxide, water vapor and a small amount of other trace gases in absence of oxygen. Methane from anaerobic digestion processes is being increasingly utilized as an alternative energy source in developing as well as developed countries. These days the energy crisis is very acute in rural communities of developing countries, where fuelwood has been and still is the major source of daily used fuel. Through anaerobic digestion, clean-burning fuel for cooking can be produced from degradation of household organic wastes, solving two global environmental problems at once, waste management and renewable energy.

However for the successful operation of a digester, selection of inoculating microbes is crucial. If an active inoculum is selected, failure of overall the process can be avoided during the initial/start-up phase. Previous studies have found sewage sludge to be an effective inocula. However, as mentioned, one of the most important applications of small-scale anaerobic digesters is in the rural areas of low income countries. Using sewage sludge as a seed for an anaerobic digester is not an option in these cases since most developing countries do not have wastewater treatment facilities in the rural areas. Pond bottom soil and dungs from various animals can be easily available in the rural areas of low income countries.

Methanogens are common in nature at the bottom of stagnated ponds, lakes, and swamps or wetlands, where they produce methane, which is also known as swamp gas or marsh gas, in anoxic

conditions, as well as in digestive tracts of animals. Several studies have been done to examine the animals dung as an inocula but pond bottom soil has not been examined in previous digester studies.

The main objectives of the current study were to compare the rate of methane production and cumulative methane generation from degradation of a mixture of household organic wastes (food, paper, and yard) using 2 types of pond soil and horse dung as an inocula, and to determine the effectiveness of the best inoculum from the first experiment in degrading mixtures with various proportions of food, paper and yard waste. To accomplish this, three kinds of inocula - Pond bottom soil 1, Pond bottom soil 2 and Horse dung - were obtained from two different ponds and outdoor loafing sheds near Keller, Texas. Waste samples, including Food, Paper and Yard waste, were collected from a house. Three laboratory scale reactors were prepared with selected household waste and inoculum and operated at room temperature. Duplicate runs were conducted. The pH level of reactors was controlled by adding sodium hydroxide.

For the duplicate sets of reactors in experiments, reactors inoculated with Pond1 started gas formation sooner, and peaked earlier than the Pond 2 and Horse dung reactors. For both sets of reactors, the reactor seeded with pond bottom soil obtained from Pond1 had the highest methane generation rate, as well as the largest cumulative volume of methane, followed by Pond 2 and finally the reactor seeded with Horse dung. The possible reason is that Pond 1 is an older pond compared to Pond 2, and the bottom soil obtained from the Pond1 may have more stable organic matter accumulated as well as higher nutrient concentrations, leading to higher numbers and variety of microbes. The cumulative average methane generation in 95 days, for the reactor seeded with Pond 1 was 19.4 % higher than reactor seeded with Pond 2 and 72% higher than reactor seeded with Horse dung. Similarly, reactor Pond 2 generated 44% more methane than the reactor with the Horse dung. Pond soils obtained from the Pond 1 and Pond 2 gave better performance than horse dung as an inoculum for treating household wastes.

Another experiment was the further testing of the effectiveness of best inoculum from the first experiments in degrading mixtures with various proportions of food, paper and yard waste. All reactors were inoculated with digested waste of Pond1 (B).The reactor with more Food started gas formation sooner than Paper and Yard. It is likely because of food waste decomposes faster. For all three reactors (Food, Paper

and Yard) through the 68 days of reactor operation, Food waste had the highest rate of methane generation as well as largest cumulative volume of methane, likely because food waste has the highest amount of cellulose, and more surface areas per unit volume of waste for the microbes to access, than paper and yard waste.

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Chapter 1

Introduction

1.1 Background

The sustainable management of municipal solid waste (MSW) and the need for renewable energy are two of the most pressing environmental problems faced by the world today. Solid waste generation has increased rapidly due to increases in the worldwide population levels over the last five decades (Singh et al., 2011). Other factors which have increased the rate of municipal solid waste generation are booming economies; rapid, uncontrolled and unmonitored urbanization; and a rise in human living standards (Guerrero et al., 2013). Current global municipal solid waste (MSW) generation levels are approximately 1.3 billion tons per year and are expected to increase to approximately 2.2 billion tons per year by 2025, which represents a significant increase in per capita waste generation rates, from 1.2 to 1.42 kg per person per day (World Bank Report, 2012). According to a study done by World Bank, it has been estimated that per capita waste generation rates in the countries such as China, India, Indonesia, Thailand, Bangladesh, Cambodia, Vietnam, and Malaysia will climb by 1-2 times between 1998 and 2025 (Singh et al., 2011).

Currently, 90-95% of total generated wastes in most of the developing countries are disposed of in open areas, street curbs, etc. and in landfills in developed countries (Pudasaini, 2014). Improving solid waste management, especially in the rapidly growing cities of developing countries, is becoming a more urgent issue these days. If these wastes are not managed properly, it will lead to the severe deterioration of environmental quality causing serious threats to human health.

Although landfills alleviate many health issues associated with open dumping of solid waste, landfills pose health and environmental issues of their own. In particular, landfills are sources of methane and carbon dioxide emissions, which are called greenhouse gases (GHGs). Carbon dioxide is the most prevalent greenhouse gas, and methane ranks second. In 2013, methane accounted for about 10% of all U.S. greenhouse gas emissions from anthropogenic sources. Globally, methane accounted about 16% of total greenhouse gas emissions (IPCC, 2014). Over 60% of total CH₄ emissions comes from human activities, such as industry, agriculture, and waste management activities (EPA, 2015). Methane's lifetime

in the atmosphere is much shorter than carbon dioxide (CO₂) but it is more efficient at trapping radiation compared to CO₂. The comparative impact of CH₄ on climate change is at least 28 times greater than CO₂ over a 100-year time horizon, and methane can stay in the atmosphere as long as 12 years (EPA / IPCC Report, 2013).

Figure 1-1 shows the US methane emissions by source. US landfills and manure management totaled about 28% of overall methane emissions. Emissions of methane in the United States decreased by 15% between 1990 and 2013. During that period, emissions from agricultural activities have increased and exploration and production of natural gas and petroleum products have decreased. In 2013, U.S. total greenhouse gas emissions totaled 6,673 million metric tons of carbon dioxide equivalents.

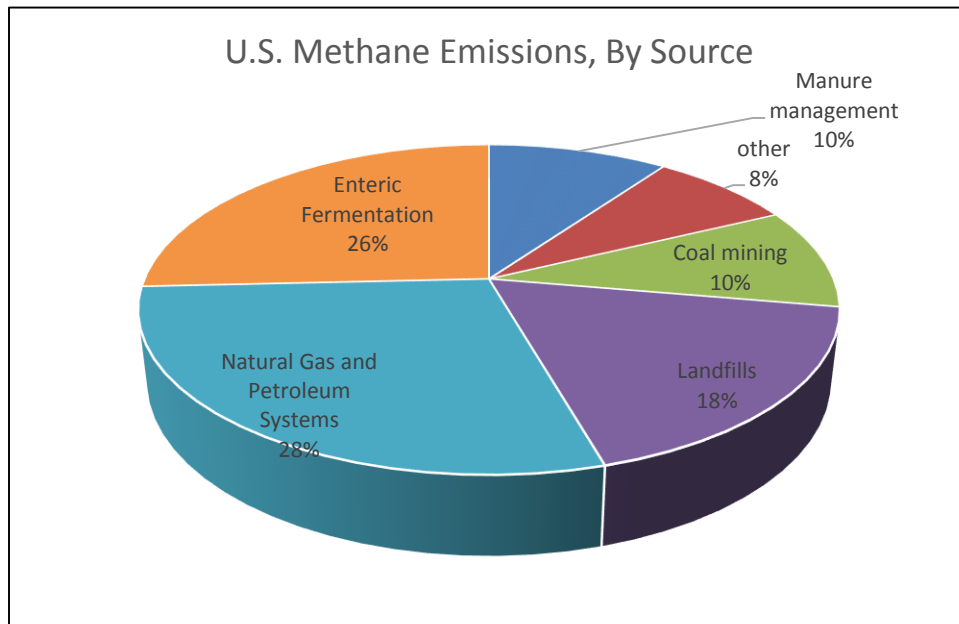


Figure 1- 1 US methane emissions by source (USEPA, 2015)

Fortunately, methane from anaerobic decomposition of waste in landfills, as well as anaerobic digesters, can be used as a renewable source of energy, for both developed and developing countries. Burning methane to generate electricity converts the methane to carbon dioxide, which has 28 times lower climate change potential. Energy is a basic tool for development (Anushiya, 2011). These days the energy

crisis is very distressing in the rural community of developing countries. Fuelwood has been and still is the major source of daily used fuel in the rural mass of peoples. Globally over 1.3 billion people (18% of global population) are without access to electricity and 2.6 billion people (38% of global population) are without clean cooking facilities. More than 95% of these people are either in sub-Saharan African or developing Asia (IEA, 2015). Bioenergy accounts for roughly 10% of world total primary energy supply today. Most of this is consumed in developing countries for cooking and heating. About 3.5% of road transportation fuel is provided today by biofuels (IEA, 2015).

1.2 Anaerobic Digestion

Anaerobic digestion is a biochemical process in which various microorganisms transform high molecular weight complex organic compounds to methane, along with carbon dioxide, water vapor and a small amount of other trace gases such as hydrogen sulfide, hydrogen, nitrogen, and carbon monoxide. Anaerobic digestion (AD) has the opportunity to be an essential part of the solution to the pressing global environmental concerns of waste management and renewable energy. Depending upon the source of organic materials used in AD, biogas contains typically 60- 70 % of methane (Ras et al, 2007). Figure 1-2 shows the schematic of the anaerobic digestion process.

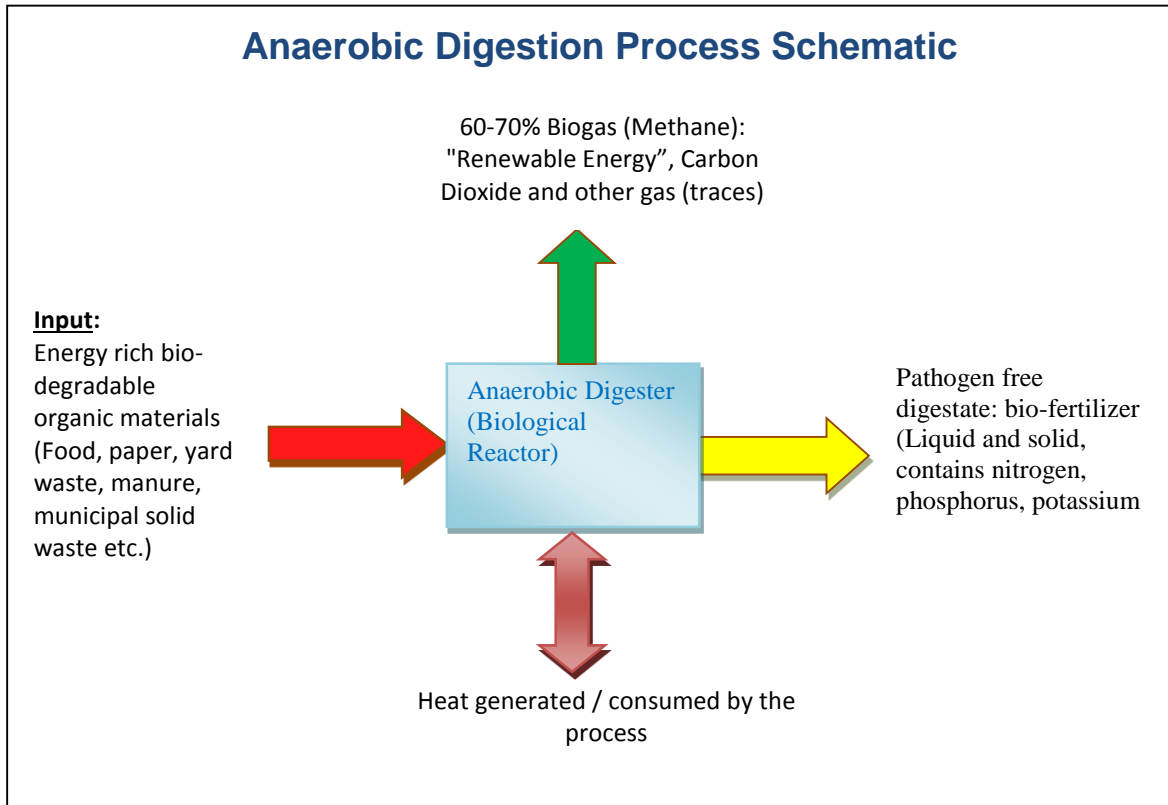


Figure 1- 2 Anaerobic digestion process schematic

Biogas is a renewable, high-quality fuel, produced from degradation of different organic raw materials, and can be captured and used for various energy services. The volume of sludge produced by an anaerobic process is less than 20 % of the volume generated by an aerobic process (van Lier, 2008). So, AD has been successful in reducing the volume of waste going to landfills. The problems associated with waste disposal can be alleviated by the generation of useful products, which also decreases the release of the potent greenhouse gas methane from landfill sites. The process of converting organic material into biogas can serve as a substitute for fossil fuels and artificial fertilizers. Biogas technology has been developed and widely used over the world because it has a lot of advantages, including reducing the dependence on non-renewable resources, high energy-efficiency, environmental benefits, available and cheap resources to use as feedstock, relatively easy and cheap technology for production, and nutrient rich digestate as a fertilizer (Zhang, 2012). It has already been proven that this technology is easily

implemented, cost-efficient, small-scale, and completely decentralized renewable alternative, which is technically feasible and economically viable (Zhang, 2012; SSWM, 2015). Figure 1-3 shows the various advantages of biogas plants.

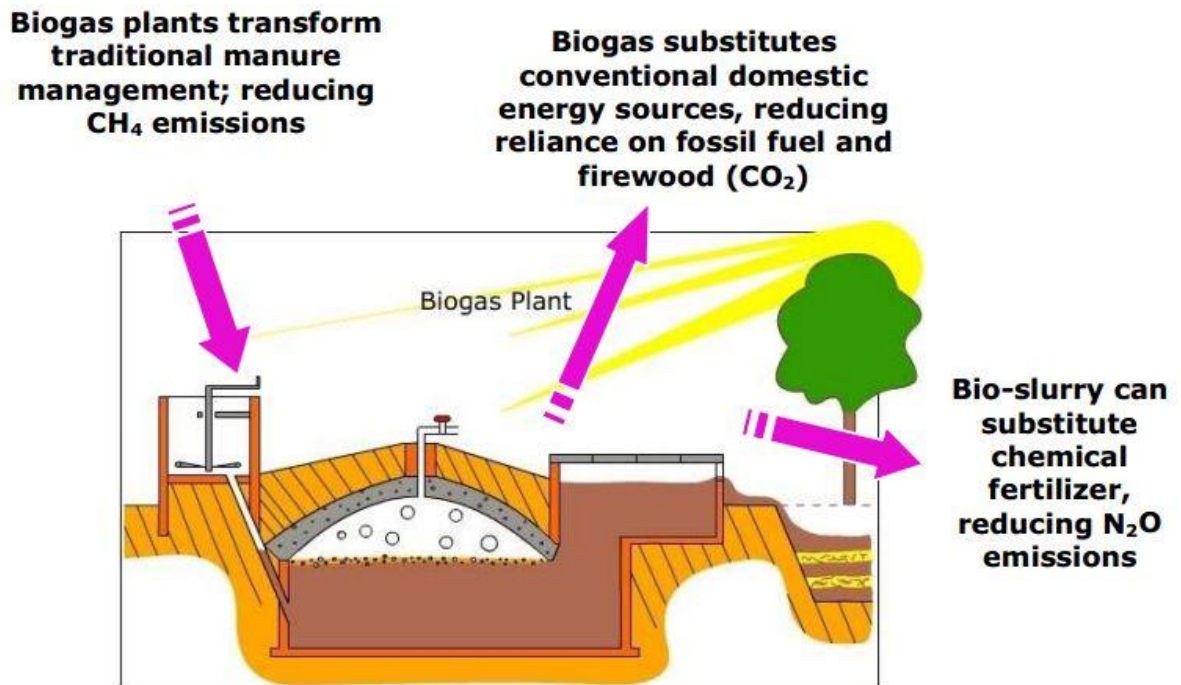


Figure 1- 3 Biogas plant (PBPO, 2006)

Advantage of anaerobic digesters include: (Laurel, 2011)

- Improved indoor air quality: saves women and children from exposure to smoke in the kitchen
- Time savings for women and children in collecting firewood,
- A cost-effective and simple technology that is ideal for small scale applications.
- Provides a non-polluting and renewable source of energy.
- An efficient way of energy conversion (preventing deforestation).

- Produces nutrient –rich organic effluent, which can supplement or even replace chemical fertilizers.
- Household wastes and bio-wastes can be disposed of usefully and in a healthy manner, which leads to improvement in the environment, and sanitation and hygiene.
- Provides a source for decentralized power generation.
- Leads to employment generation in the rural areas.
- Any biodegradable organic matter can be used as the substrate.
- Anaerobic digestion inactivates pathogens and parasites and is quite effective in reducing the incidence of water-borne diseases.
- Environmental benefits on a global scale: Biogas plants can lower the greenhouse effects on the earth's atmosphere. The plants lower methane emissions by entrapping the harmful gas and using it for energy.

1.3 Thesis objectives

One of the most important factors affecting anaerobic degradation of waste to produce biogas is a selection of organic material as inoculum. The source of inocula not only affects the amount of biogas production but also influences the overall performance of the reactor. Previous studies have found sewage sludge to be an effective inocula. However, one of the most important applications of biogas systems is in rural areas of low-income countries, where clean-burning fuel for cooking can be produced from degradation of household organic wastes. Most developing countries do not have wastewater treatment facilities in the rural areas, so using sewage sludge as a seed for an anaerobic digester is not an option. Dungs from various animals can be easily available, and have been examined as inocula in previous studies. Pond bottom soil is also a readily available source of inocula, and can be found with in walking distance and has not been examined in previous digester studies. People in the rural areas of low income countries often do not have accessibility to transportation facilities. Even if they are accessible to agricultural

roads, they likely do not have a car . Thus, peoples of those areas would prefer a locally and easily available source of inoculum to use as seed for biogas plant, such as dung or pond bottom soil microbes..

The **overall goal** of this study was thus to compare the effectiveness of inocula readily-available in developing countries for anaerobic reactors treating household waste. **Specific objectives** were:

- 1) To compare the rate of methane production and cumulative methane generation from degradation of a mixture of household organic wastes (food, paper, and yard) using 2 types of pond soil and horse dung as examples of inocula readily-available in developing countries.
- 2) To determine the effectiveness of the best inoculum from the first experiment in degrading mixtures with various proportions of food, paper and yard waste. Methane production (quantity and rate) from each type of waste will be measured.

1.4 Thesis outline

The remainder of the thesis is outlined in the following manner:

- The second chapter reviews the literature on greenhouse gas emissions, composition of municipal solid waste, landfill gasses and factors affecting landfill gas production, degradation phases of organic waste, biogas and its importance, benefits and types, effects of inoculum sources in the performance of digester etc.
- The third chapter describes sample collection, experimental setup, methodologies, and procedure for a laboratory test to address the present research objectives.
- The fourth chapter presents, analyzes and discusses experimental results and findings.
- The fifth chapter summarizes the main conclusions of this study and provides some recommendations for future research work.

Chapter 2

Literature Review

2.1 Background

This chapter include background information on greenhouse gases (GHG), sources of GHG emissions, landfills, degradation of wastes and generation of landfill gases, municipal solid wastes and composition, biogas, degradation of waste in anaerobic digesters (AD) and factors affecting performance of AD, importance of biogas in developing countries, global overview of biogas and effect of inoculum on overall performance of digesters.

2.2 Greenhouse Gases

A greenhouse gas (GHG) is a gas in an atmosphere that absorbs and emits radiation within the thermal infrared range. When sunlight strikes the Earth's surface, some of it is re-radiated back toward space as infrared radiation. Greenhouse gases are transparent to the short-wave incoming radiation but opaque to outgoing long-wave radiation. As a result, GHG absorb this outgoing infrared radiation and trap its heat in the atmosphere. There are many chemical compounds in the atmosphere that exhibit the greenhouse properties. Some are naturally occurring, such as carbon dioxide (CO₂), methane (CH₄), nitrous oxide (N₂O) and water vapor, and some gases are produced by humans and human activities such as hydrofluorocarbons (HFCs), perfluorocarbons (PFCs), sulfur hexafluoride (SF₆), nitrogen trifluoride (NF₃).

GHG produce a natural warming to sustain life on the Earth. Without greenhouse gases, the average temperature of Earth's surface would be about 15 °C (27 °F) colder than the present average of 14 °C (57 °F) In the past hundred years, however, the average temperature of Earth has risen by 1.5°F, and is projected to rise another 0.5 to 8.6°F over the next hundred years (US EPA, 2015). Even a small change in the average temperature of the Earth can convert to potentially dangerous shifts in climate and weather.

Global warming potential (GWP) is a relative measure of how much heat a greenhouse gas can trap in the atmosphere. GWP measures the amount of heat trapped by a certain mass of a gas over a given period of time, relative to the amount of heat trapped by a similar mass of carbon dioxide (USEPA 2015). Different greenhouse gases have different effects on global warming, which depend on their ability to absorb energy and how long they stay in the atmosphere. The time scale usually used for GWP is 100 years. Policy makers use the values GWP to compare the impacts of emissions and reductions of different gases (US EPA, 2015). Methane (CH₄) has a shorter lifetime but higher energy absorption capacity compared to CO₂, which is reflected in its GWP. The GWP of methane is estimated to be 28-36 over 100 year period. Nitrous Oxide (N₂O) has a GWP 265-298 times that of CO₂ for a 100-year timescale. The high-GWP (thousands or tens of thousands) gases such as chlorofluorocarbons (CFCs), hydrofluorocarbons (HFCs), hydrochlorofluorocarbons (HCFCs), perfluorocarbons (PFCs), and sulfur hexafluoride (SF₆) can trap substantially higher heat than CO₂ (US EPA, 2015). In 2013, U.S. greenhouse gas emissions totaled 6,673 million metric tons of carbon dioxide equivalents (US EPA, 2015).

Globally 60 % of GHG emissions comes from human-related activities. Methane (CH₄) is the second most prevalent greenhouse gas emitted in the United States from human activities. Methane accounts for 16% of the global GHG emissions and 10% of total US GHG emissions. Landfills are the third largest anthropogenic source of US methane emissions, accounting for 18.2 % in 2013 (US EPA, 2015). Figures 2-1 and 2-2 show the US and global GHG emissions, respectively.

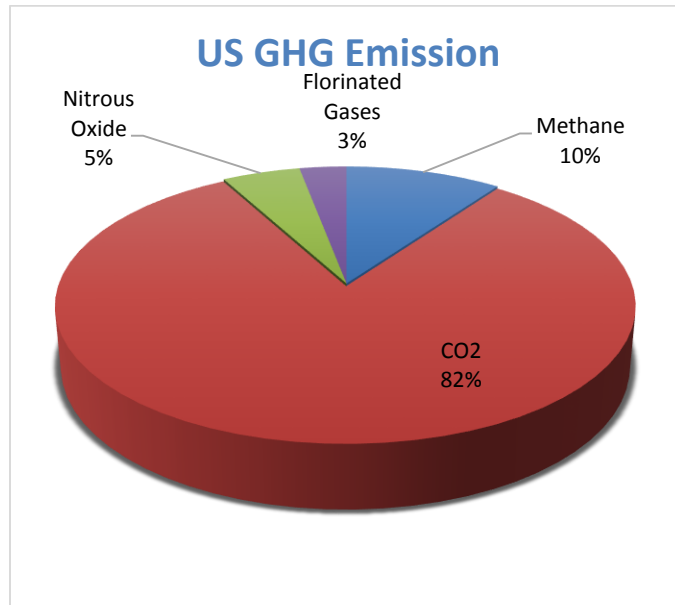


Figure 2- 1 US greenhouse emissions (US EPA, 2015)

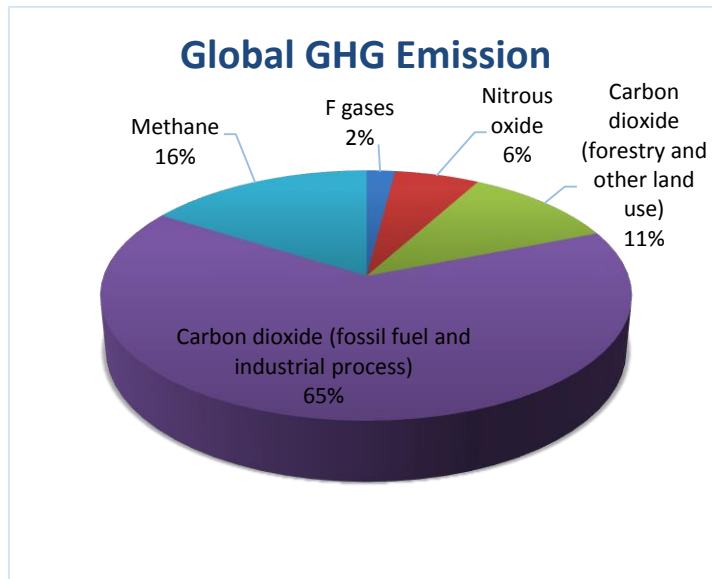


Figure 2-2 Global greenhouse gas emissions (US EPA, 2015)

2.3 Municipal Solid Waste

Municipal solid waste (MSW), more commonly known as trash or garbage, is waste consisting of everyday items that are discarded by the public, such as product packaging, yard waste, furniture, clothing,

plastics, bottles, food waste, newspapers, appliances, glass, metals and batteries. In other words, MSW is all types of solid waste generated by households and commercial establishments usually collected by local government bodies. According to US EPA (2011), municipal waste does not include industrial wastes, wastes from construction and hazardous wastes.

2.3.1 *Composition of municipal solid waste*

Most of the MSW typically being disposed of in landfills is food waste, paper (e.g., newspaper, office paper, paperboard, cardboard, etc.) and yard wastes (e.g., leaves, grass, tree limbs, etc.). Typical composition of MSW and their ranges are given in table 2-1 below.

Table 2- 1 Typical MSW Composition (% by weight) for United States (Tchobanoglous et al., 1993)

Component	Typical Range	Organics
Food Wastes	6–18	9.0
Paper	25–40	34.0
Cardboard	3–10	6.0
Plastics	4–10	7.0
Textiles	0–4	2.0
Rubber	0–2	0.5
Leather	0–2	0.5
Yard Wastes	5–20	18.5
Wood	1–4	2.0
Glass	4–12	8.0
Tin Cans	2–8	6.0
Aluminum	0–1	0.5
Other Metal	1–4	3.0
Dirt, Ash, etc.	0–6	3.0

Food waste, paper and cardboard and yard waste are the three largest sources of municipal solid waste in US. Total municipal solid waste (MSW) generation in the USA in 2013 was 254.1 million tons. Figure 2-3 shows the breakdown of MSW generation by material. Organic materials such as paper and paperboard, yard trimmings and food continue to be the largest components of MSW.

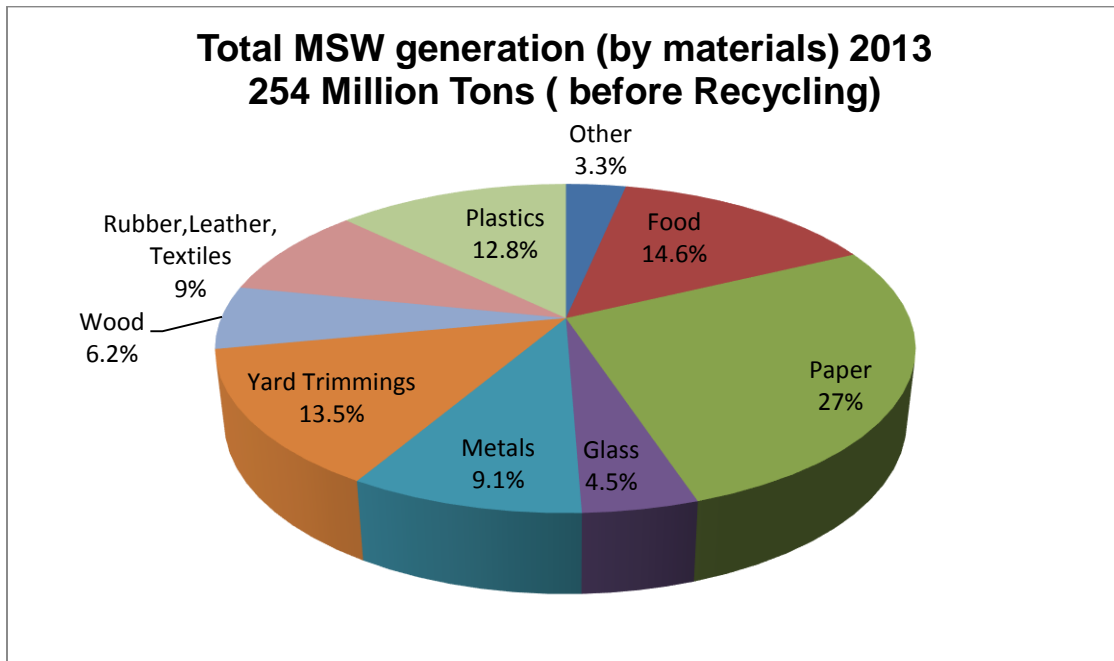


Figure 2- 3 Total MSW generation (by materials) 2013, 254 million tons (before Recycling) (EPA, 2015)

In 2013 total MSW recovery was over 87 million tons total MSW generation, as shown in Fig. (2-4). Recycling and composting MSW saved almost 1.1 quadrillions Btu of energy, which is the same amount of energy consumed by over 9.9 million U.S. households in a year. This provides an annual reduction of more than 186 million metric tons of carbon dioxide equivalent emissions, comparable to the annual GHG emissions from over 39 million passenger vehicles (EPA, 2015).

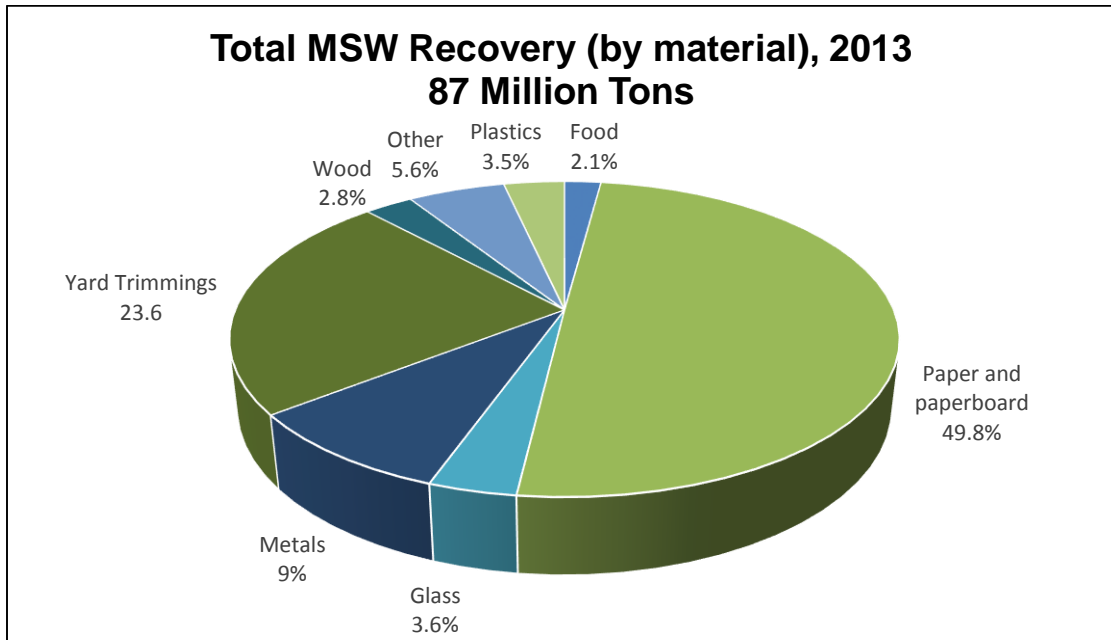


Figure 2- 4 Total MSW recovery (by material) 2013, 87 million tons (EPA, 2015)

About 34.3 percent of Total generated waste materials was recovered (recycling and composting) out of the total generated in USA with recycling rate of 34.3 percent. Figure 2-5 shows the US MSW recycling rates from 1960-2013 and Table 2-2 shows the generation, recovery and discards of materials in MSW, in 2013.

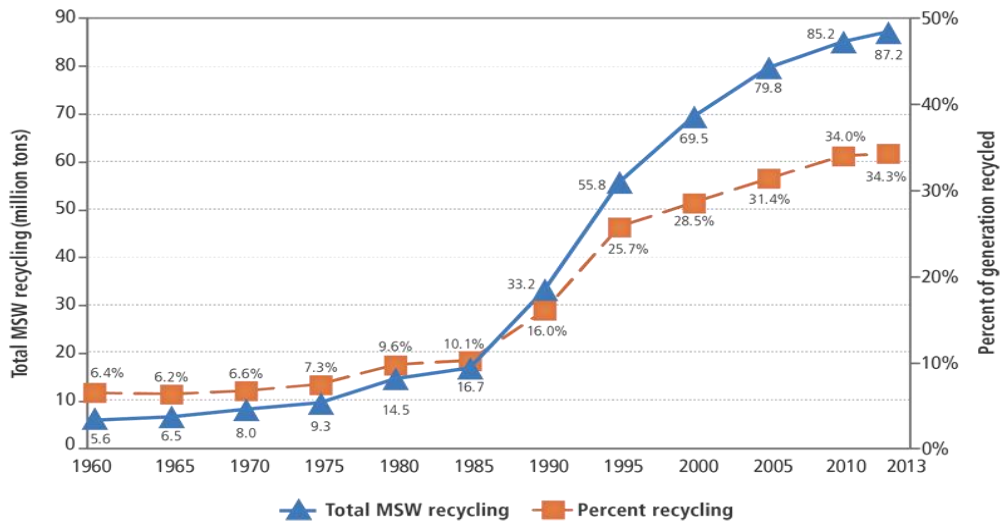


Figure 2- 5 MSW recycling rates, 1960-2013 (EPA, 2015)

Table 2- 2 Total waste Generated, Recovered and Discarded in 2013 (in millions of tons and percent of generation of each material) (EPA, 2015)

Material	Weight Generated	Weight Recovered	Recovery as Percent of Generation	Weight Discarded
Paper and paperboard	68.60	43.40	63.3%	25.20
Glass	11.54	3.15	27.3%	8.39
Metals				
Steel	17.55	5.80	33.0%	11.75
Aluminum	3.50	0.70	20.0%	2.80
Other nonferrous metals†	2.01	1.37	68.2%	0.64
Total metals	23.06	7.87	34.1%	15.19
Plastics	32.52	3.0	9.2%	29.52
Rubber and leather	7.72	1.24	16.1%	6.48
Textiles	15.13	2.30	15.2%	12.83
Wood	15.77	2.47	15.7%	13.30
Other materials	4.58	1.31	28.6%	3.27
Total materials in products	178.92	64.74	36.2%	114.18
Other wastes				
Food, other	37.06	1.84	5.0%	35.22
Yard trimmings	34.20	20.6	60.2%	13.60
Miscellaneous inorganic wastes	3.93	Negligible	Negligible	3.93
Total other wastes	75.19	22.44	29.8%	52.75
Total municipal solid waste	254.11	87.18	34.3%	166.93

2.3.2 Cellulose, hemicellulose and lignin content in MSW

Several studies have been done to find the ultimate methane potential of waste by finding the cellulose, hemicellulose and lignin content of the different waste materials (Barlaz et al. 1990; Rees, 1980; Eleazer et al., 1997; Komilis and Ham, 2003; Rao et al., 2000; Ress et al., 1998; Jones et al., 1983; Rhew and Barlaz, 1995). Typical cellulose, hemicellulose and lignin content found in municipal solid waste components are shown below in Table 2-3.

Cellulose and hemicellulose components are easily degradable under anaerobic conditions but lignin is assumed to be poorly degradable and is less affected during biological degradation. It has been reported from several researchers that as the age of the landfill waste increases, the ratio of cellulose and hemicelluloses to lignin decreases (Mehta et al., 2002; Barlaz, 2006; Bookter and Ham, 1982). According to Barlaz (2006), methane generated due to decomposition of cellulose and hemicellulose can be calculated by using the following equations:

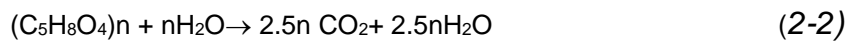
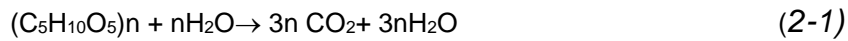


Table 2- 3 Cellulose, Hemicellulose, and Lignin Content of Different Waste Components
Reported in the Literature

Waste	References	Cellulose	Hemicellulose	Lignin
Food	Eleazer et al. (1997)	55.4	7.2	11.4
	Komilis and Ham (2003)	46.09	0.0	12.03
Grass	Komilis and Ham (2003)	39.67	16.89	17.63
	Eleazer et al. (1997)	26.5	10.2	28.4
Leaves	Eleazer et al. (1997)	15.3	10.5	43.8
	Komilis and Ham (2003)	9.48	3.24	33.88
Office paper	Eleazer et al. (1997)	87.4	8.4	2.3
Mixed Paper	Komilis and Ham (2003)	69.6	7.79	15.9
MSW	Eleazer et al. (1997)	28.8	9.0	23.1
	Barlaz (1990)	51.2	11.9	15.2

According to Eleazer et al. (1997), the bio methane potential of food waste, grass, leaves, office paper, old newsprint are 0.30, 0.14, 0.03, 0.217, and 0.074 liters per kilogram of waste, respectively. Since these waste materials are composed of primarily cellulose, hemicellulose, and lignin. Food wastes, paper waste and grass have high level of cellulose and hemicellulose content. Out of the selected wastes in this study, office paper has the highest percentage of cellulose, leaves has the highest percentage of hemicellulose and lignin. Material which contains high amount of cellulose and hemicellulose are known to be faster degradable. Yard waste such as grass , leaves and paper material will biodegrade to methane and other gasses but the rate of the biodegradation is very slow compared to other wastes at traditional landfills such as food wastes because of the lignin and cellulose content (Tchobanoglous et al., 1993). Table 2-4 shows the selected wastes biodegradable fraction based on the lignin content.

Table 2- 4. Biodegradable Fraction of Selected Organic Waste Based on the Lignin Content

(Tchobanoglous et al., 1993)

Component	Volatile solids (VS) % of total solid (TS)	Lignin content (LC), % of VS	Biodegradable fraction (BF)
Food wastes	7–15	0.4	0.82
Newsprint paper	94	21.9	0.22
Office paper	96.4	0.4	0.82
Yard wastes	50–90	4.1	0.72

2.4 Anaerobic Decomposition of Waste

A landfill is a place to dispose of refuse and other waste material by burying it and covering it over with soil, especially as a method of filling in or extending usable land. Worldwide landfills and open dumps are the most dominant types of solid waste disposal methods (IPCC, 2007). Organic matter contained in the solid waste disposal site (landfill) undergoes biological transformation and produces greenhouse gases under aerobic and anaerobic conditions. Methane, one of the prevalent GHG, is produced from the landfill, depending on the composition and characteristics of the wastes.

2.4.1 Landfills as sources of greenhouse gas emissions

Landfill gas is the byproduct of natural decomposition of municipal solid waste in an anoxic condition. Landfill gas consists of about 50-55% CH₄, about 45-50% CO₂, and a small amount of other gases. As mentioned already, in the US 18.2% of total methane emissions comes from landfills in 2013 (USEPA, 2015). If this amount of CH₄ is not collected from the landfill, it will escape to the atmosphere. The amount of methane generation from the landfills depends on various factors, such as the waste composition, compaction, unit weight, age of the waste, pH, particle size, and initial moisture content and climatic factors such as the annual rainfall and temperature.

2.4.2 Composition of landfill gas

Methane and carbon dioxide are the principal landfill gases and hydrogen sulfide, hydrogen, carbon monoxides are trace gases. Some trace gases which are in small amounts, such as hydrogen sulfide (smells like rotten eggs), can be toxic. The typical composition of landfill gas is given in Table 2-5.

Table 2- 5 Typical Composition of Landfill Gas (Source: Tchobanoglous et al., 1993, EPA 1995)

Component Percent (dry volume basis)

Methane	45-60
Carbon dioxide	40-60
Oxygen	2-5
Sulfides, disulfides, mercaptans, etc.	0.1-1.0
Ammonia	0.1-1.0
Hydrogen	0-0.2
Carbon monoxide	0-0.2
Trace constituents	0.01-0.6

The constituents of landfill gas are discussed in more detail below.

2.4.2.1 Methane

Methane is a colorless and odorless naturally occurring gas. It is flammable and explosive in high concentrations (ATSDR, 2001).

2.4.2.2 Carbon dioxide

Carbon dioxide is naturally found at small concentrations in the atmosphere (0.03%), which is colorless, odorless, and slightly acidic (ATSDR, 2001). During the initial decomposition of the landfill wastes, the concentration of carbon dioxide is high, which reduces the pH during the initial phase. When the displacement of aerobic reactions by anaerobic reaction starts, the concentration decreases accordingly. Finally, in the methane production phase, carbon dioxide is stabilized (ATSDR, 2001).

2.4.2.3 Oxygen

Oxygen comprises approximately 21% of the atmosphere. It is odorless, tasteless, and colorless. The concentration of oxygen decreases as the decomposition phase's move from aerobic to anaerobic.

2.4.2.4 Sulfides

Sulfides such as hydrogen sulfide, dimethyl sulfide, and mercaptans are naturally occurring gases that give the landfill gas mixture its rotten-egg smell. Even at very low concentrations, sulfides can cause unpleasant odors (ATSDR, 2001).

2.4.2.5 Ammonia

Ammonia is a colorless gas with a pungent odor.

2.4.2.6 Hydrogen:

Hydrogen is an odorless, colorless gas.

2.4.2.7 Carbon Monoxide

Carbon monoxide is an odorless, colorless gas.

2.4.2.8 Trace constituents: NMOCs (non-methane organic compounds)

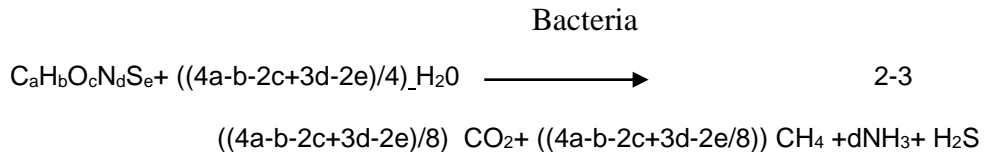
NMOCs are organic compounds (compounds that contain carbon). Methane is an organic compound but is not considered an NMOC. NMOCs may occur naturally or be formed by synthetic chemical processes. NMOCs most commonly found in landfills include acrylonitrile, benzene, 1, 1-dichloroethane, 1,2-cis dichloroethylene, dichloromethane, carbonyl sulfide, ethyl-benzene, hexane, methyl ethyl ketone, tetrachloroethylene, toluene, trichloroethylene, vinyl chloride, and xylenes (ATSDR, 2001).

2.4.3 *Phases of anaerobic degradation of organic wastes*

Anaerobic digestion is a series of complex and interconnected biological processes that is carried out by different species of microorganisms in the absence of the oxygen. After the municipal waste is deposited in the landfill site, the conversion of solid waste to methane and carbon dioxide takes place by several microorganisms and by a series of chemical reactions (aerobic and anaerobic). Landfills generally accept MSW over a 20-30 year period, so that wastes in the landfill may be undergoing several decomposition steps at the same time. Generally, decomposition of landfill waste takes places in four

phases and the composition of gases produced changes in each of the phases (US EPA, 2015). The older waste in one area might be in a different phase of decomposition than the waste which has been dumped recently in another area of the same landfill (ATSDR, 2001).

The process of converting organic waste to methane and carbon dioxide can be expressed as following equation 2-3 (Cooper et al., 1992).



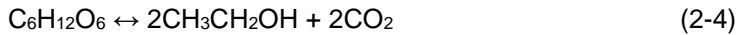
The degradation of organic waste occurs in four Phases/ stages (Figure 2-6 below): hydrolysis acidogenesis, acetogenesis and methanogenesis

2.4.3.1 Hydrolysis (Phase I, Aerobic or Lag phase)

This is the first step for the most fermentation processes. The participating bacteria cannot directly process the organic substrate. The substrate particularly has organic material consisting of proteins, carbohydrates and lipids which must first be broken down into soluble polymers or monomers, like amino acids, sugars and fatty acids (Gujer and Zehnder, 1983). This process is called hydrolysis (polymer breakdown). The process (hydrolysis) is usually the rate-limiting step in the process of anaerobic digestion of particulate organic substrates (Zeeman and Sanders, 2001). This is because the bacteria responsible for the liquefaction of complex compounds are operating at a very slow rate at this step, compared to the following steps, and are also highly dependent on digester conditions, such as availability of substrate, bacterial population density in the inoculum used, temperature and pH (Evans, 2001). Aerobic bacteria break down the long chain of complex carbohydrates, proteins, and lipids that form the content of organic waste. While breaking down the substrate aerobic bacteria consume oxygen and mainly produce carbon dioxide as a by-product. The aerobic phase continues until available oxygen is depleted. Phase I decomposition can last for days or months, depending on the amount of oxygen present in the landfill waste (ATSDR, 2001).

2.4.3.2 Acidogenesis (Phase II, Transition phase)

Anaerobic decomposition starts after all the available oxygen in the landfill waste has been used up. In the second step, acidogenic bacteria transform the products of the first reaction into short chain volatile acids, ketones, alcohols, hydrogen and carbon dioxide. The principal products of this step are propionic acid ($\text{CH}_3\text{CH}_2\text{COOH}$), butyric acid ($\text{CH}_3\text{CH}_2\text{CH}_2\text{COOH}$), acetic acid (CH_3COOH), formic acid (HCOOH), lactic acid ($\text{C}_3\text{H}_6\text{O}_3$), ethanol ($\text{C}_2\text{H}_5\text{OH}$) and methanol (CH_3OH), among others. The landfill becomes highly acidic. When moisture present in the landfill mixes with acid formed in the first phase, nitrogen and phosphorus are consumed by certain species of bacteria; then carbon dioxide and hydrogen are produced. Hydrogen, carbon dioxide and acetic acid will skip the acetogenesis stage and be utilized directly by the methanogenic bacteria in the final stage. Three typical acidogenesis reactions where glucose is converted to ethanol, propionate and acetic acid, respectively, are represented by equations 2-4, 2-5 (Ostrem, 2004) and 2-6 (Bilitewski et al., 1997).



Figures 2-6 and 2-7 show degradation of pyruvic acid and acetic acid through butyric acid pathway

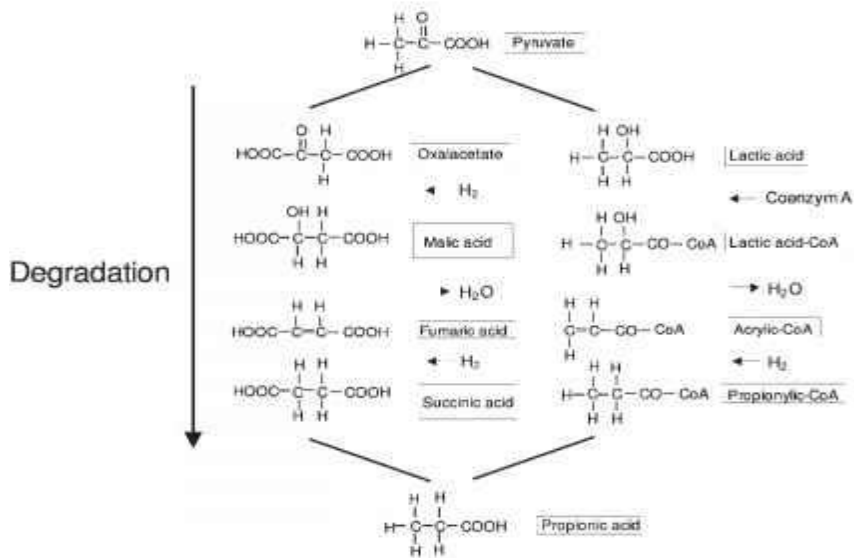


Figure 2- 6 Degradation of pyruvate acid (Free energy planet, biogas from waste, 2015)

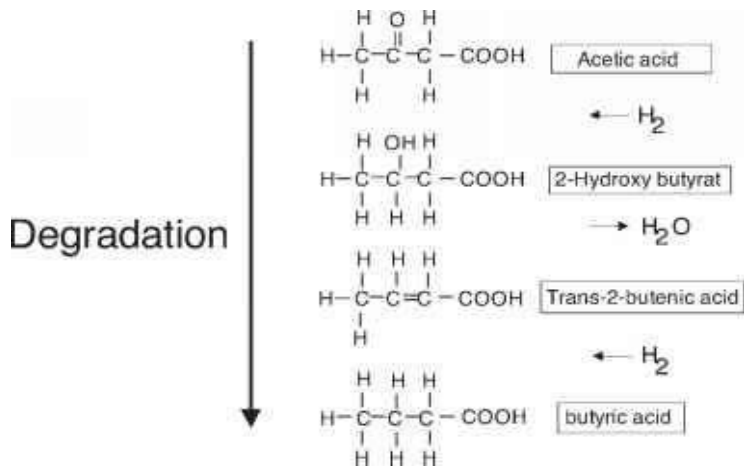


Figure 2- 7 Degradation of acetic acid via butyric acid pathway (Free energy planet, biogas from waste, 2015)

2.4.3.3 Acetogenesis (Phase III, accelerated methane production phase)

In third stage (acetogenesis phase), the rest of the acidogenesis products, i.e. the propionic acid, butyric acid and alcohols, are converted by acetogenic bacteria into acetic acid (CH_3COOH)/acetate (CH_3COO^-), hydrogen and carbon dioxide. In this process the pH becomes more neutral so the methane-

forming bacteria start to produce methane. Methanogenic bacteria can consume carbon dioxide and acetate (compound that was created by the acid forming bacteria) .In this phase, methane and acid-producing bacteria have a symbiotic, mutually beneficial relationship (Barlaz et el, 1990; Rees, 1980). The final products of the acetogenesis process (acetate, H₂ and CO₂) are precursors of methane formation (Metcalf and Eddy, 2003). During the third stage, propionate (equation 2-7), butyric acid (equation 2-8), ethanol (equation 2-9) and glucose (equation 2-10) among others are converted to acetate (Ostrem, 2004).

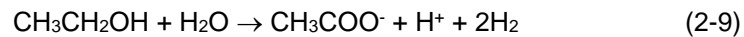
Propionate → acetate



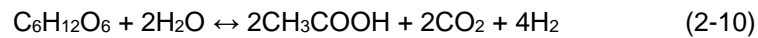
Butyrate → acetate



Ethanol → acetate



Glucose to acetate



2.4.3.4 Methanogenesis (Phase IV decelerated methane production phase)

The fourth step methanogenesis is carried out by a group of microorganisms known as methanogens (strict anaerobes) . Microorganisms responsible for methane production are classified as archaea. Many of the methanogenic bacteria found in anaerobic digesters are similar to those found in the stomachs of ruminant animals and inorganic sediments taken from ponds, lakes and rivers. There are two groups of microorganisms that produce methane. One group, termed acetoclastic methanogens, splits the acetate to methane and carbon dioxide; the other group, called hydrogen utilizing methanogens, use hydrogen as the electron donor and CO₂ as electron acceptor to produce methane (Metcalf and Eddy, 2003). Bacteria named acetogens are also able to oxidize hydrogen and form acetic acid, but the acetic acid is converted to methane so the impact of this reaction is very small. The following equations are the two main ways of methane production by methanogenic bacteria.



The composition and production rates of landfill gas are relatively constant in this phase. The landfill gas contains approximately 50% to 55% methane by volume, 45% to 50% carbon dioxide, and 2% to 5% other gases such as sulfides (EPA 2015). In this phase the landfill gas is produced at a stable rate, which is expected to last a long time (about 20 to 50 or more years) after the waste is placed in the landfill. Gas production duration in this phase depends upon the type of organic waste received at the landfill. The pH of this phase is similar to the third phase (Barlaz et al., 1990; Tchobanoglous et al., 1993).

About 72% of methane formation in anaerobic digestion is derived from acetate formation (Figure 2-8).

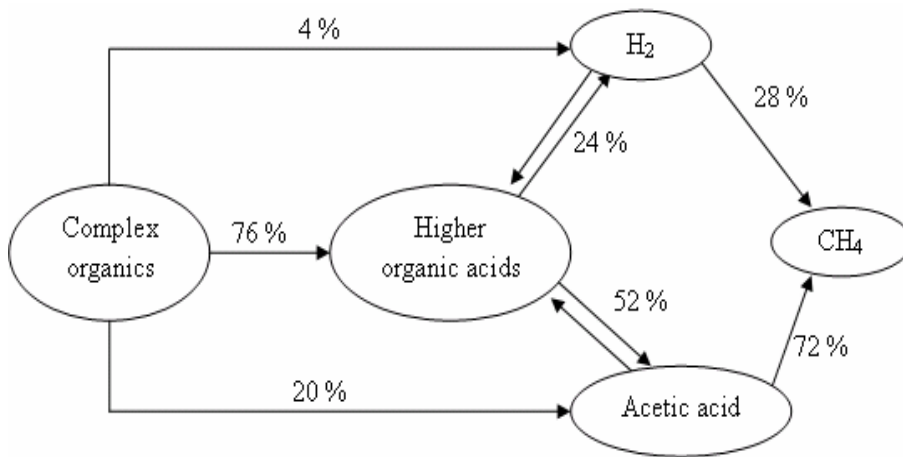


Figure 2- 8 Carbon and hydrogen flow in anaerobic process (the given percentage values are based on COD) (Metcalf and Eddy, 2003)

Table 2-6 shows the reactions that are carried out by methanogens during the anaerobic digestion process.

Table 2- 6 Reaction Carried out by Methanogens (Demirel et al., 2008)

Hydrogen	$4\text{H}_2 + \text{CO}_2 \rightarrow \text{CH}_4 + 2\text{H}_2\text{O}$
Acetate	$\text{CH}_3\text{COOH} \rightarrow \text{CH}_4 + \text{CO}_2$
Formate	$4\text{HCOOH} \rightarrow \text{CH}_4 + \text{CO}_2 + 2\text{H}_2\text{O}$
Methanol	$4\text{CH}_3\text{OH} \rightarrow 3\text{CH}_4 + \text{CO}_2 + 2\text{H}_2\text{O}$
Carbon monoxide	$4\text{CO} + 2\text{H}_2\text{O} \rightarrow \text{CH}_4 + 3\text{H}_2\text{CO}_3$
Trimethylamine	$4(\text{CH}_3)_3\text{N} + 6\text{H}_2\text{O} \rightarrow 9\text{CH}_4 + 3\text{CO}_2 + 4\text{NH}_3$
Dimethylamine	$2(\text{CH}_3)_2\text{NH} + 2\text{H}_2\text{O} \rightarrow 3\text{CH}_4 + \text{CO}_2 + 2\text{NH}_3$
Methylamine	$4(\text{CH}_3)\text{NH}_2 + 2\text{H}_2\text{O} \rightarrow 3\text{CH}_4 + \text{CO}_2 + 4\text{NH}_3$
Methyl mercaptans	$2(\text{CH}_3)_2\text{S} + 3\text{H}_2\text{O} \rightarrow 3\text{CH}_4 + \text{CO}_2 + \text{H}_2\text{S}$
Metals	$4\text{Me}^0 + 8\text{H}^+ + \text{CO}_2 \rightarrow 4\text{Me}^{++} + \text{CH}_4 + 2\text{H}_2\text{O}$

Following figure 2-9 and 2-10 shows the flow chart and phases of anaerobic degradation of organic waste respectively.

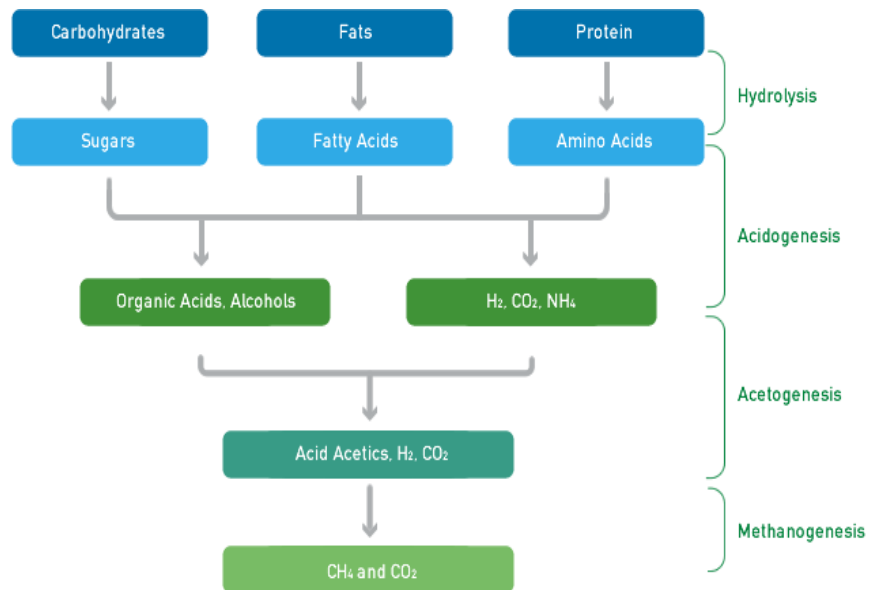


Figure 2- 9 Flow chart of anaerobic digestion

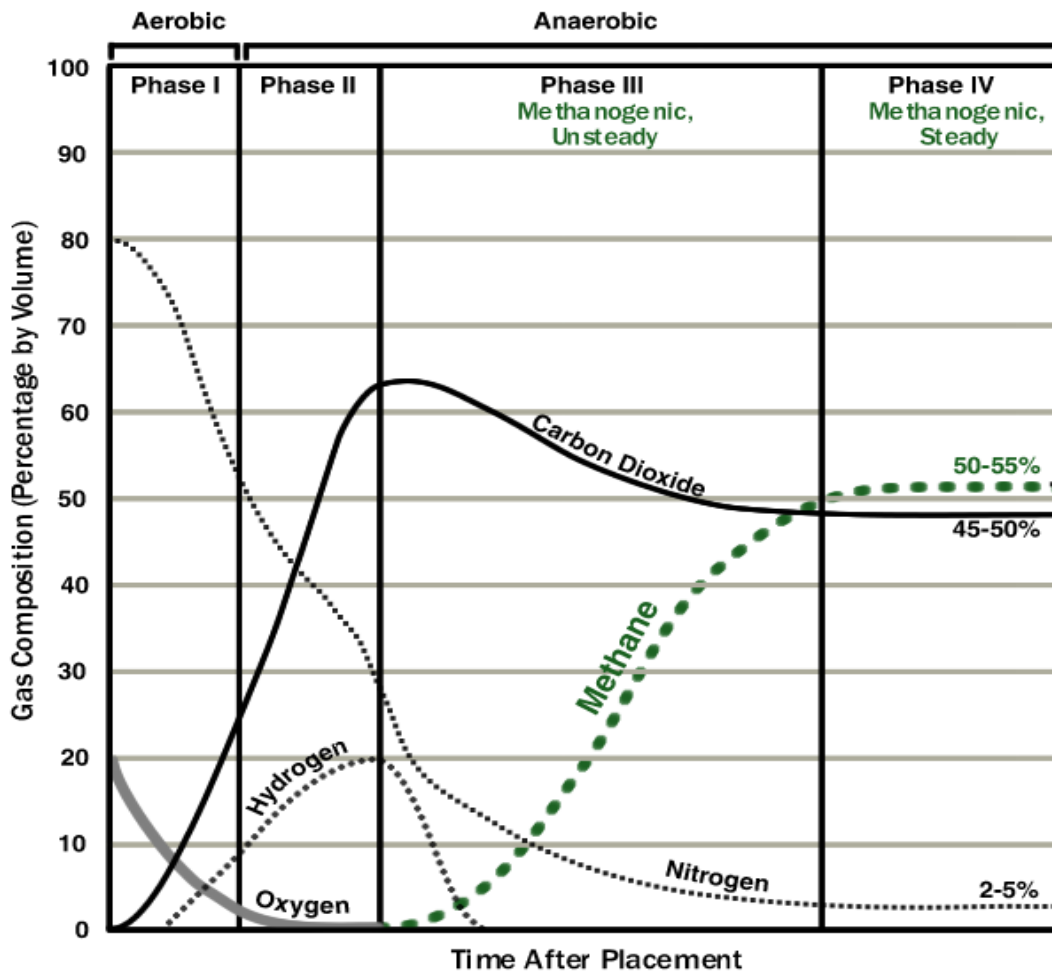


Figure 2- 10 Degradation phases in landfills (EPA, 1997)

2.4.4 Factors affecting landfill gas production

Landfill gas production depends substantially on the composition of the wastes in the landfills. Some organic waste in the landfill contains high amount of nutrients such as sodium, potassium, calcium, and magnesium which help bacteria live. When such kind of nutrients are present in the landfill wastes, the landfill gases increases accordingly. However, some landfill wastes contain material that inhibits methane-producing bacteria, such as high salt concentrations or high toxic substances, causing less gas production.

Generally municipal solid waste can be categorized as bio- degradable and non-biodegradable. Some materials like food, paper, yard, and wood waste, which have high amounts of cellulose and hemicellulose, are decomposable, whereas glass, plastics, metals construction and demolition debris are not easily decomposable and fall into the non- biodegradable category. Food waste in particular decomposes quickly. Figure 2-11 describes the relative rate of biodegradation of waste materials in the landfill.

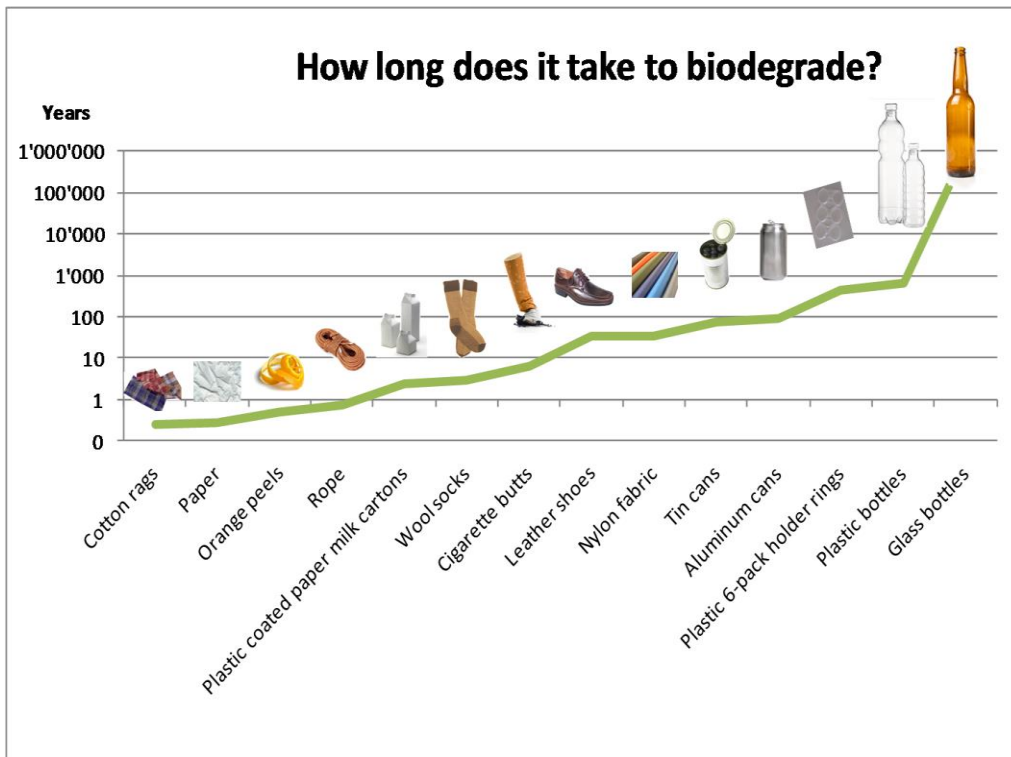


Figure 2- 11 Graphical representation of relative rate of degradation of different waste material in a landfill

Source (<https://enviropolicyintro.wordpress.com/tag/education/>)

Another important factor is a presence of oxygen in the landfill. The more oxygen present in a landfill, the longer the aerobic Phase I lasts. Only after the oxygen is used up can methanogenic bacteria begin to produce methane. Also, if the deposited waste is loosely buried or frequently disturbed, more oxygen is

available, and aerobic bacteria live longer and produce carbon dioxide and water for longer periods. Alternatively, if the waste is more compacted, aerobic bacteria are replaced by anaerobic bacteria in Phase III and methane production will begin earlier.

The presence of a certain amount of moisture in a landfill accelerates gas production, up to a point. The moisture content in the waste encourages bacterial growth and helps to transport nutrients. According to Rees (1980), maximum methane production has been obtained at the moisture content of 60-80%. If the water content is too high, however, gas transport is blocked, lowering methane production.

As warm temperatures increase, bacterial activity also increases up to a point, causing the rate of gas production to increase. Colder temperatures inhibit bacterial activity. Generally, bacterial activity drops off dramatically if the temperature is below 50° Fahrenheit (F). The bacterial activity releases heat, stabilizing the temperature of a landfill between 77° F and 113° F, although temperatures up to 158° F have been noted. Higher temperatures also promote volatilization and chemical reactions. In general, emissions of NMOCs double with every 18° F increase in temperature (ATSDR, 2001; EPA, 1993).

Landfill gas production also depends on the ages of the wastes buried in the landfill. More recently buried organic waste will produce more gas than older waste. Landfills usually produce appreciable amounts of gas within 1 to 3 years. The highest gas production usually occurs 5 to 7 years after wastes are deposited. Most of the gas is produced within 20 years after waste is dumped; however, small quantities of gas may continue to be emitted from a landfill for 50 or more years (ATSDR, 2001). Generally, more recently buried waste such as waste buried less than 10 years produces more landfill gas through bacterial decomposition, volatilization, and chemical reactions than older waste (buried more than 10 years) (ATSDR, 2001)

2.5 Biogas

Biogas is a combustible gas used as cooking gas in agricultural communities produced by anaerobic fermentation of different form of organic-rich substrates and is mainly composed of methane and

carbon dioxide. Solids remaining after the fermentation process is completed are rich in nutrients and used as organic fertilizer.

Biogas production by anaerobic digestion is popular for treating biodegradable waste because valuable fuel can be produced while destroying disease-causing pathogens and reducing the volume of disposed waste products. The methane in biogas burns more cleanly than coal, and produces more energy with less emissions of carbon dioxide. The capturing of methane and use as energy source is an important role of waste management, which reduces greenhouse gas emissions.

2.5.1 *Factors affecting anaerobic digestion (Biogas Production)*

Biological processes take place during anaerobic digestion are influenced largely by the conditions inside and outside of the bioreactor. In order to maintain favorable conditions for bacterial activity and to optimize reactor performance (methane production), which is the ultimate goal, some parameters need to be taken care of. These parameters, if not monitored, can cause instability in the digestion process and lower the potential biogas yield. The following are some of the factors that affect methane production.

2.5.1.1 Composition of Substrate

The amount of methane production depends greatly on the substrate composition, along with its particle size and degradability. Theoretical methane production can be calculated as follows.

$$Q_{\text{waste}i} = q_{\text{waste}i} * M_{\text{waste}i} * f_{\text{TS}} * f_{\text{oTS}} * 0.75$$

Where

$Q_{\text{waste}i}$ = Gas production rate (volume/day) for waste i

$q_{\text{waste}i}$ = Maximum specific yield of biogas for waste i (maximum biogas produced per total organic solid, vol/mass)

$M_{\text{waste}i}$ = waste feed rates for waste i (mass /day)

f_{TS} = fraction of the waste by weight that is solids

f_{oTS} = fraction of the waste by weight that are organic (volatile)

0.75 = factor to account for practical biogas yield

The smaller the waste particle size, the more surface area is exposed to bacteria, and the faster methane will be produced. Shredding the waste can increase the rate of methane production, particularly for wastes with a high content of structural materials (e.g. cellulose, lignin), which make it difficult for microbes to access and degrade the substrate. The yield for substrates can be increased by up to 20% by shredding (Deublein and Steinhauser, 2008). Table 2-7 shows extent of decomposition of different wastes and their bio methane yield.

Table 2- 7 Extent of Waste Decomposition and Methane Yield (Eleazer et al., 1997)

Waste	Methane Yield (ml of CH ₄ /dry gram)	Extent of decomposition
Food	300.7	84.1
Grass	144	94.3
Leaves	30.6	28.3
Office paper	217.92	54.6
Old newsprint	74.33	31.1
MSW	92	58.4

2.5.1.2 Retention time

In an anaerobic digester, there are two types of retention times, solids retention time and hydraulic retention time. Solids retention time (SRT) refers to the average time spent by the bacteria (solids) in the bioreactor. Hydraulic retention time is defined as the average amount of time liquid waste stays inside the reactor. If SRT is too low, there will be chances of organism washout, damaging the process. Alternatively if it is too long, the system becomes food limited. SRT is same as HRT when there is no solids recycle (Vesilind, 1998). A longer SRT stabilizes the overall process, lowers the amount of effluent produced, and also increases biogas production (Rittmann & McCarty, 2001). Generally, SRT higher than 20 days is needed (Metcalf and Eddy, 2003). High SRT has higher organic load removal capacity, reduced digester

volume, shock loads resistance and microorganism acclimation to toxic compounds. High SRT can be achieved either by increasing the digester volume or by increasing the bacterial population (Gerardi, 2003).

2.5.1.3 Temperature

Temperature is one of the major parameters that affects the methane production process. According to the temperature they operate, bacteria are generally divided into two groups, mesophilic (30°C to 35°C) and thermophilic (50°C to 60°C). Since most of the methanogens are mesophilic, mesophilic AD is the most widely used (Deublein and Steinhauser, 2011). In a mesophilic digester, maximum conversion is considered to occur at 35°C. When temperature decreases 11°C, the production of biogas will fall by 50% (Zhang, 2012). In general bacterial growth rates double for each 10°C rise in temperature over a temperature range.

2.5.1.4 pH

Anaerobic processes are extremely sensitive to pH changes. A neutral pH value is highly preferred. If the pH drops below 6.8, the methanogenic activity is inhibited, and pH values below 6.2 are toxic. Generally acidogens prefer pH 5.5-6.5; methanogens prefer 7.8-8.2. If both species coexist, the preferred pH range is 6.8-7.5 (Khanal, 2008). When acidogens and methanogenic bacteria have reached equilibrium, the pH will naturally stabilize around 6.8 to 7.2.

2.5.1.5 Moisture content

Moisture content is also an important factor in the rate of waste degradation. It has many functions in the degradation process: it dissolves metabolite and influences enzyme and nutrient transport to microorganisms and the accessibility of bacteria to substrate surfaces. Many studies have confirmed that methane generation rate increases as substrate moisture content increases, up to a point (Barlaz et al., 1990; Filipkowska et al., 2004; Chan et al., 2002; Mehta et al., 2002; Wreford et al., 2000). If the moisture content is too high, transport of gases such as methane is inhibited, which negatively impacts gas production.

According to Filipkowska et al. (2004), minimal water content in wastes, at which fermentation starts, is about 25%, and optimal moisture for efficient landfill stabilization ranged from 60 to 70%. Results also confirmed that waste moisture that is too high inhibits the fermentation process.

2.5.1.6 Organic Loading Rate

OLR is defined as the amount of BOD or COD applied to the reactor volume per day (Tchobanoglous et al., 2003). Organic loading rate is related to hydraulic retention time by the following equation:

$$\text{OLR} = (Q) * (C_{vs}) / \text{Volume of reactor} \quad (2-14)$$

Where OLR = Organic loading rate

Q = volumetric flow rate (m³/d)

C_{vs} = concentration volatile solids (kg VS/m³)

V_{reactor} = reactor volume (m³)

HRT = hydraulic retention time.

For high-rate anaerobic digestion, the recommended organic loading is 1.6- 4.8 kg VSS/(m³*d) (Rittmann & McCarty, 2001). If the organic loading rate in the digester is too high, the two methanogenesis pathways can be inhibited, resulting in the formation of volatile fatty acids in the reactor.

2.5.1.7 Mixing

Mixing is another very important parameter that affects the performance of the digester. Mixing increases the rate kinetics of anaerobic digestion, accelerating the overall biological conversion process. Mixing allows uniform heating of the reactor and can be done mechanically through motorized impellers or turbines within the reactor or pneumatically by injecting gas (in anaerobic digestion, methane and carbon dioxide gas) via spargers at the bottom of the reactor (Tchobanoglous et al., 2003).

2.5.1.8 Concentration of toxic substances:

The microbial community could be disturbed if some toxic substances are introduced into the reactor, in addition to pH and temperature considerations. Obviously, oxygen has to be kept out of the bioreactor (in its molecular form O₂). The presence of a toxic substance does not mean that the process cannot operate.

Some toxic compounds inhibit anaerobic methanogenic reaction rates, but with a diverse microbial population and low enough loading, the process can be sustained. Acclimatization to toxic concentrations is also possible. Some toxic and inhibitory compounds are ammonia, calcium magnesium, metals, sodium chlorides, sulfate etc.

2.5.1.9 Nutrients

Microbes need nutrient to grow. The AD process runs best with C/N ratio between 15:1 and 30:1 (optimally 20:1). If waste does not meet the requirement, nutrient imbalance occurs so additional adjustment is needed. Crop residues or leaves which contain high carbon can be added to improve the digester performance.

2.5.2 *Importance of Biogas in Developing Countries*

Energy poverty is one of the greatest challenges these days, especially for developing countries. Globally over 2.6 billion people (38% of global population) are without clean cooking facilities (IEA, 2015). A sustainable renewable source of energy is needed to substitute for biomass sources that do not burn cleanly, such as cow dung. The importance of biogas for the less developed countries is discussed below.

2.5.2.1 Indoor Air Pollution

About half the world's population and up to 90% of rural households in developing countries still rely on unprocessed biomass fuels such as wood, dung cakes, crop residues (World Resources Institute, 1998). Because of their involvement in cooking, women's exposure is much higher than men's (Behera et al., 1988). Especially young children are often with their mom, carried on their mothers' backs while cooking, and thus spending many hours breathing in indoor smoke (Albalak, 1997). Indoor air pollution is a critical public health problem in developing countries. Particulate matter released by the burning of wood, coal, animal dung cakes, hay is a primary public health concern because of its ability to affect the upper airways of the respiratory system (Mihelcic et al., 2009).

Figure 2-12, a & b shows the traditional cooking stove and the biogas stove with clean energy (Nepal). Using traditional fuel wood stove under conditions of limited ventilation, leads to high exposures to

indoor smoke and large associated health risks, particularly for women and children. Indoor smoke from using fuel wood contains a range of potentially harmful substances, from carcinogens to small particulate matter, all of which cause damage to the lungs. Worldwide the Indoor smoke causes about 21% of lower respiratory infection deaths, 35% of chronic obstructive pulmonary deaths and about 3% of lung cancer deaths. About 64% of these deaths occur in low-income countries, especially in South-East Asia and Africa (WHO 2009).



Figure 2- 12 a. Using traditional fuel wood.

b. Using biogas for cooking

2.5.2.2 Deforestation Caused by Fuel Wood Use

Agricultural expansion and mechanization, the growth of grazing operations, mining, and fuel collection are the main causes of deforestation worldwide (Douglas & Simula, 2010). Deforestation is a very big problem in developing countries. Most of the rural areas of those countries depend on fuel-wood for cooking and lighting, which ultimately requires cutting of trees. Deforestation also leads to a decrease in the fertility of land by soil erosion. Household anaerobic digestion addresses unsustainable deforestation by providing an alternative cooking fuel, biogas, instead of traditional cooking fuel such as firewood. After successful introduction of household biogas in Nepal (one of the developing countries), wood fuel consumption was observed to decrease by 53%, with each household saving about 250 kg of firewood per

month, or 3 tons of firewood per year (Katuwal & Bohara, 2009). Figure 2-13, a & b shows the collection of firewood for energy source in a rural area of Nepal.



Figure 2- 13 a. Women collecting firewood (Nepal) b. A year supply of firewood for a family

2.5.2.3 Empowerment of Women

In developing countries energy poverty is also directly related to gender issues. In rural communities, women and young girls are typically responsible for collecting fuelwood for household uses such as cooking and heating. They have to spend several hours for fetching and collecting heavy fuel wood loads, causing several health problems and restricting them from other important productive, social and educational activities (Clancy et al., 2002). Moreover, burning of traditional biomass fuels such as firewood, coal and dung cakes, and causes emission of harmful gases, associated with health issues ranging from mild respiratory illnesses to lung cancer, with infants, children and pregnant women being the most affected (Ezzati, 2005).

As mentioned above, women and girls do most of the cooking and therefore are more exposure to indoor rural air pollution in comparison with men. 40- 45% of the Chronic Obstructive Pulmonary Disease experienced by women in developing countries is caused by indoor air pollution by the use of biomass as cooking fuel (Mihelcic et al., 2009). To promote gender equity and empower women is one of the Millennium

Development Goals (MDG's). Anaerobic digestion technology can empower women by saving their time so they can be involved in social and educational activities.

2.5.2.4 Energy Production and an Alternative Cooking Fuel: Methane

An energy produced from anaerobic digestion in the form of biogas can be used as a clean- cooking fuel as well as for lighting. Since rural peoples in developing countries depend on agriculture, every household has at least one head of cow/buffalo for milk and manure. The anaerobic decomposition of waste products, human excreta and cattle manure is a cheap way of getting energy and also at the same time handling household waste products (Gautam et al., 2009). Dung cakes, crop residues, firewood, and charcoal are all locally-producible fuel sources. LPG and natural gas require transportation; by the time it reaches consumers in rural areas, the fuel cost will be several times more than its original price. Electricity is often not reliable in such developing countries, and does not come from sustainable sources. So in such countries, biogas, an eco-friendly substitute for energy, is very important and essential.

2.5.2.5 Treatment of human / animal waste and uses of slurry as fertilizer

Runoff of animal and human waste into streams and other water bodies adversely affects the surface water quality. Also waste runoff creates contamination problems for communities living downstream. Sanitation hygiene of the household increases with the installation of toilet-attached biogas plants. According to Sasse (1988), an effluent from one kilogram of digested dung can yield up to an extra 0.5 kg nitrogen compared to fresh manure, and also an estimated N: P: K content in the effluent is 2.7:1.9:2.2, respectively. Small scale household biogas is very important for treating the human and animal wastes, improving sanitation hygiene and utilization of bio-slurry as fertilizer.

2.5.2.6 Mitigation of methane release

The biogas plants of sizes 4, 6 and 8 cubic meter mitigates about 3, 4 and 5 tons of carbon dioxide per biogas plant per year in rural areas (Shrestha et al., 2003). A biogas plant of size of 6 cubic meters can displace three tons of fuelwood, which is the equivalent of 38 liters of kerosene annually, and reduces 4.9

tons of carbon dioxide equivalents per year (Devkota, 2007). So small scale household size biogas plants help to reduce methane emissions to the atmosphere.

2.5.3 *Biogas around the world (Source: World Bioenergy Association 2015)*

2.5.3.1 Experience with domestic biogas technology in developing countries

The implementation of biogas plants has taken place in countries where governments have been involved in the subsidy, planning, design, construction, operation and maintenance of biogas plants around the world. Several countries in Asia, especially China and India, are the largest biogas countries, having effectively popularized the biogas technology. China has the highest number of household biogas plants installed in rural areas. According to the World Bio Energy Association, 26.5 million biogas plants had been installed by 2007, whose biogas yield reached 10.5 billion m³ (equivalent to more than 100 million tons of standard coal) (Chen et al., 2010). China and India produced 2.8 million and 150,000 biogas plants respectively in 2011, arriving at the cumulative numbers of 42.8 million and 4.5 million units installed of all sizes. According to Gautam et al. (2009), it is estimated that more than 111,000 biogas plants have been installed in Nepal. The Netherlands Development Organization SNV supports national programs for domestic biogas plants for households in developing countries, including countries in Asia such as Nepal, Vietnam, Bangladesh, Bhutan, Cambodia, Lao PDR, Pakistan and Indonesia; and countries in Africa such as Rwanda, Senegal, Burkina Faso, Ethiopia, Tanzania, Uganda, Kenya, Benin and Cameroon. SNV installed more than 475,000 plants in the first half of 2012. Financial support was provided by several national and international organizations.

2.5.3.2 The United States

The U.S. has more than 2,200 sites producing biogas, 239 anaerobic digesters on farms, 1241 anaerobic digesters at wastewater treatment plants (860 currently use the biogas they produce) and 636 landfill gas projects. In comparison, Europe has over 10,000 currently operating digesters; some communities are fossil fuel free because of biogas they produce (ABC 2014)

In 2011, there were about 180 operational biogas recovery systems on American commercial livestock farms, which produced enough electricity to power the equivalent of 47,000 homes (EPA, 2010). The AgSTAR program of EPA reported in 2010, about 8,000 U.S. farms could support biogas systems providing about 1,600 megawatts of energy, reducing 1.8 million metric tons of methane emissions, which is the equivalent of taking 6.5 million cars off the road.

The US methane potential from landfill waste, animal manure, wastewater, and industrial, institutional, and commercial organic is estimated at about 7.9 million tons per year, which is equivalent to about 420 billion cubic feet or 431 trillion Btu (NREL 2013). Table 2-8 shows the estimated methane generation potential of selected sources in the United States.

Table 2- 8 Methane Generation Potential in United States (NREL. 2013)

Source	Methane Potential (tons/yr)
Wastewater	2,339,339
Landfills*	2,454,974
Animal manure	1,905,253
IIC organic waste	1,157,883
Total	7,857,449

The following map (Figure 2-14) illustrates the methane generation potential by county from the following biogas sources such as landfills; animal manure; wastewater treatment; and industrial, institutional, and commercial organic waste (NREL, 2014).

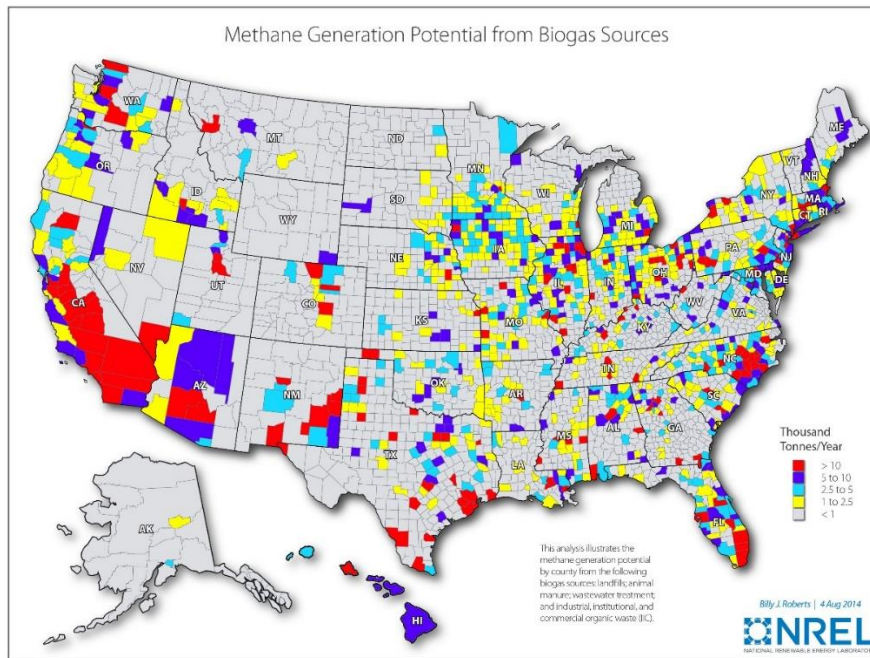


Figure 2- 14 Estimated methane generation potential for select biogas sources by county (NREL, 2014)

2.5.3.3 Europe

The biogas sectors are usually linked with agriculture in the European Union countries. In Germany, Denmark, Austria and Sweden, agricultural biogas plants are the most developed; in the countries like Portugal, Greece and some Eastern European countries, biogas technology is currently under development (Holm-Nielsen et al., 2009). In 2010 the European Union produced 21.1 billion m³ of biogas, corresponding to 12.7 billion m³ of biomethane (WBA 2013).

2.5.3.4 Germany: Industrial scale

One of reasons that Germany has succeeded in developing biogas plants is government subsidy programs. Since the Renewable Energy Sources Act (REEG) was enacted in 2000, application of biogas technology has significantly increased in Germany (Weiland, 2003). Germany is Europe's biggest biogas producer and is leading the market of biogas technology. The total electricity produced by biogas in 2012 was 20 TWh, which is equal to the supply of 5.7 million houses with electricity. In Germany biogas provides more than 3% of the whole electricity consumption (WBA 2013). About 95% of all biogas plants are at farms

and the other 5% are large centralized plants, using animal manure from a group of suppliers (Weiland, 2003).

2.5.3.5 Sweden: World leader in the use of biogas for transport (source: WBA Factsheet, 2013)

Sweden is a world leader in upgrading biogas and using it for transport. Sweden has many 'biogas vehicles', including private cars, buses, and even a biogas train. At the end of 2012, there were nearly 44,000 gas vehicles, representing a 14% increase over 2011. At the same time, the number of upgrading plants has also increased.

2.5.3.6 China (Leader in small-scale household biogas plants)

As of 2013, China had about 42 million small-scale biogas plants in operation, generating biogas for households for cooking, lighting, etc. About 60,000 small, medium and large-scale biogas plants were installed for industrial purposes. Total biogas generated in 2010 is estimated at 15 billion cubic meters, which is equivalent to 9 billion cubic meters biomethane (WBA, 2013).

2.5.4 *Benefits of Bio-Gas*

Biogas plants provide several benefits at the household, local, national and international levels. These benefits can be classified according to their impact on energy security, employment, environment and poverty.

2.5.4.1 Environmental benefits

Biogas is a complete and mature technology. Anaerobic digestion reduces the volume of the wastes going to the landfills and problems associated with their disposal. Biogas production using household waste results in reduced contamination of groundwater, surface water, and other resources. In the anaerobic digestion process, harmful pathogens can be destroyed completely. Nutrient rich by-products (effluent) from biogas digesters can serve as high-quality organic fertilizer, displacing import of synthetic nitrogenous fertilizers.

2.5.4.2 Impact on the greenhouse effect

Production of sustainable biogas plant can significantly reduce greenhouse gas emissions. Annually, worldwide 30 million tons of methane emissions are generated from the different animal waste

management systems like solid storage, anaerobic lagoon, liquid/slurry storage, and pastures. These emissions could be cut in half through anaerobic treatment (WBA 2013). It is estimated that through anaerobic treatment of animal waste and energy use, about 1324 million tons of CH₄ emissions can be avoided worldwide per year (Cassada et al., 1990).

2.5.4.3 Economic and social benefits

Increased employment: promoting the production of biogas from household organic wastes helps to create permanent jobs in local and regional development.

Sustainable energy resources: the development of biogas plant represents an important step away from dependence on fossil fuels, at the same time contributing to the development of a sustainable energy supply and enhanced energy security in the long-term.

Decentralized energy generation: Biogas technology can be established locally, using local materials and local human resources without import of raw any materials.

Sustainable waste management: Utilizing organic wastes reduces the amount of wastes that must be taken care of in another way, for example by combustion or transport to landfills.

2.5.5 *Types of biogas plants*

There are several types of biogas plant according to their design and uses such as Fixed Dome Biogas Plants, Floating Drum Plants, Low-Cost Polyethylene Tube Digesters, Balloon Plants, Horizontal Plants, Earth-Pit Plants, and Ferro-Cement Plants. The following are three basic types of biogas plants that are mostly used all over the world.

2.5.5.1 Fixed dome type biogas plant

In a fixed dome digester, the gas holder and the digester are combined (Absar, 2015). Gas is stored in the upper part of the digester. The pressure inside the digester varies as the amount of gas collected. A fixed dome digester is usually constructed of masonry and built below the ground level, protecting it from physical damage, saving space and providing insulation, which makes it suitable for cold regions (GTZ / GIZ, 1999). Advantages of this type of digesters are: the digesters have no moving parts, no steel is needed, no rusting occurs and hence a long life of the plant (20 years or more) can be expected (GTZ / GIZ, 1999).

It can be built by using locally available materials and hence its construction costs are low. The disadvantages are that special sealants are required and high technical skills are needed for construction; otherwise, plants may not be gas-tight (porosity and cracks), which causes complication of gas use (GTZ / GIZ, 1999). Figure 2-15 shows the typical fixed dome biogas reactors used in a biogas plant.

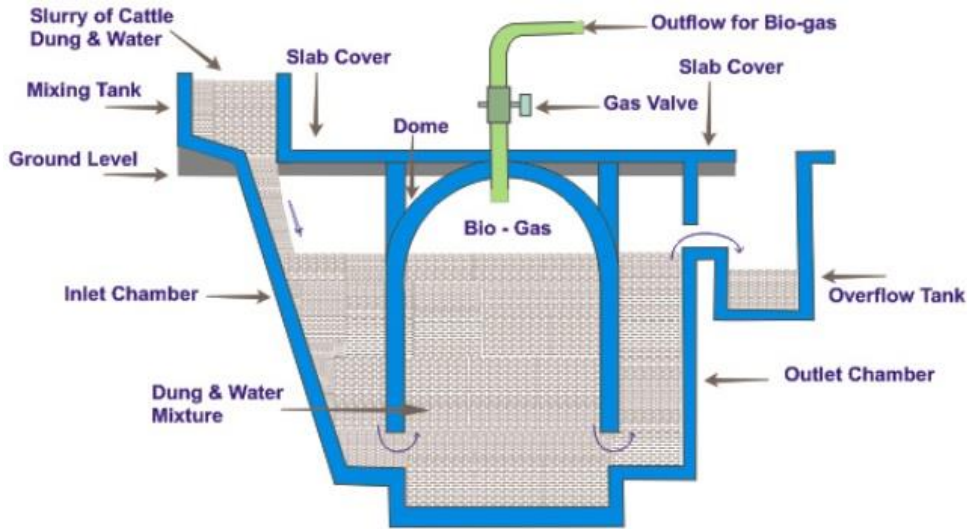


Figure 2- 15 Fixed dome type biogas plant (Biogas Technology, 2011)

2.5.5.2 Floating gas holder type biogas plant.

A floating gas holder type biogas plant consists of an underground brick masonry digester connected with an inlet and outlet, covered by a floating steel gas holder. Depending upon accumulation and discharge of gas, the gasholder moves up and down. This type of construction is expensive compared to the fixed dome type biogas plant; therefore, its use is usually restricted to large-scale sewage treatment plants. When the pressure in the holder rises accordingly, the generated gas is let out through the gas supply pipe. Alternatively, when the pressure is decreased, the holder lowers to stop the supply of the biogas. Floating-drum digesters are easy to operate and easy to maintain in terms of gas tightness by removing rust and repainting regularly (GTZ / GIZ, 1999). The design life of this type of digester is 5-15 years (Laurel, 2011). Figure 2-16 shows the floating dome type biogas digester.

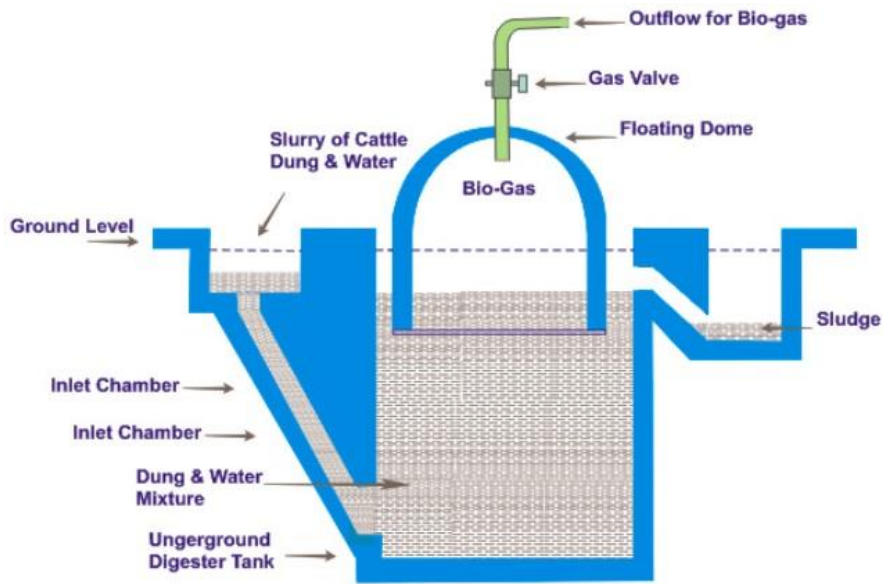


Figure 2- 16 Floating dome type biogas plant (Biogas Technology, 2011)

2.5.5.3 Fixed dome with expansion chamber type biogas plant:

The shape of this type of biogas plant has a curved bottom and hemispherical top which are joined at their bases without a cylindrical portion in between (Absar, 2015). Displaced slurry following digestion moves to the outlet displacement chamber, as there is no displacement space on the inlet side. An inlet pipe connects the mixing tank with the digester as shown in Figure 2-17. This type of biogas plant is very cheap compared with the other two types of biogas plants (Absar, 2015).

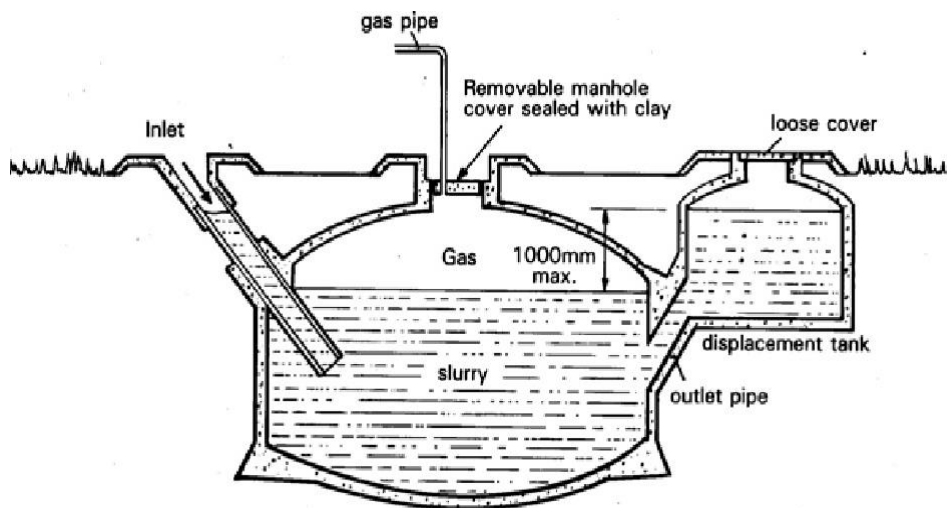


Figure 2- 17 Fixed dome with expansion chamber type biogas plant (Biogas Technology, 2011)

2.6 Inoculum

An inoculum is a source of materials used for inoculation. For anaerobic digesters, the inoculum is used to provide the source of methanogens and other microbes that degrade waste. Inoculum biomass is important not only to start up the process but also for the overall performance of the reactors. For the better performance of the reactor, the microbial loading rate must be high enough. Methanogens are abundant in anaerobic freshwater such as swamps; the stomachs of ruminants such as cow, pig, goat; and sewage sludge. Methane is produced naturally by the anaerobic degradation of organic matter, also called swamp gas. Finding new organisms that can effectively convert biomass into biomethane is an active area of research.

2.6.1 *Inoculum used for anaerobic digesters*

The sources of inocula used by many researchers are anaerobic digesters treating municipal wastewater sludge and manure. The inocula from different sources are normally used under the same operating conditions such as mesophilic temperature and around neutral pH. The respective microbial communities vary in many aspects like profiles of extracellular enzymes, species profile, biofilm-forming behavior, nutritional requirements, and physiological characteristics (Jensen et al., 2009). The inocula from anaerobic digesters treating municipal wastewater sludge are expected to have diverse groups of active

microorganisms and being suitable for many different sources of microorganisms. Mateescu et al. (2011) found that sewage sludge inocula produced greater methane than cattle dung.

Several studies have compared the impact of various kinds of dung as seed. Gopi et al. (2015) compared methane production of five kinds of animal manure as waste and inocula (both) using batch reactors operated for 90 days. Horse dung had the lowest methane production (155 ml/g VS), whereas swine manure had the highest (323 ml/g VS). Olowoyeye (2013) compared biogas production using six different animal dungs and found that sheep dung has the highest potential for production of methane, and horse dung has the lowest methane production.

2.6.2 *Effect of inoculum source and quantity.*

In an anaerobic digester, inocula play an important role during startup by balancing the populations of syntrobacter and methanogens, making syntrophic metabolism thermodynamically feasible in anaerobic digestion. The source of inocula not only affects the amount of biogas production but also influences the kinetics of the process of anaerobic digestion (Shah et al, 2014; Li et al., 2011). In anaerobic biodegradability of solid waste, the usage of a highly active anaerobic inocula or animal inoculum waste will reduce experimental time significantly, reduce the amount of inocula required in full-scale batch reactors and also consequently reduce corresponding digester volume (Obaja et al., 2003; Neves et al., 2004).

The performance of biogas production particularly from lignocellulosic biomass is greatly affected by microbial activities and chemical composition of inoculum's consortia (Griffin et al., 1998). In overcoming the acidification, the selection of inocula is crucial and plays an important role to start up a full-scale digester. If an active inoculum is selected, failure of overall the process can be avoided during the initial/start-up phase (Li et al., 2011).

A higher percentage of inoculum has been found to give a higher rate of biogas production (Forster et al., 2008). The amount of biogas produced is proportional to the initial inoculum added (Castillo, et el 1995). The selection of waste/inocula ratio is crucial, as well as the assessment of anaerobic biodegradability of solid wastes (Lopes et al., 2004). The waste/inocula ratios and wetting procedures

proposed by several researchers are different. The inocula/waste ratio for acidogenic fermentation of organic wastes is approximately 30% (Sans et al., 1995).

2.6.3 *Inoculum to be used in this study*

As discussed above, inoculum source is one of the important operational parameters for the successful operation of a digester. Without proper inoculum source, it may take long for the methane to be formed, or little or none may be formed. Previous studies, as mentioned, have found sewage sludge to be an effective inocula. ***However, one of the most important applications of biogas systems is in rural areas of low-income countries, where clean-burning fuel for cooking can be produced from degradation of household organic wastes. Most developing countries do not have wastewater treatment facilities in the rural areas, so using sewage sludge as a seed for an anaerobic digester is not an option.*** Dungs from various animals can be easily available, and have been examined as inocula in previous studies. ***Pond bottom soil is also a readily available source of inocula, and has not been examined in previous digester studies.***

Methanogens are common in nature at the bottom of stagnated ponds, lakes, and swamps or wetlands, where they produce methane, which is also known as swamp gas or marsh gas, in anoxic conditions. The pond bottom soil and the accumulated sediments are integral parts of ponds. Concentrations of nutrients, organic matter and microorganisms in the pond bottom are several orders of magnitude greater than in the water. The amounts of nutrients in a 1-cm layer of the pond bottom are normally about 10 or more times higher than the equivalent amounts in a 1-m deep water column. The bottom becomes a favorable site for microbial development due to the availability of organic matter. Methanogens in pond bottom soil in particular are important to ponds ecosystems because they remove excess hydrogen and make use of fermentation products that have been formed by other forms of anaerobic respiration. Munsiri et al. (1996) reported a higher concentration of nutrients in bottom soils of older compared to newer ponds. It was found that the bacteria count increased with the nutrient concentrations and with smaller sediment grain size (Burford et al., 1998).

The **overall goal** of this study was thus to compare the effectiveness of inocula readily-available in developing countries for anaerobic reactors treating household waste. Although horse dung has not performed as well as other types of animal dung in previous studies, it was used in this study due to its being readily available to the researcher. **Specific objectives** were:

- 1) To compare the rate of methane production and cumulative methane generation from degradation of a mixture of household organic wastes (food, paper, and yard) using 2 types of pond soil and horse dung as examples of inocula readily-available in developing countries.
- 2) To determine the effectiveness of the best inoculum from the first experiment in degrading mixtures with various proportions of food, paper and yard waste. Methane production (quantity and rate) from each type of waste will be measured.

Chapter 3

Methodology

3.1 Introduction

The first objective of this study was to compare the rate of methane production and cumulative methane generation by degradation of household wastes using horse dung and pond soil as inocula.

The second objective was applying the digestate from the first experiment (one which has a better rate of methane production) as inoculum to investigate the substrate (food waste, paper waste or yard waste) which has more potential in methane production (quantity and rate).

The typical composition of MSW includes food, paper, cardboard, plastics, textiles rubber, leather, yard, wood, glass, tin cans, aluminum, other metal, and dirt. Since most of the organic MSW typically being disposed in landfills or open dumps are food, paper (e.g., newspaper, office paper, paperboard, cardboard,) and yard wastes (e.g., leaves, grass, tree limbs), those constituents were chosen for this study (USEPA, 2007). Food, agricultural and paper wastes are the main waste components found in developing countries as well.

Three laboratory scale batch reactors were used for this study. Two sets of experiments were conducted for the biodegradation of household wastes. In experiment set "A" the three reactors were seeded with different types of inoculum: horse dung (reactor1), pond bottom soil 1 (reactor 2) and pond bottom soil 2 (reactor 3). The percentage of inoculum was 10% (by weight) for all three reactors. The reactors were filled with waste of composition 50% food, 30% yard and 10% paper by weight. This study is more focused on the rural community of developing countries. In those areas, there is no energy available and even if it is available people cannot afford that. So the people of rural community do not have facilities to save leftover food; instead it will be feed to the animals or dumped in the street corners. Thus, the amount of waste that comes from the households in rural community of developing countries is more food than yard and very low amount of papers. The waste composition for testing was chosen based on waste generated in the rural community of developing countries. One duplicate "B" run was conducted for set "A". For experiment set "C", all three reactors were inoculated with the same type of inoculum (digestate from set

“B”) with different proportions of waste mixes in each reactor. The inocula used for the “C”, set of experiments was digested waste from a reactor which was inoculated with Pond 1 which had the highest rate of methane production in the first experiments. Experiment “C” is the further testing of better-digested waste from the first experiment. To find out the effectiveness of the seeds obtained from Pond 1(B) on Food, Yard and Paper waste, the three reactors were filled with different proportion of waste compositions, so that performance of seeds on Food, Yard and Paper waste can be determine. The first reactor was filled with more Food waste (50%), second reactor was filled with more Yard waste (50%), and third reactor was filled with more Paper waste (50%), with remaining waste as shown in Table 3-1. All sets of experiments were conducted at room temperature (25 -35 °C) and the total mass of waste used was 3.06 kg for each reactor. Table 3-2 summarizes experiments conducted in present study.

Table 3- 1 Summary of the Experiments

Waste type	Experiment A and B (Duplicates)			Experiment C		
	Reactor 1	Reactor 2	Reactor 3	Reactor 1	Reactor 2	Reactor 3
Inoculum	Horse dung	Pond soil	Pond soil	Digestate	Digestate	Digestate
Inoculum %	10%	10%	10%	10%	10%	10%
Food	50%	50%	50%	50%	20%	20%
Yard	30%	30%	30%	20%	50%	20%
Paper	10%	10%	10%	20%	20%	50%

3.2 Reactor Preparation

Three laboratory scale reactors (Wheaton 356889 celstir spinner flask, 6000 ml glass double sidearm with 4mm screw caps, 258 mm*404 mm) were used for this experiment. Previously used reactors were cleaned by using soap and tap water before use them. Tygon tubes, two way valves and connections were leak checked before use. Transparent sealant was applied as necessary and dried for 6-8 hours. Figures 3-1 and 3-2 show the reactor and the tygon tubes used for this experiment, respectively.



Figure 3- 1 Glass reactor used for this experiment



Figure 3-2 Tygon tubes with two-way valve

Six-layered aluminized gas sample collection bags with storage volume of 22 L (Cali 5-Bond Bag, Calibrated Instruments, Inc.) were used to collect the biogas generated from the reactor. As shown in Figure 3-3, gas sampling bags were installed for each reactor to collect the gas produced from the reactors.



Figure 3-3 Gas-bag used to collect the generated biogas

3.3 Inoculum Collection

The inoculums used and tested for this experiment were horse dung and ponds soils. Figure 3-5 shows the inocula used for this study. The horse dung was collected from outdoor loafing sheds in Keller, Texas. The dung had been stored on site in static piles for approximately one week prior to being collected for this study.

Pond bottom soils were collected from two different ponds near Keller, Texas. Pond soil 1 was obtained from a smaller pond, whereas pond soil 2 was obtained from a bigger pond almost twice the size of pond 1 (Figure 3-4, a and b). Soil from the bottom of ponds was collected by using a scoop and small bucket. Ponds near the author's home were chosen, to simulate what persons in developing countries

would do, which is obtain soil from the nearest pond, because they likely do not have a car to access ponds farther away.



Figure 3- 4 a. Pond 1

b. Pond 2



Figure 3-5 Different inoculum used for this study

3.4 Waste Collection

3.4.1 *Food waste*

Food wastes such as leftover food (rice, curry, cereals, and breads, tortillas, and noodles chips) and expired food (store purchased) were collected from the kitchen of my own house. To increase the carbon-nitrogen ratio, some egg waste was also added. Collected food wastes were kept in the refrigerator until enough accumulated for the 3 reactors of the next set of experiments. Figure 3-5 shows the collected food wastes for this experiment.



Figure 3-6 Food waste

3.4.2 *Paper waste*

Paper waste was collected from a house as well as an office (office paper). The composition of papers were newspapers, mail, magazines, office papers, etc. Large pieces of paper were shredded to smaller pieces because it has been reported to increase bio-availability, which can lead to faster degradation (Buivid et al., 1981). Figure 3-7 shows shredded paper waste (newspaper, office paper).



Figure 3-7 Paper waste

3.4.3 *Yard waste*

Yard waste such grass from the lawn mowers, leaves and tree/bush trimmings was obtained from my own backyard. Vegetable leaves such as beans, squash, pumpkins, eggplant, cabbage, and carrots were collected and dried, as well as green leaves from yard trimmings (mugo pine, boxwood, oak). Large pieces of yard waste (grass, leaves) were cut in order to fit into the reactors. Larger and matured branches were removed manually; only leaves and the new/ soft stem were used. Figures 3-8 (a) and (b) show yard wastes grass and leaves, respectively.



Figure 3- 8 a. Leaves from trees and bushes trimming



b. Grass from yard and lawn mower.

All the wastes collected and used for this experiment were fresh when obtained from the individual sources, which indicates that any form of decomposition was not started. Those individual waste components (food, paper and waste) were then mixed together.

3.4.4 Methane potential of waste used, estimation of required waste and water.

Table 3-2 presents the biomethane potential of three different substrates extracted from selected literature sources (Lim, 2011; Cho et al. 1995; Eleazer et al., 1997, Jeon et al., 2007; Chynoweth et al., 1993). BMP for the same waste component can be expressed in different ways.

Table 3- 2 BioMethane Potential and Moisture Content Values for Waste Components (Literature)

Type of substrate	Biomethane potential m ³ /kg of VS in waste	g of VS/ g of dry solids	Moisture content (%)
Food	288-540	0.90	50-80
Yard	14-283	0.85	4-8
Paper	75-370	0.89	30-80

Table 3-3 shows the amount of selected waste (Food, Paper and Yard) which is calculated as per percentage value provided in table 3-1 and water needed for each reactor for first sets of experiment , A and B (duplicates) and experiment “C”.

Table 3- 3 Amount of Selected Waste and Water needed for each Reactor

Reactor	Experiment A and B (Duplicates)			
1,2 &3	Food (kg)	Yard (kg)	Paper (kg)	Water added (L) (90%)
	1.7	1.02	0.34	2.75
Experiment C				
1	1.7	0.68	0.68	2.75
2	0.68	1.7	0.68	2.75
3	0.68	0.68	1.7	2.75

According to Deublein and Steinhauser (2008), 75-90% of water is recommended with 10-25% dry matter.

In this study, the water requirement is calculated as per the following relation:

$$0.90 = M_{\text{water}} / M_{\text{TOTAL}} \quad (\text{For all the reactors 2.75 liters of water was added.})$$

Figure 3-9 (a) and (b) show the laboratory scale reactor setup (photograph) and schematic.



Figure 3- 9 (a) Photo of reactor set-up

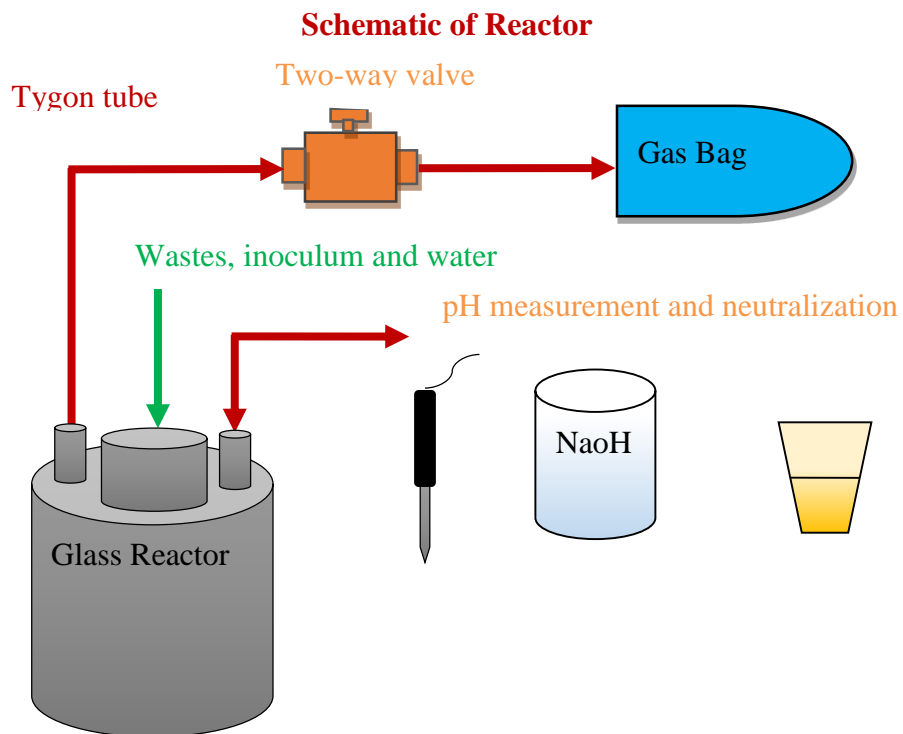


Figure (3-9) (b) Schematic of Reactor

3.5 Reactor Setup/ Procedure

The amount of required waste mass as per given in Table 3-4 was measured and mixed them together manually to homogenize the constituents. The reactors were filled with the 10 % inoculum (Pond1 Pond 2 and Horse (in duplicate reactors, "A" and "B") and digestate (in experiment "C") and then waste was added to the reactor by hand in layers and compacted by using a compacting tool. The calculated volume of tap water was added to the waste to make sure that the waste was near the saturation limit. If the water is not sufficient, it will affects acclimatization of micro-organisms to the waste composition in the reactor, which results in a longer lag period. To ensure good microbial contact and faster start-up, about 2.75 liter of water (90 %) was added to each reactor such that the waste was near the saturation limit. Then the waste and water mixture (slurry) was stirred by using a metal rod manually. The empty space of about 0.5 feet in the top portion of reactor was reserved for gas headspace.

Figures 3-10 (a) and (b) shows reactors being inoculated with microbes seed and loading of waste mixtures and compaction done by using compacting tool respectively.



Figure 3- 10 (a) Reactor inoculated with microbe's seed (b) loading of selected waste mixture

Wastes and inoculum were loaded through the main (central) opening of the reactor. One arm of the reactor was connected to the gas bag with a Tygon tube and two-way valve. The threaded cap having two holes where delivery tubes were inserted remained above the layer of the slurry. Another arm was used for pH measurement and neutralization (adding NaOH) to the slurry. After the reactors were filled, the main opening and both arms were sealed using plastic tape, threaded cap and transparent sealant in thick layers to ensure no leakage.

Figure 3-11 (a) (b) (c) and (d) shows applying sealant at the screwed cap of the central opening of digester, using thread sealed tape, and application of sealant on the joints of the delivery tube, respectively, to make sure there was no leaking.



Figure 3- 11 (a) Applying transparent sealant to the reactors



(b) Using thread sealing tape



(c) Using sealant at the T-joint of a delivery tube

Figure 3-12(a) and (b) shows the reactors after the setting up was completed (gas production phase). Figure (a) shows experiment set "A" in which reactors were inoculated with Horse dung, Pond soil 1 and Pond soil 2. Experiment set "A" has a duplicate set "B". Figure (b) shows experiment set "C", in which reactors were inoculated with digestate from a reactor which was inoculated with Pond 1.

Figure 3-12 (a) and (b) show the reactors after set up



Figure 3- 12 (a) Experiment set (A)

(b) Experiment set (C)

3.6 Measurement

3.6.1 *Gas collection and measurement (Composition and Volume)*

The amount of gas production was measured by pumping gas out of the collection bag through a standard SKC 44*R Universal Sample Pump at 1.9 L/min connected to a Calibrator (Mesa Labs Bios Defender 510 medium dry call gas flow instrument), to get a minute by minute gas pumping rate. LANDTEC GEM5000 with infrared gas analyzer was used to measure the composition of biogas produced such as % methane, (CH₄), % carbon dioxide (CO₂), % oxygen (O₂), and percentage of other gasses. A stopwatch was used to measure the time of gas flow.

Generated gas from each reactor was collected and measured on a regular basis. The frequency of gas measurement depended on the amount of the biogas generated. During the initial phase of degradation the gasses were measured once a week except the reactor inoculated with horse dung (horse dung was measured in every two weeks or more as it had long lag time and slower gas formation in its initial stages than the other reactors). As the degradation progressed, and rate of gas production increased rapidly, the gas bags were emptied every four-five days to avoid excessive buildup. When the gas production decreased, the sampling frequency was also reduced accordingly. The degradation time of waste and rate of formation of gas can be highly dependent on several factors such as waste constituents, type and percentage of inoculum used, amount of water added, pH, temperature. When the gas production from each reactor reached an asymptotic value or dropped to a low constant value, gas measurement was stopped. Figure 3-13 shows instruments used for gas composition and volume measurement for this study.



Figure 3-13 Gas composition measurement instrument Landtec GEM 5000 (middle) and volume measurement instruments, Calibrator (Left) & SKC Sampler (Right)

Figure 3-14 and Figure 3-15 show the process of measuring gas composition percentage by using the Landtec GEM 5000 infrared analyzer. While measuring the composition of gases, LANDTEC GEM5000 was connected to Mesa Labs Bios Defender so that the amount of gas released during the process can be calculated and added to the total biogas production for that reactor.



Figure 3- 14 Process of measuring gas composition



Figure 3-15 Gas flow measurement while measuring the gas composition

Figure 3-16 shows the gas production from different reactors. Gas production increased and reached a peak value and then production slowed down.



Figure 3- 16 Biogas generation from different reactors.

Figure 3-17 shows the highest methane percentage captured throughout this study. Methane percentage of 66.6% was captured on a reactor which was inoculated with pond soil 1.

View Data		05:36 - 01/22/16		24 / 41	
ID:		10/06/15 07:45:06			
CH ₄	(%)	66.6	Ini-SP	("H ₂ O)	-0.01
CO ₂	(%)	27.1	Ini-DP	("H ₂ O)	-0.001
O ₂	(%)	0.3	Ini-Temp	(°F)	0.0
H ₂ S	(ppm)	43	Ini-Flow	(SCFM)	0.0
CO	(ppm)	11	Ini-Power	(e3BTU/h)	0.0
H ₂	(LMH)	LOW	Adj-SP	("H ₂ O)	-0.01
Bal	(%)	6.1	Adj-DP	("H ₂ O)	-0.001
			Adj-Temp	(°F)	0.0
			Adj-Flow	(SCFM)	0.0
			Adj-Power	(e3BTU/h)	0.0

Figure 3- 17 Highest methane percentage captured in this study.

The volume of the collected gas in the gas sampling bag was measured by using a standard SKC 44*R Universal Sample Pump and Calibrator, Mesa Labs Bios Defender 510. The fixed rate of gas flow was measured during the sampling and the time until the gas bags were completely empty was recorded by using a stopwatch. The total gas volume was calculated by using the following relation.

$$\text{Total volume (L)} = \text{flow rate (L/min)} * \text{time required to empty the bag (min)}$$

Figure 3-18 shows the process of volume measurement by using SKC sampler and calibrator.



Figure 3- 18 Volume of gas is measured by using SKC sampler and calibrator

3.6.2 *pH measurement*

pH was measured by using HQ40d Portable pH Hach meter. The pH meter was calibrated each time before use and was rinsed and wiped after use for measuring the pH of slurry. At the initial stage, pH was measured daily. It was very essential to measure the pH during the initial stages of the reactors, because of chances of acid accumulation during the acidogenic phase (Christensen and Kjeldsen 1989). Sodium hydroxide (NaOH) addition was required in the initial startup phase of the reactor to avoid excessive acid accumulation. Since food waste was used in this study, the possibility of acid formation was higher. So the pH was maintained above 6. If the pH drops below 6, which is toxic to the methanogens, methanogenic activity is inhibited. Therefore to ensure a basic condition in the reactor and keep methanogens alive in the system, sodium hydroxide was added. 10-30 ml of NaOH was added at a time,

depending on the pH level until the pH stabilized above 6.5. Figures 3-19 and 3-20 show the instrument used to measure the pH of slurry and initial pH measurement of slurry, respectively.



Figure 3- 19 pH meter for measuring pH of waste composition.



Figure 3- 20 Initial pH measurement of slurry

The initial pH measurement of slurry was done by taking out some liquid slurry from a reactor before sealing it, which was done on the same day of the reactor set-up. The intermediate pH measurement (during gas production phase) was done by inserting the IntelliCAL™ probes into the slurry inside the reactor through one arm. pH was measured every day at the initial phase, as already mentioned, since the chances of acid accumulation are higher at the initial stage. Sodium hydroxide was added as needed to neutralize the slurry. When gas formation is started and the pH is neutralized to 6-8 to 7.2, then pH was measured once in a while (once/twice a week). After NaOH was added, to the reactor, the threaded cap on the arm was resealed by applying thread sealing tape and transparent sealant. Figure 3-21 (a), (b), and (c) show pH measurement in between the degradation process, addition of NaOH to the waste mixture and resealing of cap, respectively.



Figure 3-21 (a) pH measurement



(b) Addition of sodium hydroxide



(c) Resealing the cap of reactor

Chapter 4

Results and Discussion

In this chapter the results obtained from experiments conducted using laboratory scale reactors are presented and analyzed to evaluate the effect inoculum on the overall performance of the anaerobic reactors.

4.1 Sludge pH for duplicate reactors seeded with Pond 1, Pond 2 and Horse dung

In the initial days of the reactors, the frequency of pH measurement was higher as base needed to be added if pH dropped too low. Therefore for the first 10 days, pH was measured more frequently than the rest of the active time of the reactors. After the first 10 days, when pH stabilized about neutral, pH was measured every 1-2 weeks. The pH variation over time in all reactors for the duplicate runs are shown in Figure 4-1 (a) and (b). Figures 4-2, 4-3 and 4-4 compare pH variations with time for reactors Pond 1(A and B), Pond 2 (A and B), and Horse (A and B), respectively. The behavior of the duplicates is comparable.

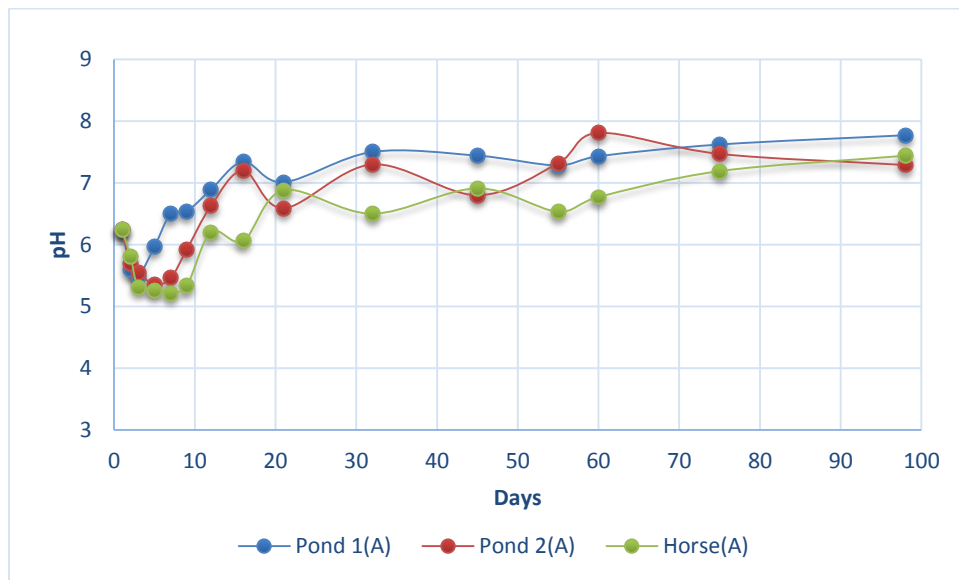
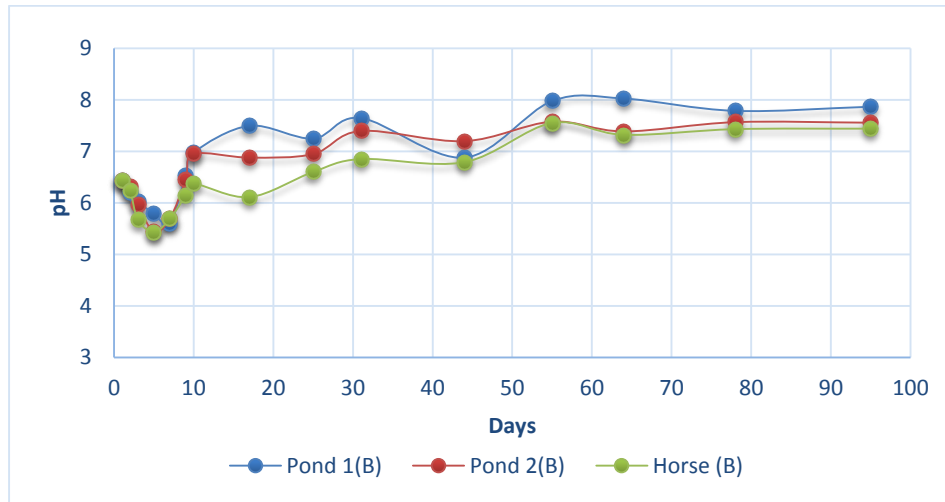


Figure 4- 1 (a) pH variation vs. time in reactors, Pond 1(A) and Pond 2(A) and Horse (A)



(b) pH variation vs. time for reactors Pond 1(B), Pond 2(B) and Horse (B)

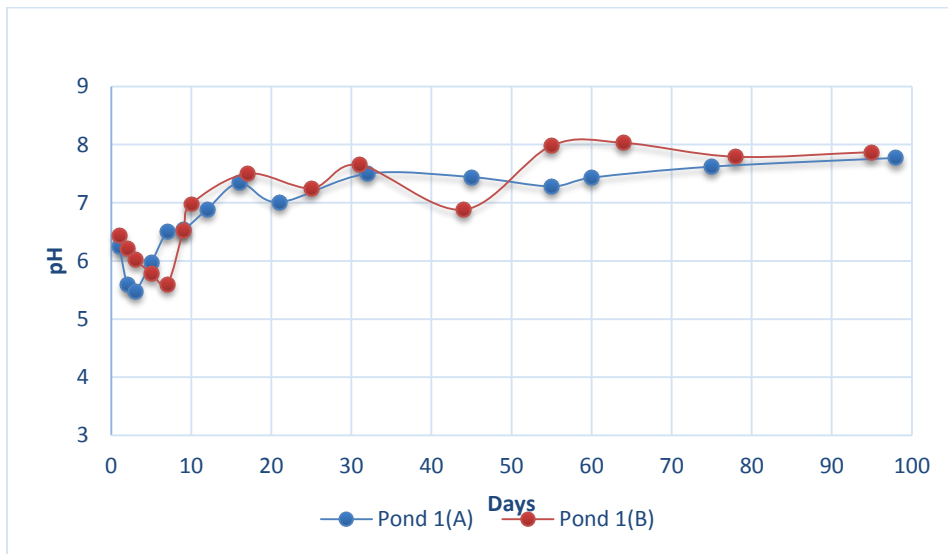


Figure 4- 2 pH variation vs. time for duplicate reactors Pond 1 (A and B)

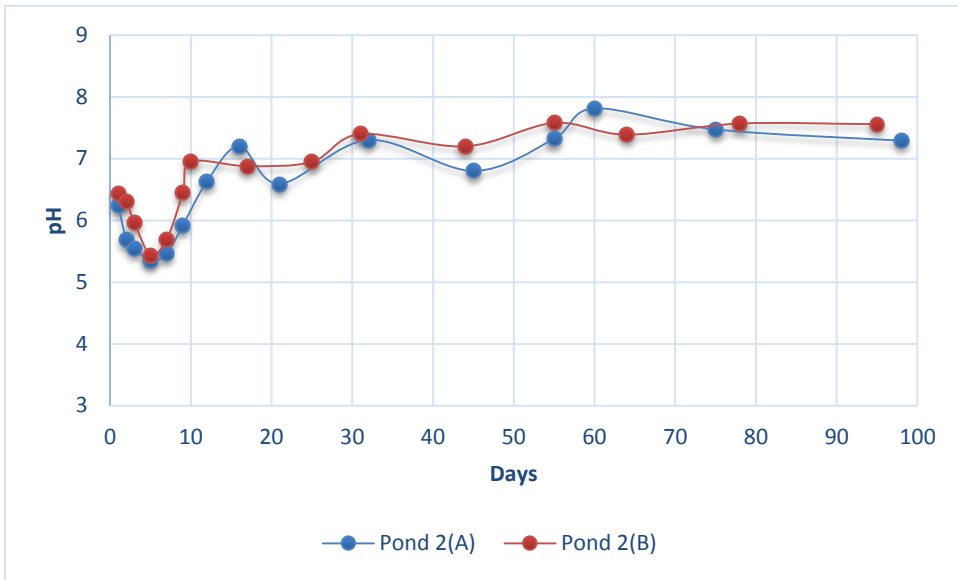


Figure 4- 3 pH variation vs. time for duplicate reactors Pond 2 (A and B)

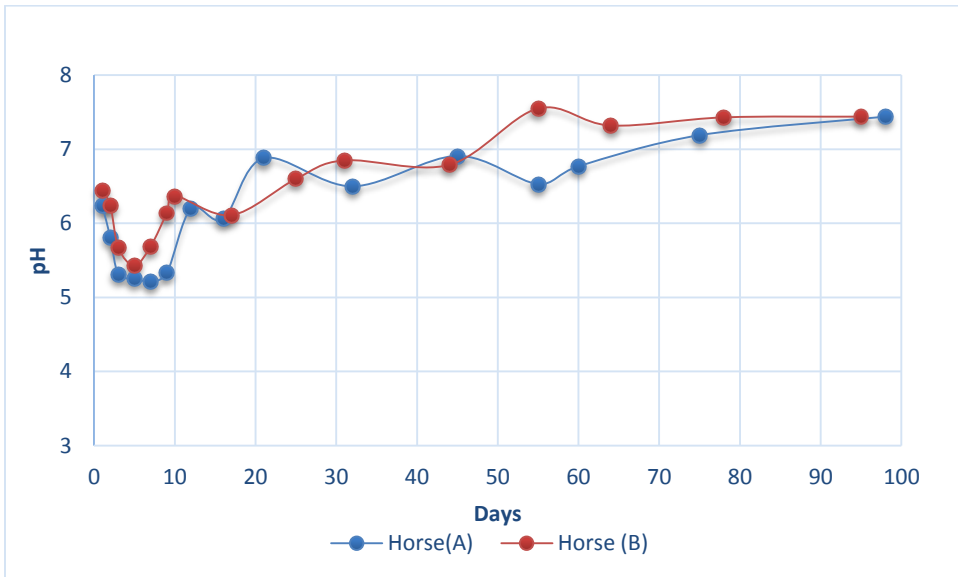


Figure 4- 4 pH variation vs. time for duplicate reactors Horse (A and B)

In the initial days, the pH level for all reactors was less than 6.5. Because of the ongoing acid accumulation state in the waste degradation process, acidic phase was existed. On the very first day, pH was 6.24 and 6.44 for all the reactors in sets (A) and (B), respectively, since all reactors were filled with the same types and proportions of the wastes.

The low pH level continued up to 7 and 9 days for pond 1 (A) and (B); 12 and 10 days for Pond 2 (A) and (B), respectively; and 21 and 17 days for Horse (A) and (B), respectively. A gradual rise in pH level was observed afterwards in all six reactors, so that values fluctuated between 6.5 and 8. This was due to the conversion of carboxylic acid into methane and carbon dioxide, which is an indication of the fourth phase of biodegradation. The maximum pH values ranged from 7.3 to 8.0 for all reactors. According to the degradation phases based on pH results, Pond 1(A), reached the methanogenesis phase quicker than other reactors in experiment A, It took only 12 days whereas in experiment B, Pond 1(B) reached in methanogenesis phase quicker than Pond 2 (B) and Horse (B). In the both runs (A and B), Pond 1 stabilizes at around neutral faster than the other two. The pH for reactor Horse took longer to stabilize than the Pond 1 and Pond 2 in the both sets.

Warith et al. (2002) recorded change in pH with time in an anaerobic bioreactor (Figure 4-5) which is in accordance with the results obtained from the reactors operated in this study. At initial stages the pH was low and then increases over time and stabilizes in between 7 and 8.

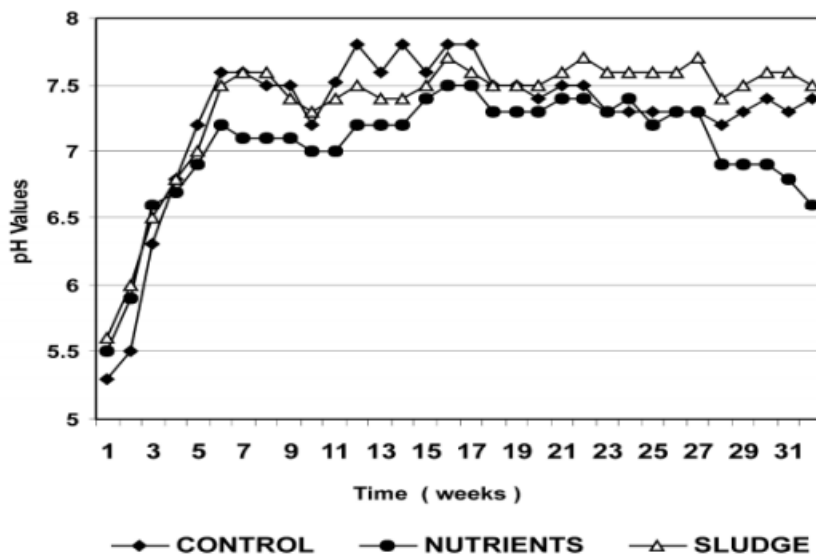


Figure 4- 5 Changes in pH of leachate with time (Warith et al., 2002)

4.2 Gas data for duplicate reactors seeded with Pond 1, Pond 2 and Horse Dung

4.2.1 Gas composition for duplicate reactors (A and B) of Pond 1, Pond 2 and Horse

Figures 4-6, 4-7, 4-8 show the composition of gases generated from the reactors Pond 1(A) and (B), Pond 2(A) and (B), and Horse (A) and (B), respectively. The duplicate generally reactors behave similarly. The carbon dioxide percent in all reactors was initially higher (around 75-85%), due to acetogenesis phase, in which volatile fatty acids, carbon dioxide and water vapor are produced. The percentage of oxygen drops quickly, leading to anaerobic conditions inside the digester. For all the reactors, the percentage of carbon dioxide decreases over time and methane increases over time, as the methanogenesis phase produces methane. For most of the reactors, carbon dioxide stabilized at 35-40%; however, some fluctuation in carbon dioxide was observed for the reactor seeded with Horse (B) after the methane peaked at day 97. For all reactors, methane stabilized at 40-60%.

In Figures 4-6 through 4-8, the “other gases” initially represents mostly molecular nitrogen present in air, which decreases over time. Other gases were less than 20 % initially and even lower (less than 10%) over time. “Other gases” can also include sulfides, disulfides, mercaptans, and ammonia generated from organic compounds containing sulfur and nitrogen, as well as molecular hydrogen and carbon monoxide.

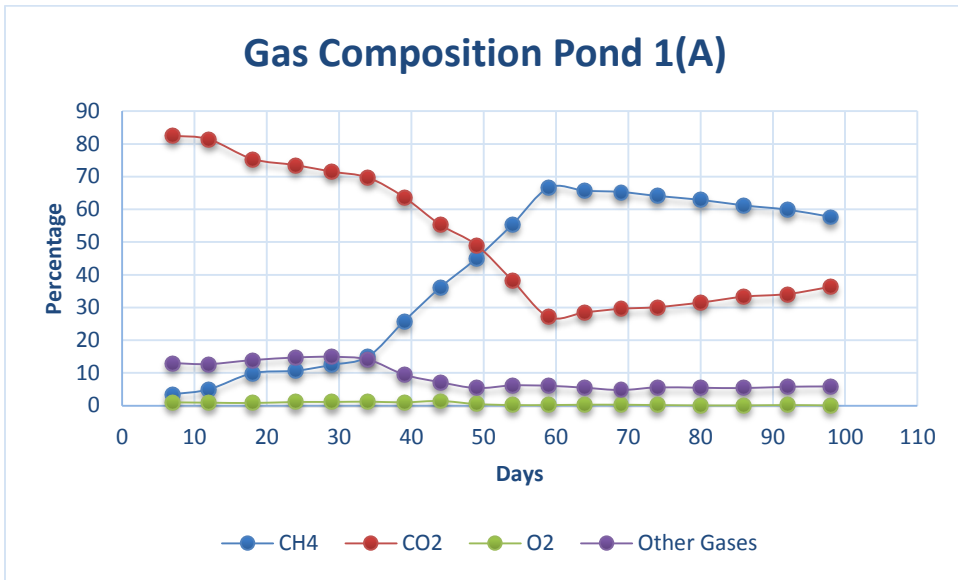


Figure 4- 6 a. Gas composition percent vs. time for Reactor Pond 1(A)

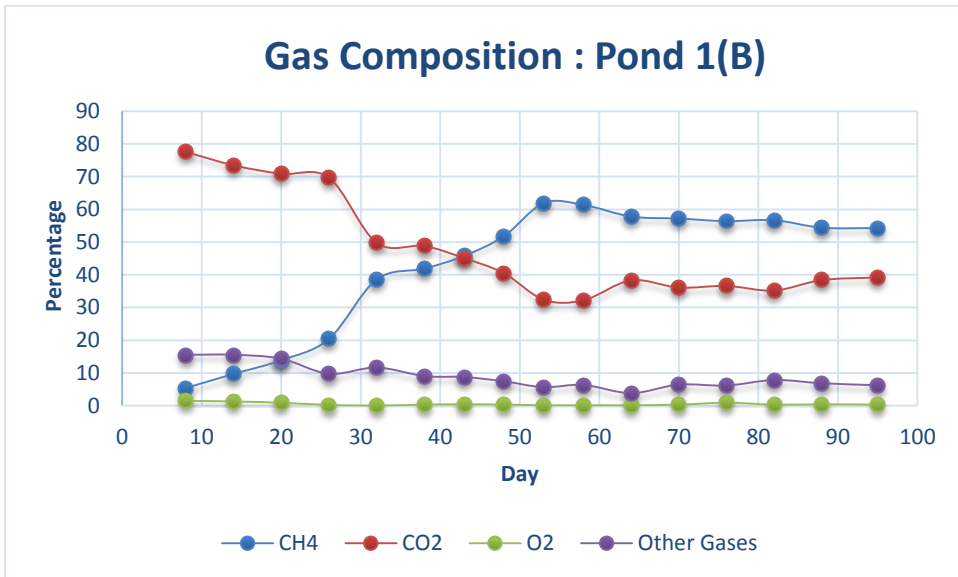


Figure 4- 6 b. Gas composition percent vs. time for Reactor Pond 1(B)

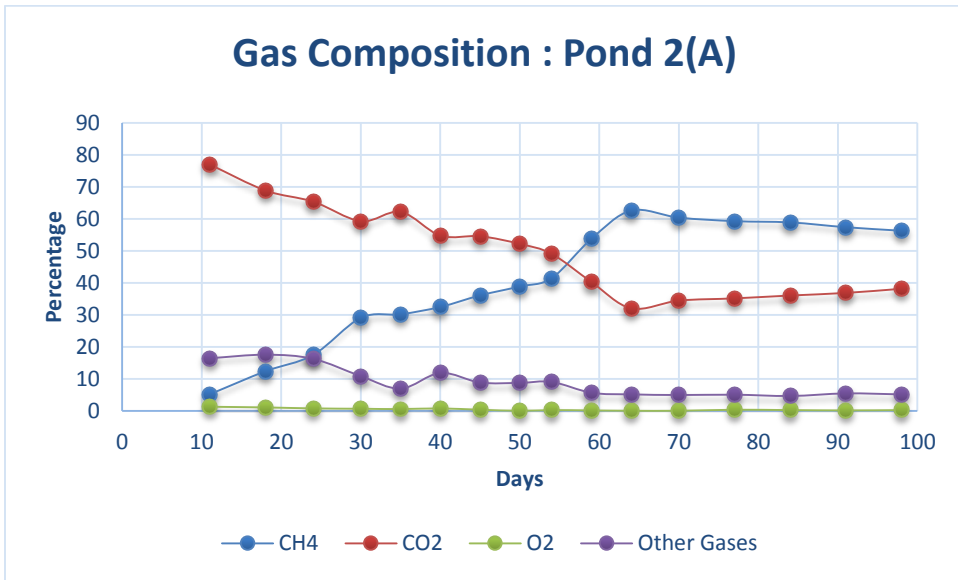


Figure 4- 7 a. Gas composition percent vs. time for Reactor Pond 2(A)

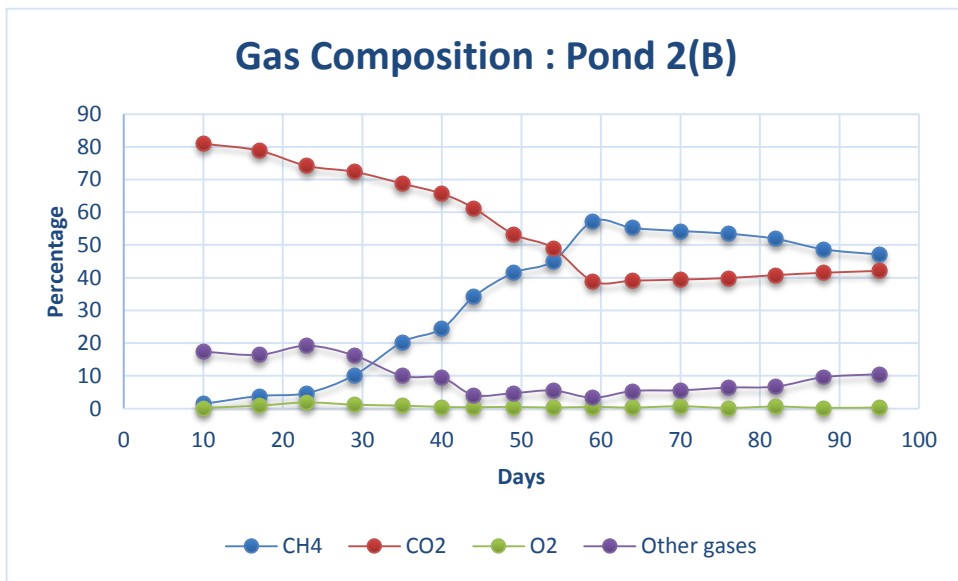


Figure 4- 7 b. Gas composition percent vs. time for Reactor Pond 2(B)

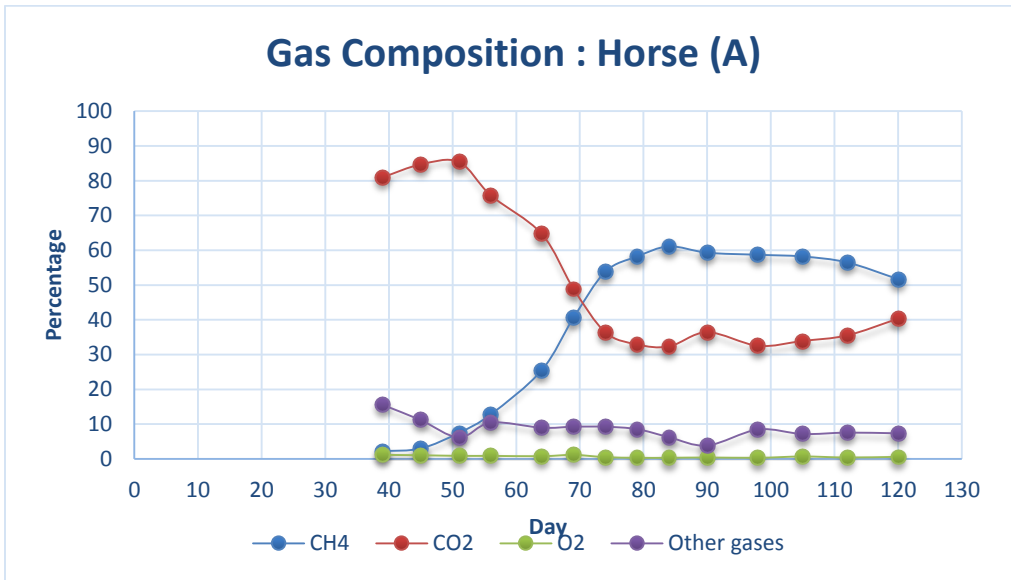


Figure 4- 8 a. Gas composition percent vs. time for Reactor Horse (A)

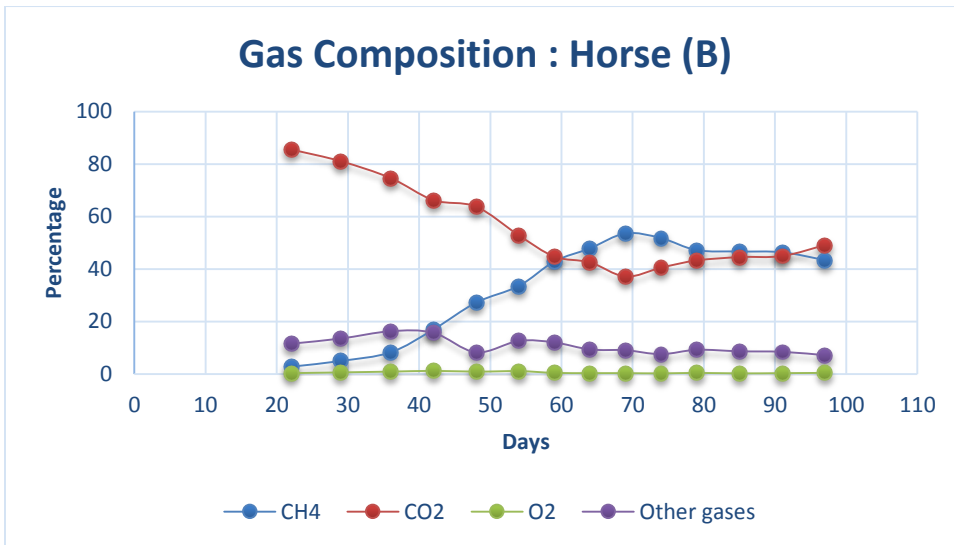


Figure 4- 8 b. Gas composition percent vs. time for reactor Horse (B)

Table 4-1 shows maximum and minimum carbon dioxide and methane percentages for each reactor, and the dates of occurrence. Maximum methane levels varied from 53% to 67% for the various reactors. For all the reactors minimum level of methane was observed at maximum level of CO₂ and vice-versa.

Table 0-1 Maximum and Minimum Methane Generation for Duplicate Reactors and Days of Occurrence

Reactor	Minimum CH ₄ %	Maximum CO ₂ %	Day of Occurrence	Maximum CH ₄ %	Minimum CO ₂ %	Day of Occurrence
Experiment A						
Pond 1(A)	3.5	82.5	7	66.6	27.1	59
Pond 2(A)	5.2	77	11	62.6	32.1	64
Horse (A)	2.2	81	39	61.1	32.3	84
Experiment B						
Pond 1(B)	5.4	77.6	8	61.9	32.4	53
Pond 2(B)	1.4	81	10	57.3	38.9	59
Horse (B)	2.7	85.5	22	53.61	37.1	69

Figure 4-9 shows the percentage methane over time for all three reactors in duplicate sets (A and B). At the very beginning, it took some time for all reactors to generate some gas. Table 4-2 shows the initial lag phase for all reactors. Pond 1(A) and (B) have the shortest lag period, likely because Pond 1 is an older pond compared to Pond 2, and thus has more stable organic matter accumulated in the bottom soil over a long time. The soil with high organic matter and nutrient concentrations provided favorable conditions for bacterial growth of different species, including methanogens in anoxic conditions, leading to the largest population of microbes that acclimated faster than Pond 2 and Horse. This hypothesis could be confirmed via tests measuring organic content of the soils and horse dung, and measures of numbers and variety of microbes. On the other hand, the Horse reactors had long lag periods but after the methane started to form, the methane percentage increased sharply. It is likely because of, less organic matter in the horse dung, leading an initial thin population of microbes; however, once the microbes acclimated to the environmental condition inside the reactors, they increase exponentially.

Similarly, methane peaks earliest for Pond 1 (A) and (B) and latest for Horse (A) and (B). The peak methane percentages for the first set of runs (A) are higher than for the second set of runs (B), for reasons that are unclear. Table 4-3 shows the average maximum methane percentage for the duplicate reactors Pond1, Pond 2 and Horse .From the both sets of reactor duplicates the average maximum methane percentage for Pond1 was higher than Pond 2, which was higher than Horse.

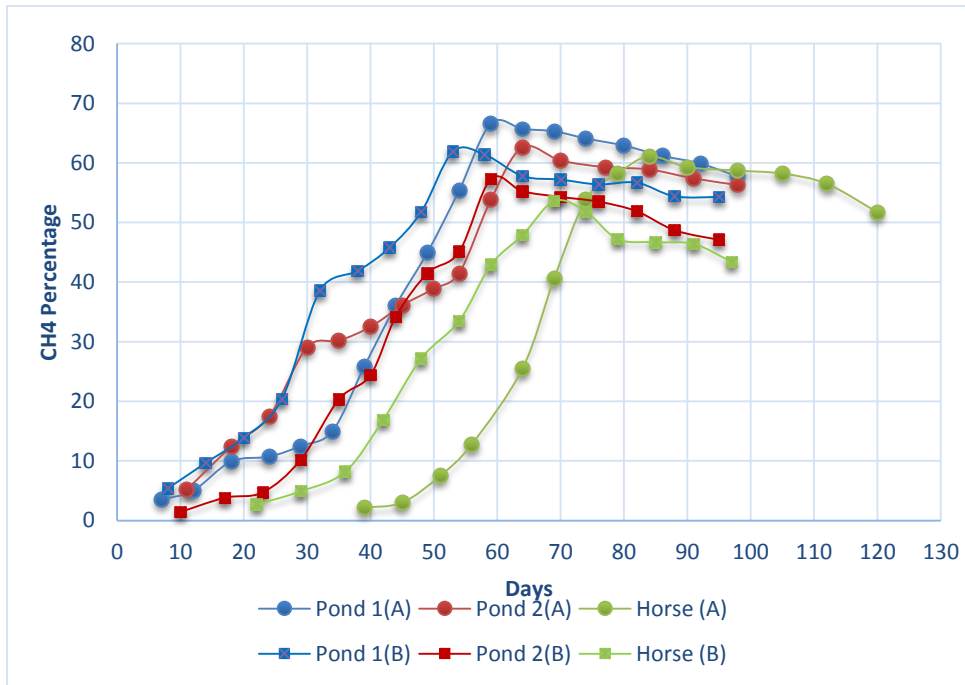


Figure 4- 9 Methane percent vs. time for duplicate reactors (A and B)

Table 0-2 Initial Lag Phase for Duplicate Reactors (A and B)

Reactor	Initial Lag Phase (Days)	
	Experiment A	Experiment B
Pond 1	7	8
Pond 2	11	10
Horse	39	22

Table 0-3 Maximum Average Methane Percentage for Duplicate Reactors

Reactor	Maximum average Methane Percentage
Pond1	64.3
Pond 2	60
Horse	57.4

Figure 4-10 shows trends of CH₄:CO₂ with time for both sets A and B. Initially the ratio of methane to carbon dioxide was very low and increased gradually over time for all reactors. Table 4-4 shows average peak methane to carbon dioxide ratios for duplicate reactors. Pond1 reactors had highest methane to carbon ratio and horse reactors had the lowest. The values peak earliest for the Pond 1 reactors and latest for the Horse reactors. The ratios then generally level off to values between 0.9 and 1.6.

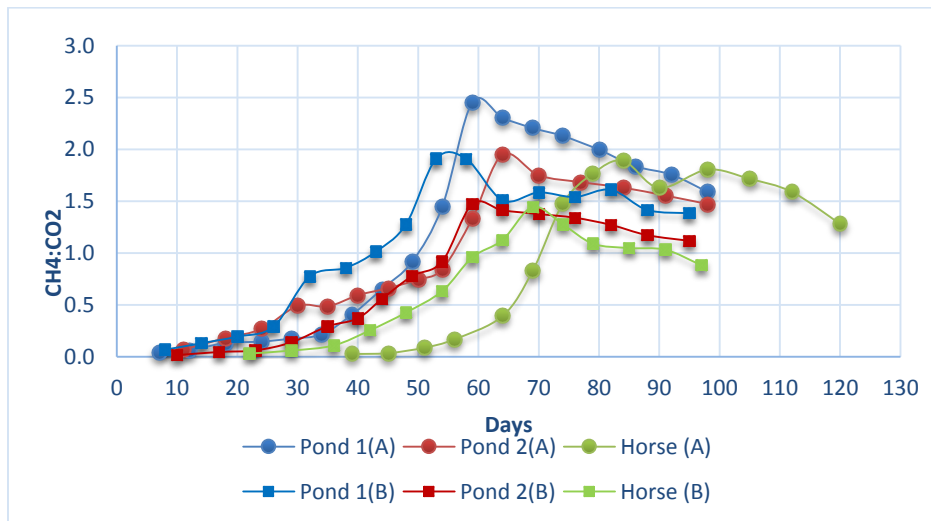


Figure 4- 10 Methane to carbon dioxide ratio vs. time for Pond 1(A &B), Pond 2(A &B), and Horse (A &B),

Table 0-4 Average Maximum of Methane to Carbon Dioxide Ratios for duplicate reactors

Reactor	Average Maximum CH ₄ :CO ₂
Pond 1	2.2
Pond 2	1.75
Horse	1.65

4.2.2 Cumulative volume and rate of methane generation

Figure 4-11 compares cumulative methane generation (liter/kg) over time for duplicate sets of reactors. Reactor duplicates generally behave similarly, except for Horse (A), which has a longer lag time than Horse (B). Figure 4-12 shows the average cumulative methane for each seed type for 95 days of reactor operation. Average cumulative methane of Pond 1 was observed highest followed by average of Pond 2. Average cumulative methane of Horse produces the lowest cumulative methane. Reactors inoculated with seeds from Pond 1 produced 19.4 % more methane than reactors seeded with Pond 2, and 72% more than the reactor seeded with Horse dung. Similarly, reactors seeded with Pond 2 generated 44% more methane than the reactor with Horse dung. So overall, Pond bottom soil as an inoculum gave the better reactor performance for producing methane using household waste.

Table 4-5 lists cumulative generation for all reactors at the end of reactor operation. It was observed that Pond 1 reactors generated the highest amount of methane and Horse reactors generated the least amount of methane.

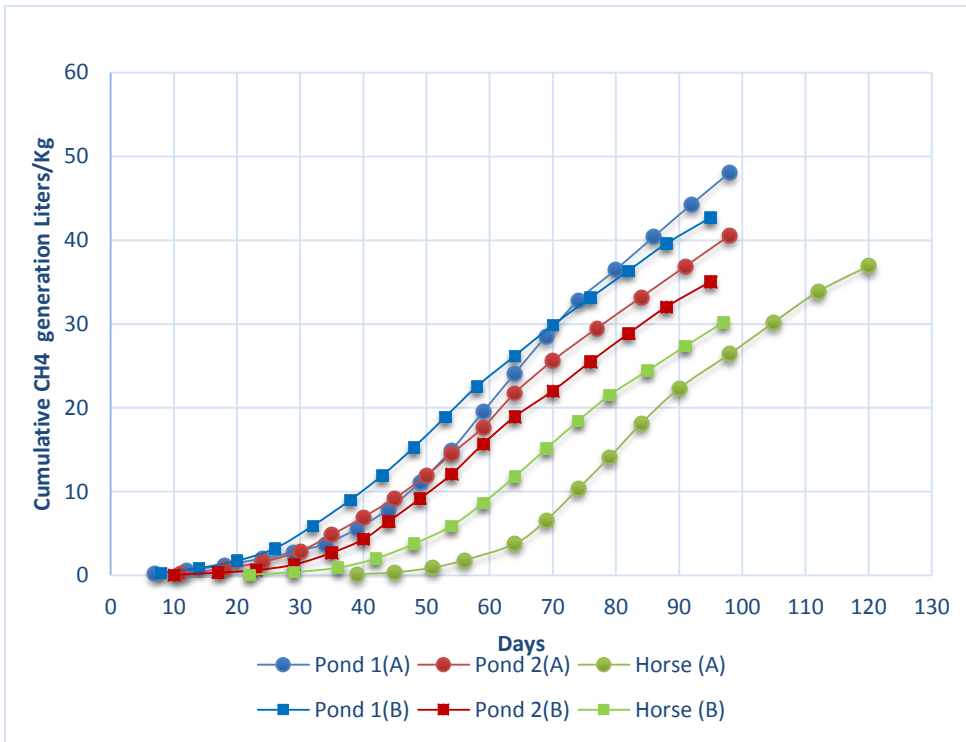


Figure 4- 11 Cumulative methane generation (liters/kg) vs. time for duplicate sets of reactors

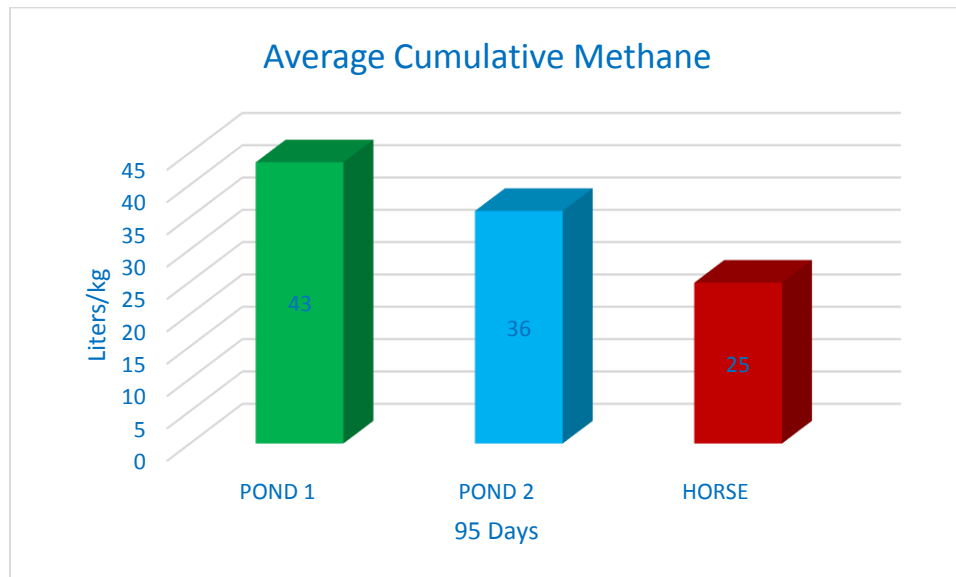


Figure 4- 12 Average cumulative CH₄ generation for Pond1, Pond 2 and Horse (95 days)

Table 0-5 Cumulative Methane Generated for Pond 1, Pond 2, and Horse (Duplicates)

Reactors	Days	Cumulative CH ₄ (liters)	Cumulative CH ₄ (liters/kg)
End of Reactor Operation			
Pond 1(A)	98	147	48
Pond 2(A)	98	124	41
Horse (A)	120	113	37
Pond 1(B)	95	131	43
Pond 2(B)	95	107	35
Horse (B)	97	93	30

All the reactors were continuously generating methane but the rate and percentage of methane was decreasing constantly. Reactors Pond 1 and Pond 2 were taken out of operation at day 98 and reactor Horse dung at 120 days for the first run; however, methane formation in all three reactors was not ceased. Similarly Pond 1 and Pond 2 were taken out of operation at day 95 and Horse at day 97 at the second set of duplicate reactors.

Figure 4-13 shows the average maximum methane generation rate (ml/kg/day) vs. time for duplicate reactors. For the reactor duplicates, Pond 1 has highest rate of methane generation, likely due to a larger amount organic matter on the pond bottom soil which becomes a favorable site for microbial development. Horse has the lowest rate of methane generation. Table 4-6 shows maximum methane generation rates and days on which they were observed. The maximum rate of methane generation (ml/kg/day) was obtained within 100 days of the reactors operation.

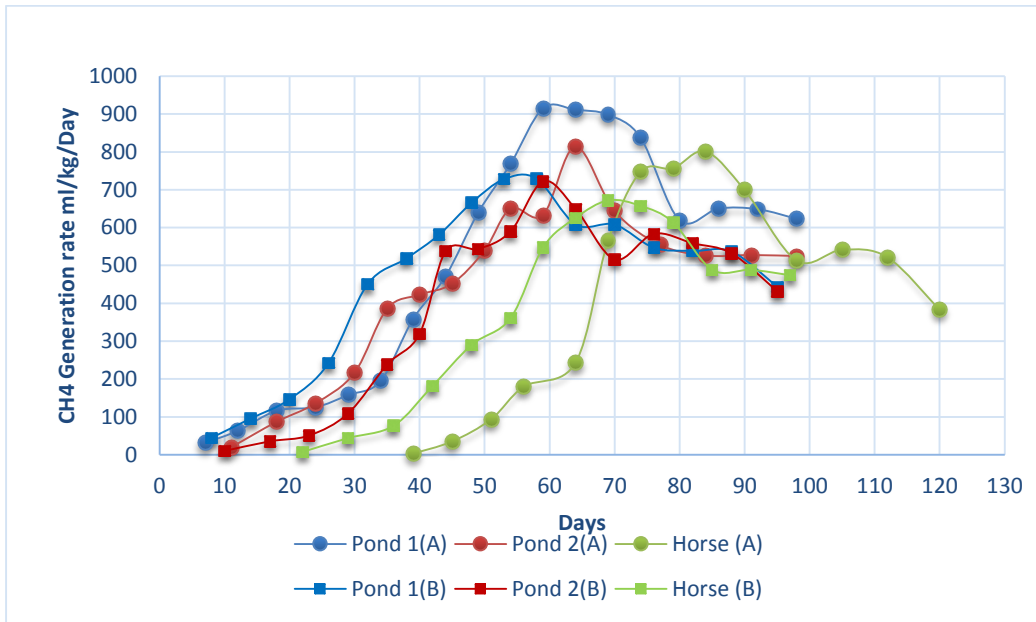


Figure 4- 13 Methane generation rate (ml/kg/day) vs. time for both sets of reactors

Table 0-6 Average CH₄ Generation rate (ml/kg/day)

Reactor	Maximum CH ₄ generation rate (ml/kg/day)
Pond 1	822
Pond 2	768
Horse	737

We shall now compare the result obtained from this study with reported data of anaerobic digestion in the literature. The rate of biodegradation of MSW in landfills was studied by Barlaz et al. (2004), in small pilot-plant columns. The methane generation rate peaked at less than one hundred days of reactor operation as shown in Figure 4-14, which is in good agreement with the results obtained in the present study.

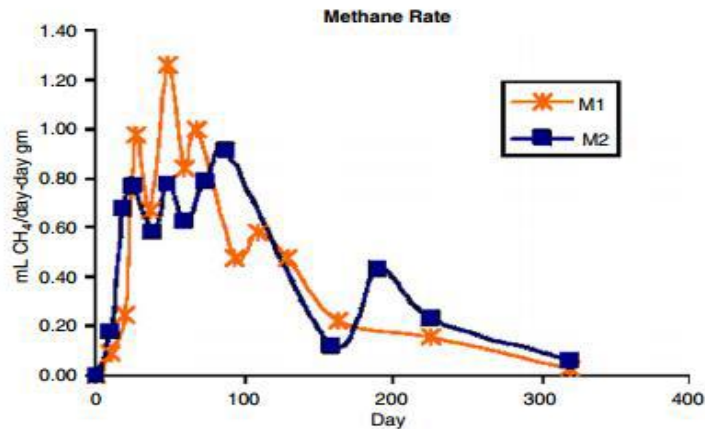


Figure 4- 14 Generation of methane in experimental reactors (Barlaz et al., 2004)

(M1 and M2 denote two different tests)

4.3 Sludge pH for reactors (Food, Paper and Yard) seeded with digestate from Pond 1(B)

From duplicate sets (A and B) of the first experiments, reactors seeded with the pond soil obtained from Pond 1 (smaller pond) was started sooner to generate methane and had higher cumulative methane generation. As shown in Fig. 4-11, cumulative methane at the initial stage of Pond 1(B) was greater than Pond 1(A). Pond 1(B) generated more methane faster so the digestate from Pond 1 (B) will be used for further testing. Figure 4-15 shows pH variation in reactors seeded with digestate from Pond 1(B) for the first 49 days of operation. In the initial days, pH level of the reactors was less than 6.5; this acidic phase existed because of the ongoing acid accumulation state in waste decomposition. The pH level continued to decrease up to 9 days for Reactor Food, 11 days for Paper, and 11 days for Yard. A gradual rise in pH level was observed afterwards in these reactors, stabilizing in the range between 7 and 8, but fluctuating between these values. pH level for Food and Yard was observed to fluctuate more than the Paper waste. The rise in pH was due to the conversion of carboxylic acid into methane and carbon dioxide in the fourth phase of biodegradation.

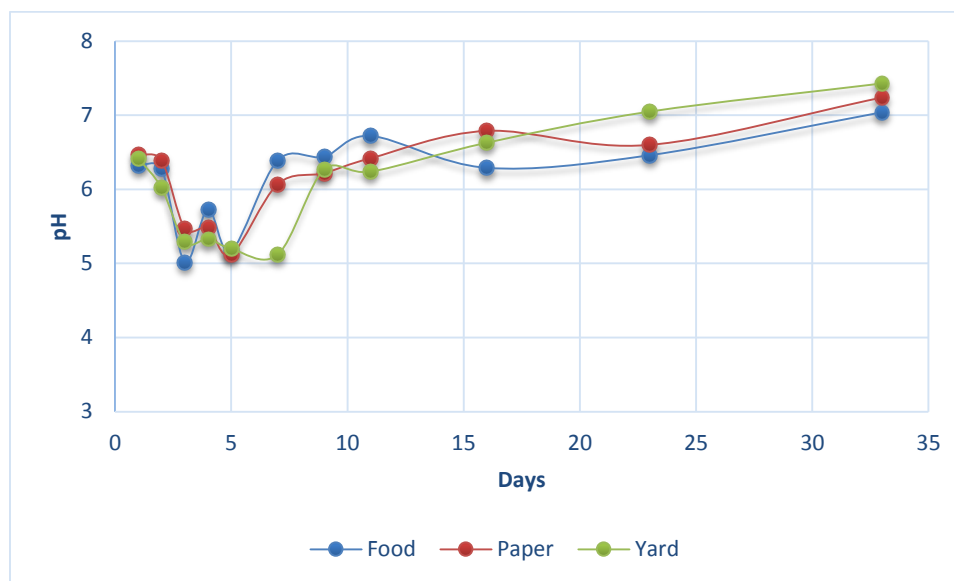


Figure 4- 15 pH variation vs. time for reactors Food, Paper and Yard

4.4 Gas data for reactors (Food, Paper and Yard) seeded with a digestate from Pond 1(B)

4.4.1 Gas composition

Figures 4-16, 4-17, and 4-18 show composition of gases generated from the reactors Food, Paper and Yard, respectively, for the first 49 days of operation. Oxygen decreased and remains low with time, which indicates anaerobic conditions inside the reactors. The “other gases” likely represent molecular nitrogen present in air, carbon monoxide, hydrogen sulfide, and hydrogen. The other gases were initially lower than 20% for Food and around 40% for Paper and Yard. The percent decreased over time to around 5 % for Food and Paper, and to around 15% for Yard.

Initially it took 3, 5 and 14 days for reactors Food, Paper and Yard respectively to form some gases. The carbon dioxide percent in all reactors was initially higher than methane percentage, which was due to the acetogenesis phase which produces carbon dioxide. Carbon dioxide for Food peaked at 80.2 % at day 5, 72% for Paper at day 11 and 74% for Yard at day 21. The percentage of carbon dioxide then decreases as methane increases over time, as the methanogenesis phase produces methane. The amount of CO₂ decreased gradually with a mild slope over time for Food, with some fluctuations. Carbon dioxide decreased gradually to 40.7 % 46.8 % and 45.5 % for Food, Paper and Yard, respectively, at day 68.

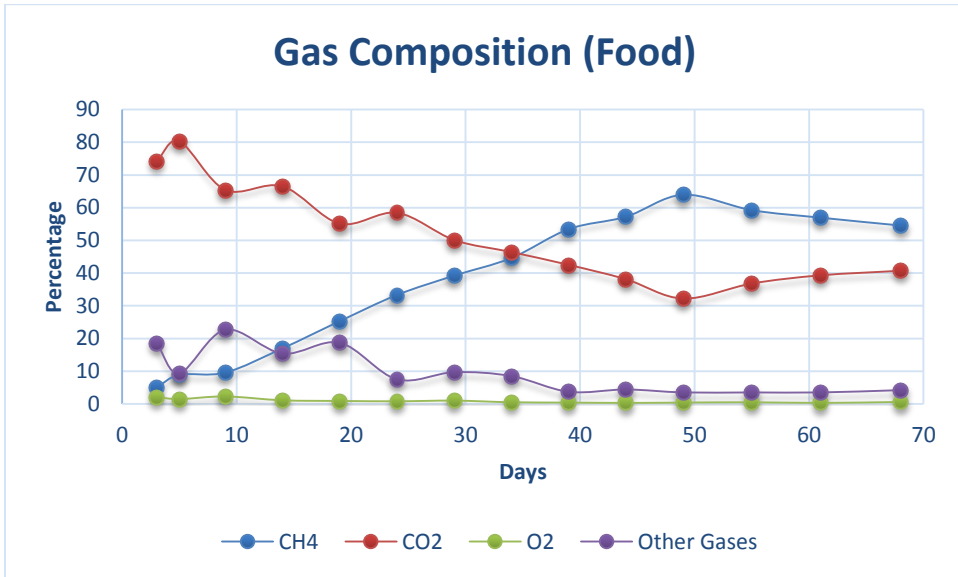


Figure 4- 16 Gas composition percent vs. time for reactor Food

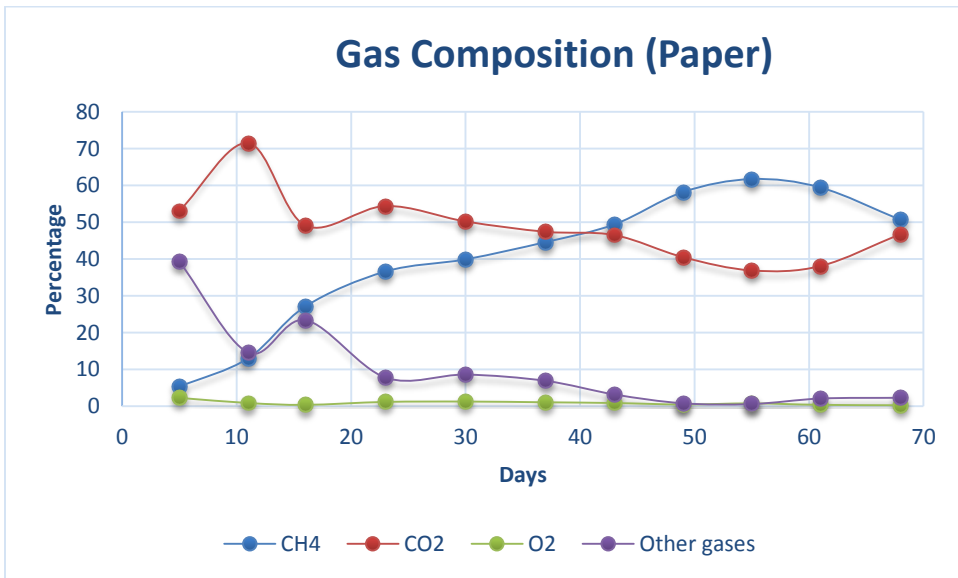


Figure 4- 17 Gas composition percent vs. time for reactor Paper

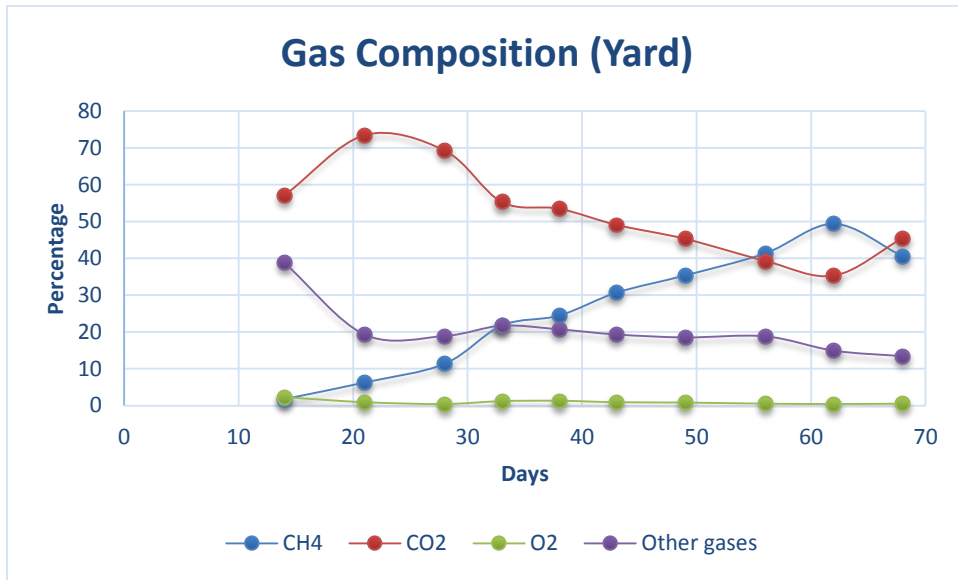


Figure 4- 18 Gas composition percent vs. time for reactor Yard

Figure 4-19 shows methane percent over time for all reactors Food, Paper and Yard. Gas formation in reactor Food started sooner than other two, likely because food wastes typically decompose faster. The highest methane percentage was also observed for the food waste reactor, perhaps because it has highest amount of cellulose and has larger surface areas per unit volume of waste for the microbes to access, compared to the reactors which have more Paper and more Yard wastes. Initially the methane percentage was 5.1 (day 3) for Food, 5.3 (day 5) for Paper and 1.8 (day 14) for Yard. Then the methane percentage increased over time. For 68 days of reactor operation, Food has the highest methane percentage followed by Paper, Yard has the lowest percent of methane. Maximum methane percentage of 63.9 (on day 49), 61.7 (on day 55) and 49.4 (on day 62) were observed for reactors Food, Paper and Yard, respectively. Then the methane was leveling off to 54.5%, 50.6% and 40.6% for Food, Paper and Yard respectively on the 68 days of reactor operation. These reactors are still being operated.

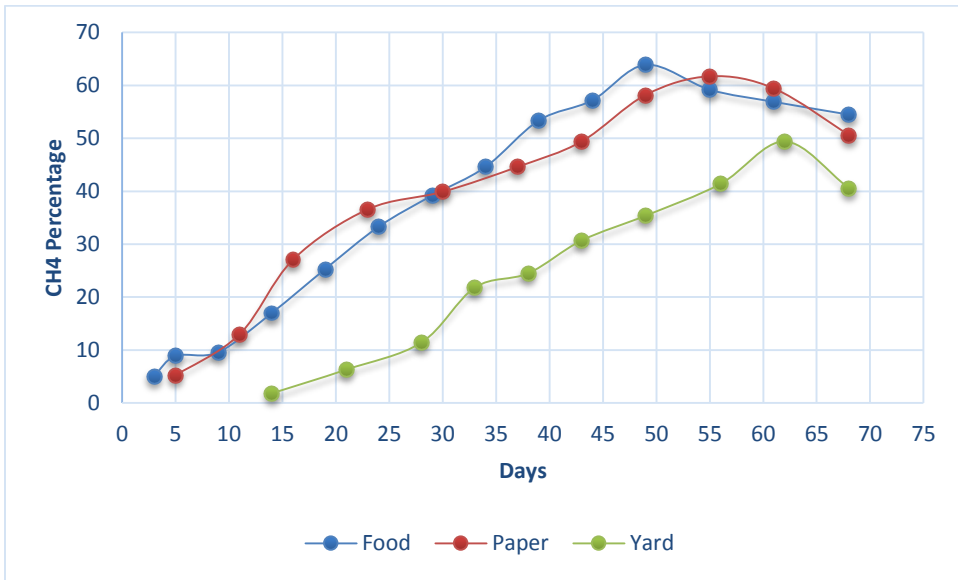


Figure 4- 19 Methane percent vs. time for reactors Food, Paper and Yard

Figure 4-20 shows methane to carbon dioxide ratio for Food, Paper and Yard. For all reactors methane to carbon dioxide ratio increases over time. Maximum methane to carbon dioxide ratios of 1.98, 1.7 and 1.4 were observed for Food, Paper and Yard respectively and levelling off to 0.89 to 1.3 through the 68 days of reactor operation.

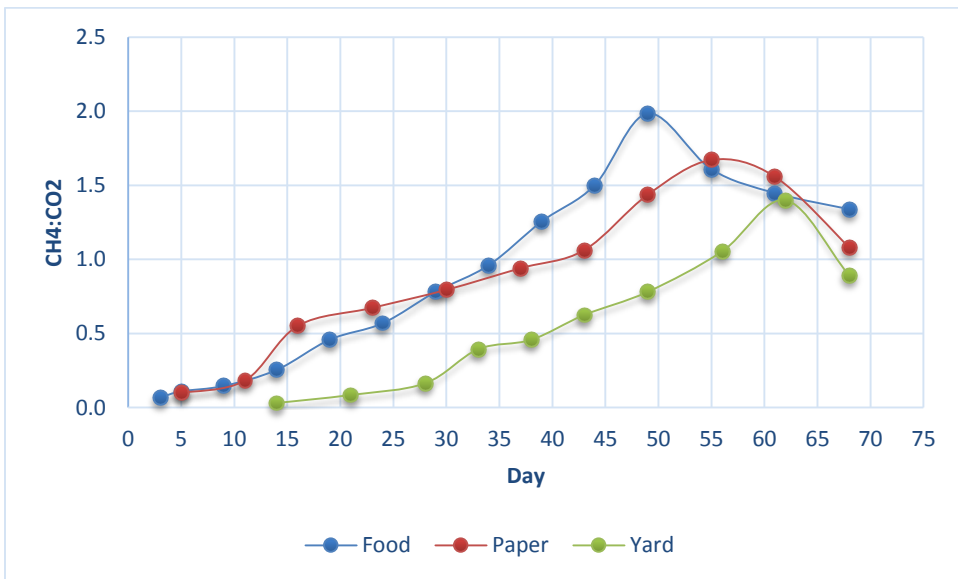


Figure 4- 20 Methane to carbon dioxide ratio for Food, Paper and Yard

4.4.2 Cumulative volume and rate of methane generation

Cumulative methane generated in liters/kg from reactors Food, Paper and Yard over time is shown in Figure 4-21. The highest amount of cumulative methane was observed on reactor Food, followed by Paper. Reactor Yard had the lowest amount of methane production during 68 days of operation. It is likely food and paper waste are more easily decomposable than the yard wastes. Table 4-7 shows the cumulative methane generation for Food, Paper and Yard for 68 days of reactor operation. Cumulative methane of Food is 1.13 times more than Paper and 2.0 times more than the reactor Yard.

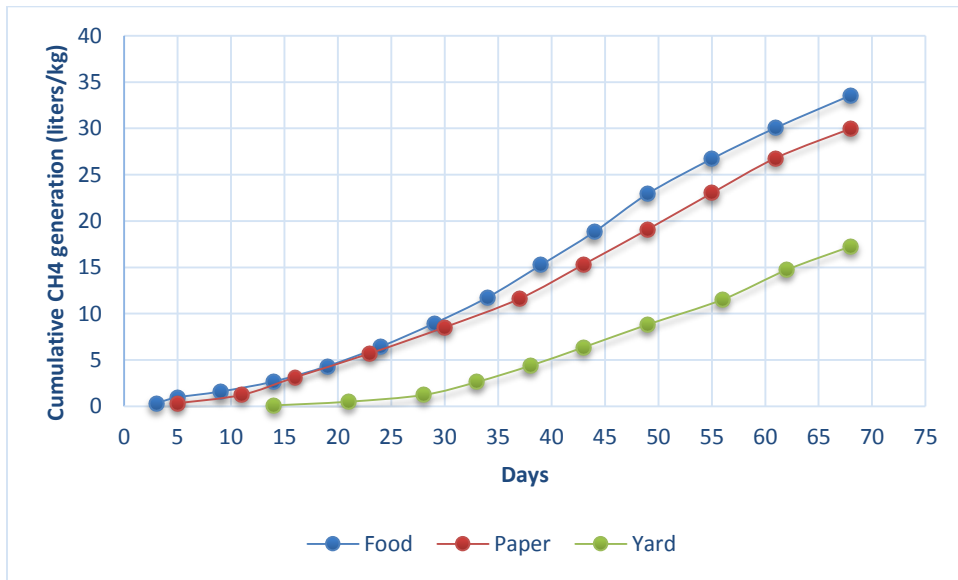


Figure 4- 21 Cumulative methane generation in reactors Food, Paper and Yard to 68 days

Table 0-7 Cumulative Methane Generated for Food, Paper and Yard

Reactor	Cumulative CH ₄ (liters)	Cumulative CH ₄ (liters/kg)
Food	103	34
Paper	92	30
Yard	53	17

Figure 4-22 shows methane generation rate (ml/kg/day) for Food, Paper and Yard. In all reactors methane generation rate increases with time. Methane generation rate for Food is higher than paper, which is higher than Yard. For Food, Paper and Yard, maximum methane generation rate 823 (on day 49), 655 (on day 55) and 535 (on day 62) ml/kg/day was observed and levelling off to 425 to 500 ml/kg/day through 68 days. These reactors are still being operated.

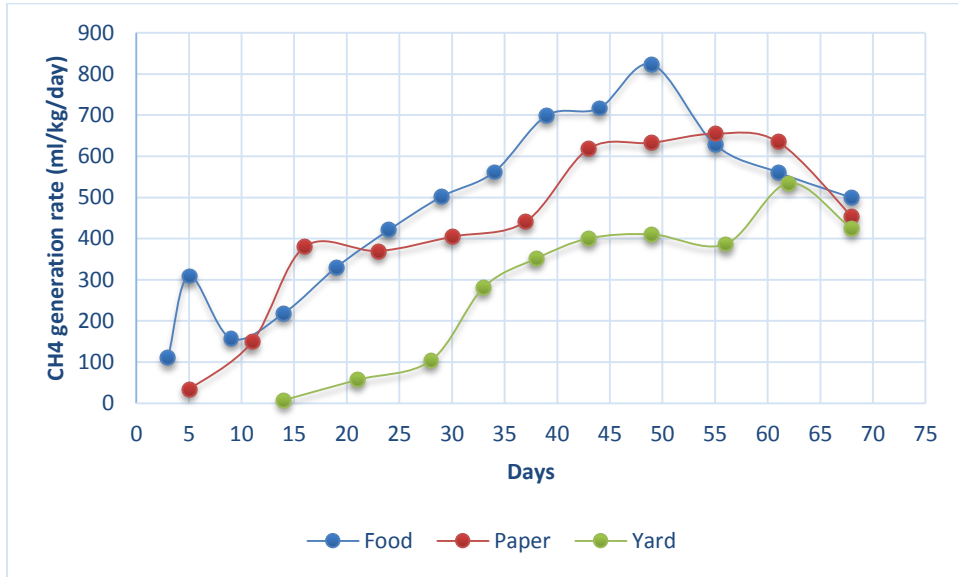


Figure 4- 22 Methane generation rate (ml/kg/day) for Food, Paper and Yard to 68 days

Chapter 5

Conclusions and Recommendations

5.1 Conclusions

The main objectives this study were to compare the rate of methane production and cumulative methane generation from degradation of a mixture of household organic wastes (food, paper, and yard) using 2 types of pond soil and horse dung as an inocula and to determine the effectiveness of the best inoculum from the first experiment in degrading mixtures with various proportions of food, paper and yard waste. The results obtained from the current study can be summarized as follows:

1. For both sets of reactors in experiments A and B, the reactor seeded with pond bottom soil obtained from Pond1 (smaller pond) had the highest methane generation rate, as well as the largest cumulative volume of methane, followed by Pond 2 (larger pond) and finally the reactor seeded with Horse dung. The possible reason is that Pond 1 is an older pond compared to Pond 2, and the bottom soil obtained from the Pond1 may have more stable organic matter accumulated as well as higher nutrient concentrations.
2. For the both sets of reactors in experiments A and B, reactors inoculated with Pond1 started gas formation sooner than the Pond 2 and Horse. A reactor inoculated with Horse dung took longest time to start up the gas formation, possibly due to less organic matter in the horse dung, leading to an initial thin population of microbes as well as it taking a longer to acclimate with the environment inside the reactor. When gas formation started, it increased quickly.
3. Methane production rate in Pond1 (A) was higher than Pond 1 (B); similarly Pond 2 (A) was higher than Pond 2 (B). It may be because of the microbe populations differ in the different times of the year such as August and November in the same pond. Pond soil obtained in August was observed to give better performance on the reactors than pond soil obtained in the November. Also the recorded temperature for the duration of reactor operation in experiment "A" was higher than the

experiment “B.” Since microbial activity typically increases with temperature, the higher August temperatures may explain the greater methane generation for experiment set (A).

4. The cumulative average methane generation in 95 days for the reactor seeded with Pond 1 was 19.4 % higher than reactor seeded with Pond 2 and 72% higher than reactor seeded with Horse dung. Similarly, reactor Pond 2 generated 44% more methane than the reactor with the horse dung. Pond soils obtained from the Pond 1 and Pond 2 gave better performance than horse dung as an inoculum for treating household wastes.
5. In experiment C, for all reactor inoculated with digested waste of Pond1 (B), reactor with more Food started gas formation sooner than Paper and Yard. It is likely because of food waste decomposes faster.
6. For all three reactors (Food, Paper and Yard) in experiment C through the 49 days of reactor operation, Food waste had the highest rate of methane generation as well as largest cumulative volume of methane, it is because of food waste has highest amount of cellulose, more surface areas per unit volume of waste for the microbes to access, than paper and yard waste. Reactor operation will be continued.

5.2 Recommendations for future studies

Based on the results obtained from the present study, and to increase its accuracy, the following recommendations are made for future study.

1. Performance of reactors can be investigated by using different percentage of inoculum such as 10%, 20%, 30% and different waste to inoculum ratios.
2. In this study, the experiments were conducted at room temperature, so further study on effect of inoculum for reactor performance can be done at fixed temperature such as 30^oC, 35^oC, and 40^oC. In addition, temperatures representative of rural communities of developing countries can be explored.

3. Investigate and compare reactors performance by recirculating the leachate and see if horse dung accelerates the methane formation.
4. Reactor performance of different inocula (Pond soil and Horse dung) can be investigated by utilizing single waste such as Food, Yard or Paper only instead of using mixed waste.
5. Pond soil as inocula can be compared to sewage sludge and other types of dung as inocula.
6. Chemical pretreatment of waste samples can be done to accelerate rate of decomposition.
7. Reactor performance can be investigated by using pond bottom soil as microbe's seed, from the same pond but different depth/place and different seasons.
8. Further study can to be done by applying horse dung along as an inoculum for different type of waste composition and find out whether methane producing slower on all the reactors.
9. Identification of the microbial communities (acidogens, methanogens, etc.) in the reactors seeded with the different inocula.
10. Quantifying organic matter in soil via Standard Method APHA 2540-E (volatile solids).

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Biographical Information

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