EFFECT OF MANURE AND ENZYME ON THE DEGRADATION OF ORGANIC FRACTION OF MUNICIPAL SOLID WASTE IN BIOCELL

by

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Abstract

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More than half of total Municipal Solid Waste (MSW) generated is composed of organic degradable waste for developed countries and the percentage is even higher for developing countries. Handling of organic fraction of MSW has always been challenging due to environmental and economic reasons. Among all the MSW management alternatives, Sustainable Biocell concept has potential to solve environmental and economic concerns related to organic waste. This potential of Sustainable Biocell can be enhanced by accelerated decomposition of waste. Accelerated decomposition of organic fraction of MSW can be achieved by the addition of nitrogen rich, macro and micro nutrients such as sludge and manures. Several studies have been conducted by using either sludge or animal's manures as inoculum for degrading organic fraction of MSW in the anaerobic digester. However, the study to compare the performance of different manures combined with sludge on the degradation of organic fraction of MSW has not been conducted on laboratory simulated Biocell reactors. Also, the effects of different types of manure on degradation of organic fraction of MSW have not been compared with the lignocellulos enzyme. Therefore, this study was conducted with the objective of finding the effects of addition of sludge-manure and sludge-enzyme in the degradation of organic fraction of MSW within laboratory simulated Biocell environment and comparing them.

In this study, five sets of reactors were prepared with selected organic fraction of MSW and inoculum composition. The reactors were operated in the environmental growth chamber at 37ºC and were monitored regularly. The pH, BOD and COD tests were conducted on generated leachate along with gas composition and gas volume measurement. Based on the experimental results, reactors with Manganese Peroxidase (MnP) and pig manure as inoculum presented better result compared to reactors with cow manure and horse manure. The highest volume of methane was generated by reactors designated as A9 and A10 with MnP, produced 54.7 L/lb. and 52.03 L/lb. with peak methane yield of 1.33 and 1.57 L/lb./day respectively. The reactors designated as A5 and A6, with pig manure produced 47.26 L/lb. and 45.91 L/lb. with peak methane yield 1.45 and 1.30 L/lb./day respectively. And, the percentage of volatile solids removal was found to be 77.01% and 77.2% for reactors A9 and A10 with MnP respectively and 75.09 and 76.87% for reactors A5 and A6 with pig manure. Therefore, based on the experimental study, it was determined that MnP and pig manure have the potential to be utilized as inoculum for improving the degradation of waste. Even though MnP enzyme helped to degrade more waste and yielded the highest quantity of methane, it is not considered an economical solution for field application due to high costs of MnP enzyme. Therefore, pig manure is suggested as an economical inoculum for Sustainable Biocell landfill.

Table of Contents

List of Illustrations

List of Tables

Chapter 1

Introduction

1.1 Background

Municipal Solid Waste (MSW) management has become the subject of major concern due to environmental and economical concern. Though landfilling is least preferred option in integrated solid waste management hierarchy, it is still the most practiced approach of waste management (US EPA, 2016). This is demonstrated by the fact that 136 million tons of waste i.e. more than half (52.6%) of the total generated waste in the USA, was landfilled in 2014 (US EPA, 2016). MSW consists of degradable organic and non-degradable inorganic fraction. The composition of MSW based on degradability for different countries is shown in Figure 1-1 (Hoornweg, D., & Bhada-Tata, P., 2012; US EPA.,2016).

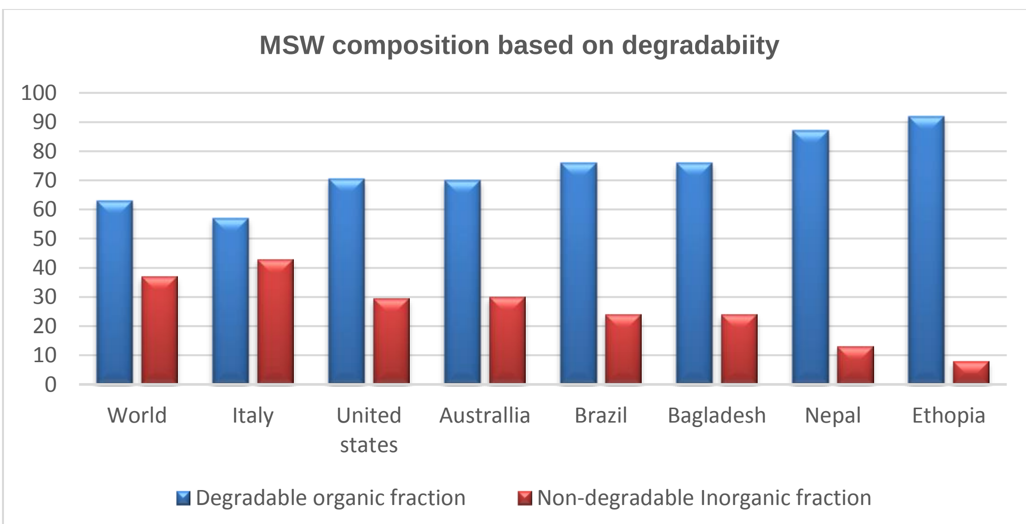

Figure 1-1 Comparison of composition of MSW based degradability. (Hoornweg, D., & Bhada-Tata, P., 2012; US EPA.,2016).

MSW composition data for different countries show that the degradable organic waste constitutes more than half of total MSW generation. Despite being such a significant fraction of MSW, proper handling and disposal of organic fraction of MSW has always been a challenge for the solid waste industry due to its adverse effect on the environment. Compositing and anaerobic digesters are considered as alternative solutions for managing the organic fraction and utilizing its energy potential. However, composting and anaerobic digester are not considered economical due to high initial investments and high operating costs. As an alternative to composting and anaerobic digester, landfilling is considered an economical alternative for managing the organic fraction of MSW.

1.2 Evolution of Biocell or Sustainable Biocell

During initial stages, landfilling started with the inadequate disposal of waste called open dump. But open dumping of organic fraction of MSW can cause environmental pollutions such as soil, groundwater and surface water contamination, uncontrolled greenhouse gases (GHG) emissions from anaerobic decomposition or burning of the waste, spreading of diseases by different vectors such as birds and insects, and space recovery (Visvanathan and Glawe, 2006). With the realization that open dumps can cause environmental pollution including groundwater contamination along with GHGs emission, the concept of conventional landfilling techniques evolved. However, conventional landfilling techniques were not found to be the effective and sustainable solution for landfill gas, long-term environmental stability, and space recovery. Then, the concept of bioreactor landfill was evolved by Pohland in the 1970s (Pohland, 1975). Though, the evolution of bioreactor landfilling concept solved the problems associated with conventional landfilling such as ground water contamination, leachate generation, waste stability, gas emissions, and space recovery problems were still unsolved. Therefore, a novel concept of sustainable landfilling was evolved called Biocell (Hettiaratchi, 2007). Biocell is operated in three steps: - anaerobic degradation followed by aerobic degradation/composting, and the landfill mining (Hettiaratchi, 2007). It is an improvement over bioreactor landfill. Biocell has the ability to handle high moisture, capture the generated gas from anaerobic decomposition. Biocell is considered as an integral part of the recently purposed sustainable management system. Therefore, Biocell can be considered as a sustainable alternative for organic waste degradation. The flow of waste management system by Hossain (2015) is presented in Figure 1-2.

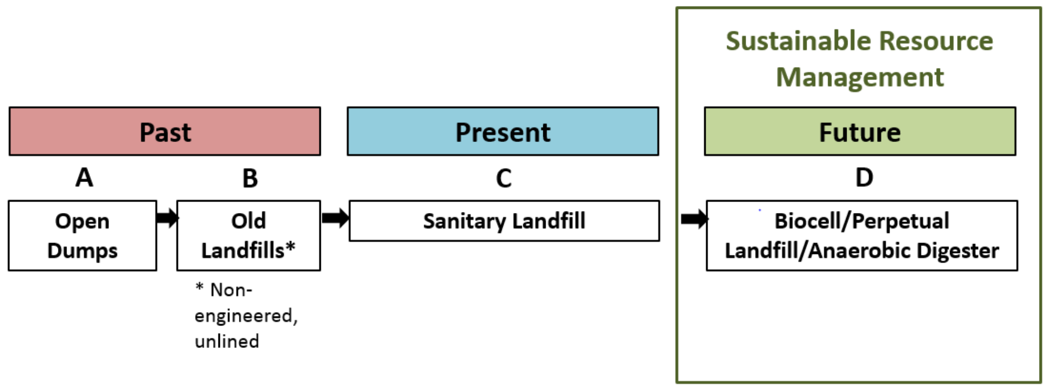

Figure 1-2 Flow diagram for waste management practices (Hossain, 2015)

1.3 Problem statement

Sustainable Biocell has potential to solve most of the problems associated with organic waste such as leachate contamination of the environment, uncontrolled fugitive GHG emissions, and scarcity of land. Since anaerobic degradation of waste is the first stage of Sustainable Biocell, faster degradation of waste is critical to shortening waste stabilization process. Therefore, enhancement of degradation process is necessary for a landfill to be sustainable.

However, the Carbon/Nitrogen ratio (C: N) ratio of organic waste is relatively high, which was one of the most frequently found problems during degradation of waste (Bujoczek, et al., 2002). To sidestep this problem, the nitrogen, other macro and micro nutrients should be provided at sufficient levels that are not present in the organic degradable waste. Several researchers have proposed co-digestion of organic fraction of

MSW, either with sewage sludge from Municipal Wastewater Treatment Plants (MWWTPs) or residues from livestock farms to resolve the problems associated with the organic fraction of MSW in Anaerobic digester (Sosnowski, et al., 2003). However, the percentages of inoculum used in these studies are higher which is not feasible for field scale study and the study to compare the performance of different manures along with sludge on the degradation of organic fraction of MSW have not been conducted on laboratory simulated reactors. Also, the effect of different types of manure on the degradation of organic fraction of MSW has not been compared with lignocellulos enzyme, Manganese Peroxidase (MnP). MnP was proved to enhance gas generation for 30 years old landfill waste as compared to other enzymes (Jayasinghe, 2013). Therefore, the decision to conduct this study was made to study the effect of enzyme and manure on the degradation of organic fraction of MSW in Biocell.

1.4 Research objective

The main objective of the current research is to determine the effect of nutrients on the degradation of organic fraction of MSW. The use of nutrients such as manure, sludge, and enzyme can enhance the waste degradation process. Therefore, it necessary to investigate the effect of different type of nutrients on waste degradation and to find the effective nutrients which can enhance gas generation. For this purpose, a systematic experimental program was undertaken to fulfill the following objectives:

1. To evaluate the effects of different types of manure and enzyme on bio-degradation of organic fraction of MSW in a laboratory simulated Biocell landfill.
2. To determine the effects of different types of manure and enzyme on gas generation of organic fraction of MSW in a laboratory simulated Biocell landfill.
3. To find out the effective and better manure and enzyme for maximum gas production.

1.5 Thesis outline

The overall thesis is organized in the following chapters:

Chapter 1 includes the overall introduction along with the problem statement and objective of the study.

Chapter 2 presents a literature review on MSW generation and its properties, problems associated with organic fraction of MSW, Sustainable Biocell concept, waste degradation process, manure management, and effect of nutrients on decomposition.

Chapter 3 describes the experimental program and all the adopted methodologies to fulfill the research objectives. It includes collection of MSW and inoculum, construction of laboratory simulated landfill reactors, and monitoring of reactors.

Chapter 4 mainly focuses on the results obtained from the experimental study on laboratory simulated Biocell reactors. Results and its analysis have been presented in details along with its comparison with previous literatures.

Chapter 5 summarizes the results and main conclusions obtained from this study along with the recommendations for future work.

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Chapter 2

Literature Review

2.1 Municipal solid waste

Municipal Solid Waste (MSW) is leftover that has no use for the owner but is the potential source of energy. According to US EPA (2016), MSW, commonly known as garbage or trash, is a subset of solid waste and is defined as durable goods (e.g., appliances, tires, and batteries), nondurable goods, containers and packaging, food wastes, yard trimmings, and miscellaneous organic wastes from residential, commercial, and industrial non-process sources.

2.2 Municipal solid waste generation and disposal

MSW generally originate from homes, institutions, commercial sources, and small businesses. Total MSW waste generation in the United States from 1960 to 2014 is shown in Figure 2-1. In the United States, 258 million tons of trash was generated in 2014. Among 258 million tons of waste, 89 million tons (about 34.6%) were recycled and composted, over 33 million tons (about 12.8%) of MSW were combusted with energy recovery, and 136 million tons (52.6%) were landfilled (US EPA, 2016). This shows that the highest percentage of MSW generated was landfilled. Figure 2-2 shows management of MSW in the United States in 2014.

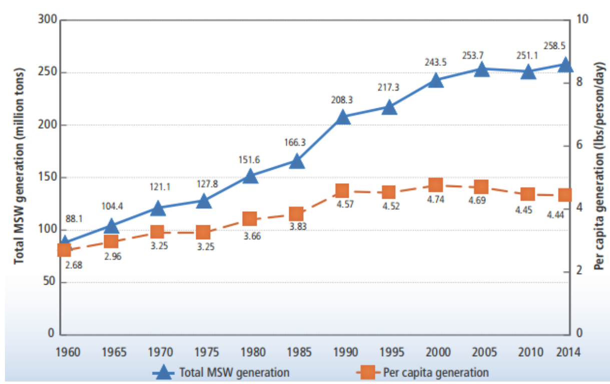


Figure 2-1 MSW generation rates from 1960 to 2014 (US EPA, 2016)

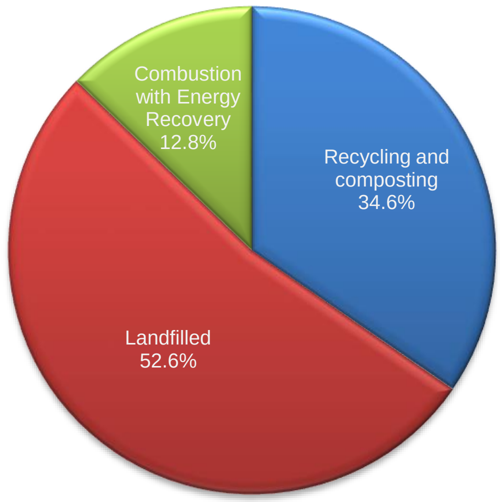


Figure 2-2 MSW management in the United States in 2014 (US EPA, 2016).

2.3 Composition of Municipal Solid waste

Waste composition is the term used to describe the individual components that make up a MSW stream and their relative distribution, usually based on the percentage by weight. (Tchobanoglous, et al., 1993). The MSW generally consists of paper, plastics, yards & wood waste, leathers & textiles, C&D, glasses, metals and fines & soils. MSW is also classified based on its degradability such as- degradable and non-degradable waste. Table 2-1 shows the classification of MSW by Landva and Clark (1990).

Table 2-1 MSW classifications by Landva and Clark (1990)

Organic waste	Putrescible	Food waste, Animal waste, Garden waste, Material contaminated by such waste
	Non-putrescible	Paper, Wood , Textiles, Leather, Plastic, Paint,oil, Grease,Chemicals , Organic Sludge.
Inorganic waste	Degradable	Metals(corrodable to varying degree)
	Non- Degradable	Glass, Ceramics, Mineral soil, Rubbles, Tallings, Slimes, Ash, Concrete, Masonry (C & D)

In the United States, the MSW generated in 2014 consisted of 26.6 % papers and paperboard, 13.3% yard trimmings,14.9% food waste, 12.9% plastics, 9% metals, 9.5% rubber, leather & textiles, 6.2% wood, 4.4% glass, and 3.2 % other miscellaneous wastes (US EPA, 2016). Figure 2-3 shows the breakdown of MSW generation by the material. Organic materials such as paper, food, yard and wood waste, and textiles was the largest component of MSW which is equivalent to 64.3% of total MSW generation in 2014 (US EPA, 2016).

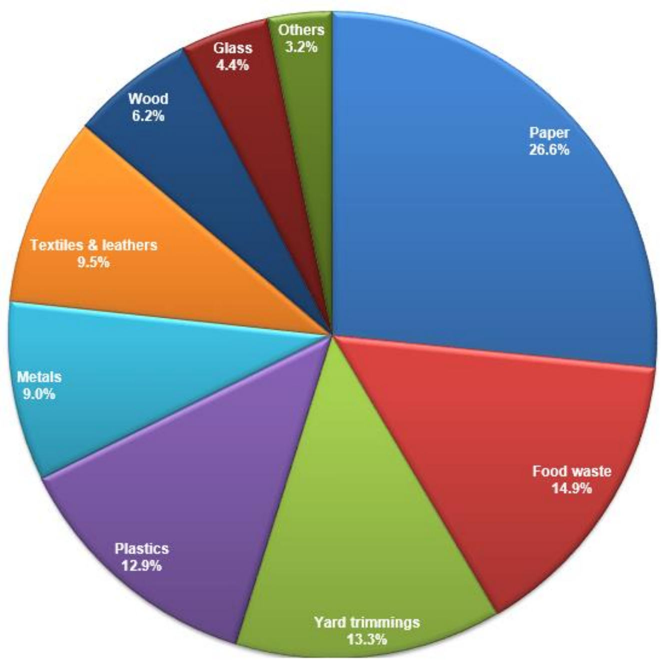


Figure 2-3 MSW composition in United states in 2014 (US EPA ,2016)

Composition of MSW varies from places to places and from countries to countries. The composition of MSW depends on many factors including regional and seasonal variation, population density, income of the average people, culture climate, extents of waste diversion, the existing legislation, and frequency of collection of waste. (Tchobanoglous, et al., 1993). Table 2-2 shows the composition of MSW based on economy of the country (Tchobanoglous, et al., 1993).

Table 2-2 MSW composition based on economy of country (Tchobanoglous, et al., 1993)

	Components	Low-income countries	Middle income countries	Upper income countries
Organics	Food wastes	40-85	20-65	6-30
	Paper/Cardboard	1-10	8-30	20-45
	Plastics	1-5	2-6	2-8
	Textiles	1-5	2-10	2-6
	Rubber/Leather	1-5	1-4	0-2
	Yard wastes/ Wood	1-6	1-10	11-25
Inorganic	Glass	1-10	1-10	4-12
	Tin cans/ Aluminums /Other metals	1-5	1-5	3-15
	Dirt, ash etc.	1-40	1-30	0-10

Table 2-2 shows that organic MSW constitutes the highest percentage of the total generated waste for low, middle and high income countries. Low-income countries have the highest proportion of organic waste as compared to middle and high income. Therefore, irrespective of the economy, organic fraction is the major portion of MSW generation.

2.4 Organic fraction of municipal solid waste

The Organic Fraction of Municipal Solid Waste (OFMSW) is defined as the degradable portion of the waste which contains carbon compounds derived from animal and plant materials. Typical organic components of MSW are food wastes, paper, cardboard, textiles, rubber, leather, yard wastes, wood etc. (Tchobanoglous, et al., 1993). Disposal of MSW containing OFMSW pollutes the environment and causes the health hazards. The problem associated with the OFMSW is described in the sections below.

2.4.1 Problems associated with disposal of OFMSW

The presence of organic waste in the MSW causes environmental pollution including ground water contamination and greenhouse gases (GHGs) emission. The moisture content of OFMSW is comparatively higher because of the presence of food, paper, and yard waste. The biodegradation of waste generated leachate which when infiltrate into the groundwater sources causes the contamination. Contamination of ground water sources can cause the heath related hazards.

Organic part of the MSW is the potential source of landfill gases (LFGs). The major constituents of LFGs are methane (CH_4) and carbon dioxide (CO_2). The organic substance present in waste disposal sites can undergo biological transformation to produce carbon dioxide (CO_2) under aerobic conditions and a mixture of methane (CH_4) and carbon dioxide (CO_2) under anaerobic conditions (US EPA, 2010). Methane (CH_4) and carbon dioxide (CO_2) are major GHGs. It is a well-established that the presence of methane and carbon dioxide in the atmosphere contribute to global warming and climate change. Methane is a particularly potent GHG and is currently considered to have a global warming potential (GWP) 25 times that of CO_2 when a time horizon of 100 years is considered.(UNEP, 2010).

Therefore, proper management of solid waste is required for reducing the environmental and health related issues. Even though disposal of MSW is the last choice of solid waste management hierarchy, the disposal of MSW in landfills is the most used method (USEPA, 2016). Recycling, compositing, landfilling, and anaerobic digestion are the commonly used solid waste management techniques. Among them, the predominant solid waste disposal and management in the United States is landfilling and considered as one of the most economic and sustainable options for solid waste disposal system.

2.5 Landfill

Landfill is the place where collected refuse and other waste material is disposed. Usually, waste is buried and covered with soil in the landfill.

2.5.1 Historical aspect of landfilling practice

The open dumps were used for waste disposal in early age. With the realization of environmental issues and health related hazards, sanitary landfills were evolved along with the concept of integrated waste management techniques, followed by landfill bioreactors and then to Sustainable Biocell. (Hettiaratchi, 2007) Historical aspects of landfilling in North America is described in following sections.

2.5.1.1 Evolution of conventional landfill

Open dumping practices lead to environmental pollution including ground water contamination and greenhouse gas emission. Dry tomb sanitary landfilling concept was introduced to control the environmental pollution which is also known as first generation sanitary landfill or conventional landfill. Liquids are banned from the landfill to control the formation of leachate and waste is covered with the daily, intermediate and final cover. All efforts were carried out to prevent the formation of leachate thereby trying to keep waste in dry condition. With time, leachate collection system and bottom-liner were being introduced provided to prevent subsurface leachate from penetrating into the ground water. But in these landfills, the rate of degradation of waste is slow due to the lack of moisture required for microbial activity. It takes more than hundred years to complete the decomposition of waste. According to US EPA federal regulations (1991), 30 years post-closure monitoring period is specified which further extends the lifespan of conventional landfills (Barlaz et al.,2002). With the realization that sanitary landfill could still cause environmental pollution as well health risks with the exposer to the moistures during its extended life periods, it was tried to keep as landfill’s footprint as minimum as possible. For achieving minimum footprint of the landfill, the other waste management techniques should be adopted to reduce the waste production and disposal.

2.5.1.2 Evolution of integrated waste management techniques

With the concept of treating the generated waste as resources such as recycling and composting, the concept of integrated waste management techniques evolved (Hettiaratchi, 2007). It further helped to keep as landfill's footprint as minimum as possible. A hierarchy of integrated waste management system is shown graphically in Figure 2-4.

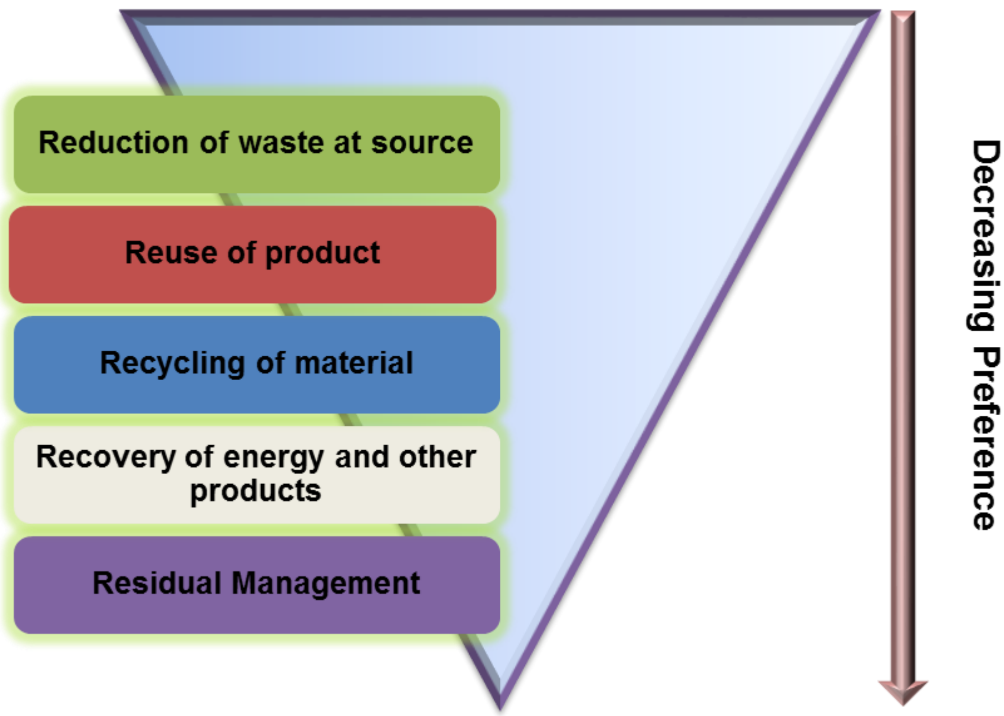


Figure 2-4 Integrated waste management hierarchy

The process of landfilling or residual management is the last but the most important part of integrated waste management. Ground water contamination problem was controlled in conventional landfill techniques but also various problems are still associated with these types of landfilling concept (Hettiaratchi,2007). The problem associated with the integrated waste management and first generation landfill is listed below:

- Requirement to find new space for landfilling. This landfilling concept is not considered as a sustainable solution.
- Long-term liability issue. It is necessary to monitor potential environmental impacts for a long time until the waste is fully stabilized.
- The first-generation landfill does not address the landfill gas issue explicitly.

2.5.1.3 Evolution of bioreactor landfill

The concept of bioreactor landfill was introduced by Pohland in the 1970s (Pohland, 1975) to ensure the rapid decomposition of waste, and reduce post-closure monitoring period which were the major drawback of the conventional landfill. The rate of degradation of waste is enhanced by controlled recirculation of leachate which helps in circulating nutrients and bacterial populations present in leachate itself (Reinhart and Townsend, 1997). The rate of production of methane increased by the addition of moisture thereby shortening the duration of waste stabilization which also reduced the monitoring period in the bioreactor landfill. The decomposition in bioreactor landfills occurs within 5-10 years which is much less than post-closure periods conventional landfills. But still, Bioreactor landfill do not resolve the most critical issues of the conventional landfill which are the requirement of new space for landfilling in every few years. To resolve this issue, a novel concept of sustainable landfill called Biocell landfill was proposed (Hettiaratchi, 2007)

2.5.2 Biocell landfill: A novel concept of waste processing

With the aim of conserving space, resource recovery, achieving faster degradation and enhancing gas production, the trend of conventional landfilling is evolving into bioreactors while bioreactors themselves are evolving into Biocell. Biocell is also known as sustainable landfill Biocell concept (Hettiaratchi et al., 2007). Biocell or Sustainable Biocell is operated in three sequential stages starting with anaerobic degradation followed by aerobic composting and then finally landfill mining. In the first stage, the Biocell works as an anaerobic bioreactor which helps in faster degradation of waste and it also enhances methane (CH_4) production. In the second stage, the Biocell is operated as an aerobic bioreactor. The aerobic biodegradation or aerobic composting is carried out till all the waste is decomposed. The third stage is landfill mining where the resources and space are recovered thus making the landfill operation sustainable (Jayasinghe, 2013). The Biocell is a holistic approach to waste disposal; some of the direct benefits include energy recovery, CH4 emission control, minimization of groundwater contamination, and space recovery (Jayasinghe, 2013).

2.6 Landfill gas generation

Landfill gas originates from the anaerobic decomposition of organic fraction of MSW in landfill. The organic fraction of MSW includes paper, food waste, textiles and leathers, and yard trimmings and wood. Landfill gas mainly constitutes of methane, carbon dioxides and water together with some traces of other gases. Methane, Carbon dioxide and oxygen fall under principal gases whereas all others fall into trace gases such as ammonia, sulfide, non-methane organic compounds etc. Waste decomposition, volatilization, and chemical reactions are the three-process involved in the production of landfill gases. Bacterial decomposition of waste occurred in five phases which are described the section 2-7 below. The typical landfill gas components in percentage are shown below in Table 2-3.

Table 2-3 Typical landfill gas components (Tchobanoglous et. al, 1993)

Component	Percent by Volume	Characteristics
Methane	45–60	Colorless and odorless naturally occurring gas.
Carbon dioxide	40–60	Colorless, odorless, and slightly acidic gas constituting approximately 0.03% of the atmosphere
Nitrogen	2–5	odorless, tasteless, and colorless gas constituting approximately 79% of the atmosphere
Oxygen	0.1–1	Odorless, tasteless, and colorless gas comprises approximately 21% of the atmosphere
Ammonia	0.1–1	Colorless gas with a pungent odor.
NMOCs (non-methane organic compounds)	0.01–0.6	Naturally occurred or formed by synthetic chemical processes (e.g. acrylonitrile, benzene, 1,1-dichloroethane, 1,2-cis dichloroethylene, dichloromethane, carbonyl sulfide, ethylbenzene, hexane, methyl ethyl ketone, tetrachloroethylene, toluene, trichloroethylene, vinyl chloride, and xylenes)
Sulfides	0–1	Naturally occurring gases with unpleasant odors like rotten-egg even at very low concentrations. (e.g. Hydrogen sulfide, dimethyl sulfide, mercaptans)
Hydrogen	0–0.2	Odorless and colorless gas.
carbon monoxide	0–0.2	Odorless and colorless gas

2.7 Waste degradation and its phases

Soon after waste is disposed in the landfill, the different physical, biological and chemical process takes place leading to the formation of landfill gases and leachate. The overall process of converting waste to gases and leachate through decomposition is known as waste degradation process. Waste degradation process is broadly divided into two stages: Aerobic and Anaerobic stage. The organic biodegradation of waste takes place aerobically due to oxygen present in the waste voids soon after the placement of waste in the landfill. The organic fraction of MSW starts to oxidize in the presence of aerobic bacteria to produce carbon dioxide and water vapor (Themelis and Ulloa, 2007). In the absence of oxygen in waste voids, the biochemical reaction will shift to the anaerobic stage. However, the process of biodegradation of waste by anaerobic microorganisms in the absence of oxygen, producing methane and carbon dioxide is called anaerobic degradation. The processes involved in anaerobic degradation of waste are presented in the Figure 2-5.

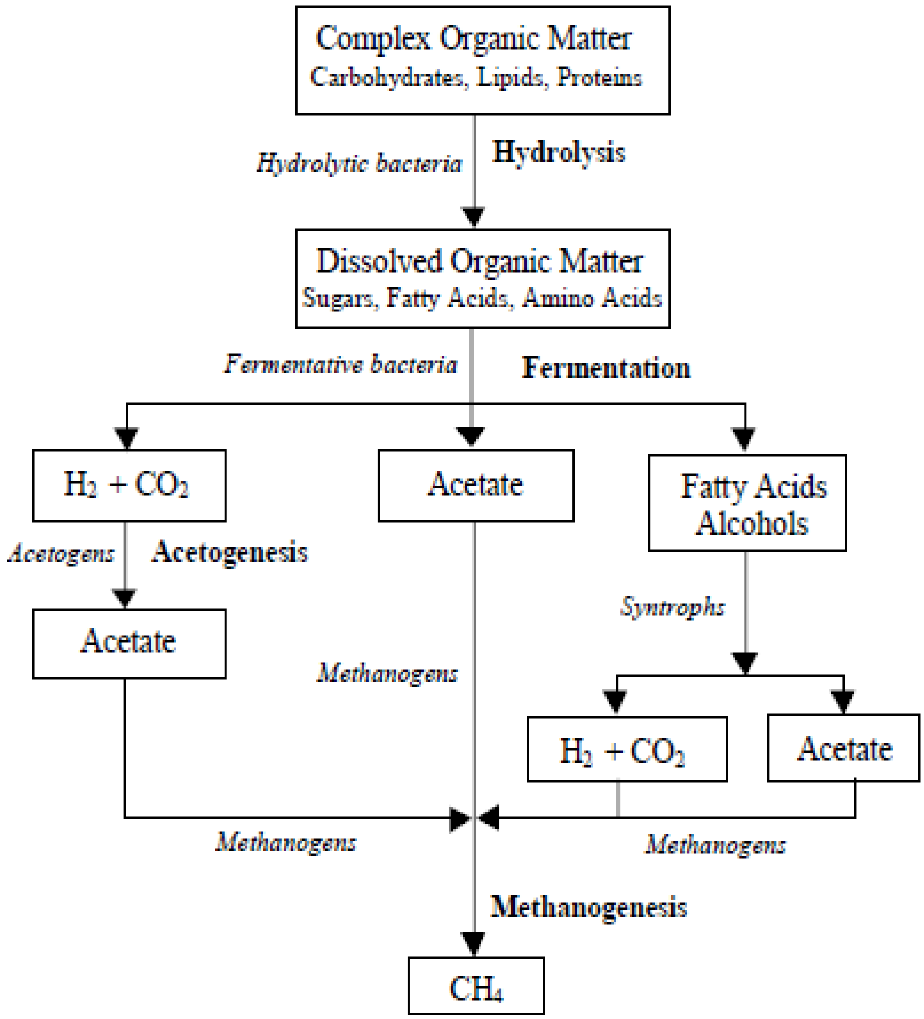


Figure 2-5 Simplified substrate degradation pathways in anaerobic waste degradation process (Jayasinghe, 2013)

Numerous studies have been conducted about the phases of waste degradation (Warith 2003; Warith et *al.,* 2005; White et *al.,* 2004; Zacharof & Butler, 2004; Barlaz et *al.,* 1989; Al-Kaabi, 2007; Kjeldsen et al., 2002 and Christensen et al., 1989). The bacterial decomposition of the waste in landfill is carried out in five sequential phases (Warith et *al.,* 2005). The rate and composition of the gas produced and leachate concentration changes with each of the five phases of decomposition, and reflect the microbially, mediated processes taking place inside the landfill waste (Reinhart and Al-Yousfi, 1996). Landfills accept waste for long periods of time, therefore waste may be experiencing several phases of decomposition at once which means older waste may be in the later phase of decomposition as compared to newly landfilled waste. The five phases of degradation of waste are shown in Figure 2-6 below (Warith et *al.,* 2005)

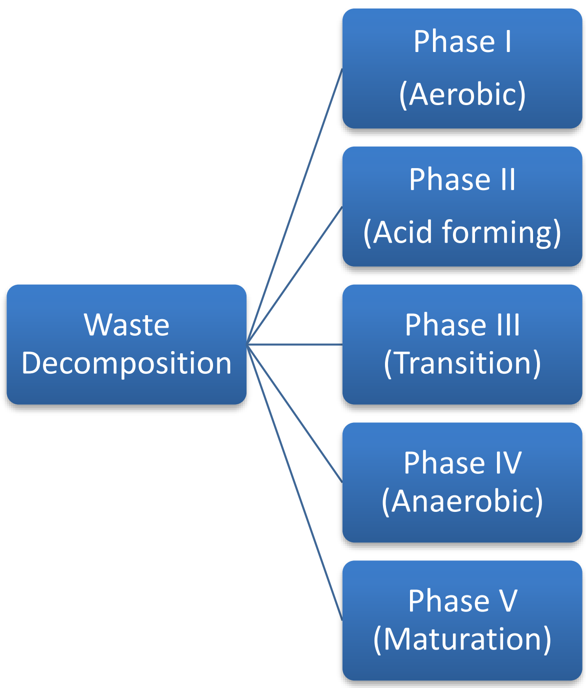


Figure 2-6 Phases of degradation of waste

Phase I: Initial adjustment phase (Aerobic phase): The first phase of waste degradation is an aerobic phase which occurs due to the presence of oxygen immediately after placing the waste. During this phase, aerobic bacteria utilizes oxygen to break down the long molecular chains of complex carbohydrates, proteins, and lipids that comprise organic waste. Carbon dioxide is the primary byproduct of this process. The duration of this phase depends upon the available oxygen in the waste voids. Aerobic phase could last from few days to few months depending upon the compaction level and available oxygen. An acclimation period/ initial lag phase is observed until sufficient moisture develops and supports an active microbial community (Reinhart and Al-Yousfi, 1996).

Phase II: Transition phase: With the depletion of trapped oxygen in this phase, anaerobic microorganisms become active thereby causing transformation from an aerobic to an anaerobic condition. The hydrolysis and fermentation of carbohydrates, fats, and proteins occur in this phase. The initial products of hydrolysis are soluble sugars, amino acids, long-chain carboxylic acids, and glycerol (Barlaz et al., 1990). The fermentative bacteria act on resulting monosaccharides forming carboxylic acids and alcohols. By the end of the transition phase, measurable concentrations of COD and volatile organic acids can be detected in the leachate.(Reinhart and Townsend 1997).

Phase III: Acid formation phase: During this phase, the hydrolytic, fermentative, and acetogenic bacteria becomes dominant. The accumulation of carboxylic acids occurs in this phase which results in the decrease of pH. The highest BOD and COD concentrations in the leachate are observed during this acidic phase (Barlaz and Ham, 1993; Reinhart and Grosh, 1998).

Phase IV: Methane fermentation phase: The methanogens and sulfate-reducing bacteria become active in this phase. The methanogenic bacteria transform acids that accumulated in the acid phase into methane and carbon dioxide. The rate of methane production increases constitutes approximately 50-60% (by volume) of gas composition (Barlaz, et al., 1990; Warith and Sharma, 1998). BOD and COD concentrations begin to decline and pH increases as acids are consumed (McBean et al., 1995).

Phase V: Maturation phase: The biodegradation of waste is stabilized and nutrients are limited in this phase. The rate of methane production decreases but the percentage of methane and carbon dioxide concentrations remains same as 60% and 40% respectively. The leachate strength remains steady with lower concentrations. The degradation of MSW can take 30 to 100 years in traditional landfill however the whole process is accelerated in Bioreactor and Biocell landfill with the recirculation of leachate and nutrients. (Heittarachi, 2007)

The phases of waste degradation shown graphically in Figure 2-7 ((UKDOE, 1993)

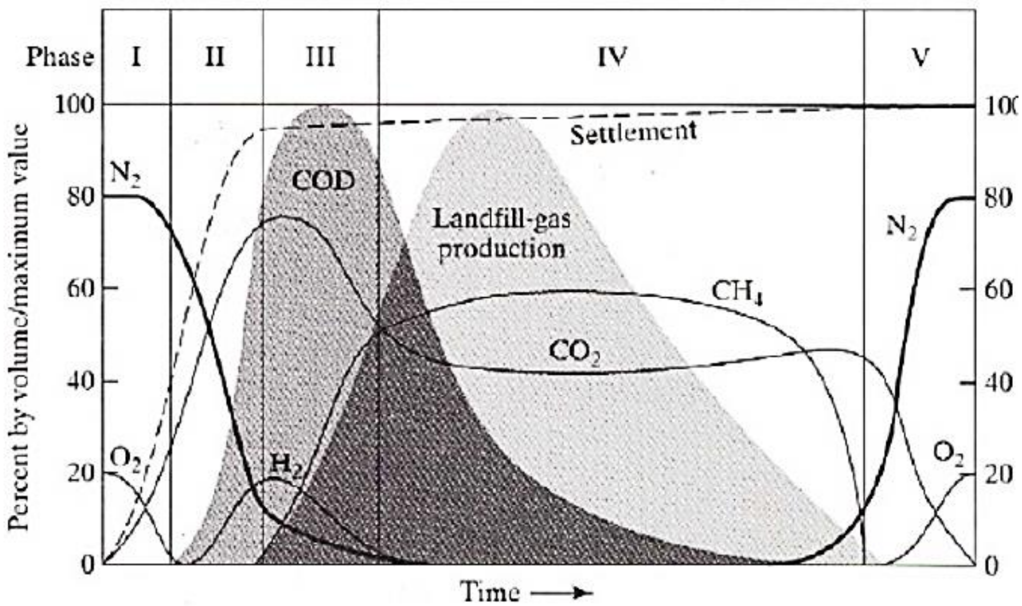


Figure 2-7 Phases of waste decomposition (UKDOE, 1993)

2.8 Factors affecting gas generation:

The final product of waste degradation is landfill gas. The rate and volume of landfill gas produced in each phase of degradation depend upon the characteristics of waste (e.g., composition and age of the refuse) and many environmental factors (e.g., oxygen availability, moisture content, and temperature). Waste composition, moisture content, pH, particle size, age of the refuse, temperature of the waste and oxygen availability are the factors controlling gas generation. (Barlaz et al., 1987; Barlaz et al., 1989, Christensen & Kjeldsen (1989); El-Fadel et al., 1996; Wraith, 2003; and Wraith et al., 2005).

2.7.1 Waste composition

The landfill wastes are composed of different types of waste components. As a result, the overall characteristics of the landfill is controlled by the waste composition. The waste composition varies with geographical conditions, depending on economic conditions, lifestyle, industrial structure and waste management techniques. Guermond et al. (2009) compared and summarized the composition of waste for various countries which is shown in Table 2-4.

Table 2-4 MSW composition for different countries (Guermond et al.,2009)

Country	City	Organics (%)	Cardboard (%)	Plastic (%)	Metal (%)	Glass (%)
Morocco	Agadir	65 – 70	18	2 – 3	5.6	0.5 – 1.0
Guinea	Labe	69	4.1	22.8(incl. textile)	1.4	0.3
Tunisia	Tunis	68	11	7	4	2
Jordan	Amman	63	11	16	2	2
Mauritania	Nouakchott	48	6.3	20	4.2	4
Turkey	Istanbul	36.1	11.2	3.1	4.6	1.2
Portugal		35.5	25.9	11.5	2.6	5.4
Greece	Palermo	31.7	23.1	11.8	2.7	8.3
Canada	Toronto	30.2	29.6	20.3	2.1	2
France	Paris	28.8	25.3	11.1	4.1	13.1

It is evident from Table 2-4, organic degradable waste constitutes the highest portion of total MSW generation. The percent of organic fraction in waste is higher for developing countries than developed countries (Guermond et al.,2009). The gas generation potential of MSW highly depends upon its composition. The higher percentage of organics corresponds to higher decomposition and loss of mass resulting in the higher landfill gas generation such as methane, carbon dioxide, nitrogen, and hydrogen sulfide. Again the presence of chemicals causes volatilization or chemical reaction producing non methanogenic organic compounds and other gases. Due to the heterogeneity of landfill waste composition, the landfill gas composition is also unpredictable.

According to Karanjekar (2013), the effect of waste composition on methane generation was studied and the result of the study is shown in Figure 2-8. The cumulative methane generation rate and volume versus time graph showed that 100% yard wastes showed a relatively early high peak and asymptotic decrease followed by 60% yard waste. The total amount of methane generated from reactors with 100% paper, 100% food and 100% textiles were low as compared to the other reactor. Minimum methane production in paper waste reactors could be due to the nutrient deficiency. The reactors with 100% and 60% food showed enhanced lag phase due to rapid hydrolysis and a late peak as compared to other reactors. Further, 100% textile waste reactors showed the lowest cumulative methane generation, with multiple and relatively low peaks. However, the reactors with 20% each (paper, food, textiles and inorganics) showed enhanced methane generation rates due to the presence of a mixture of wastes supplemented nutrients.

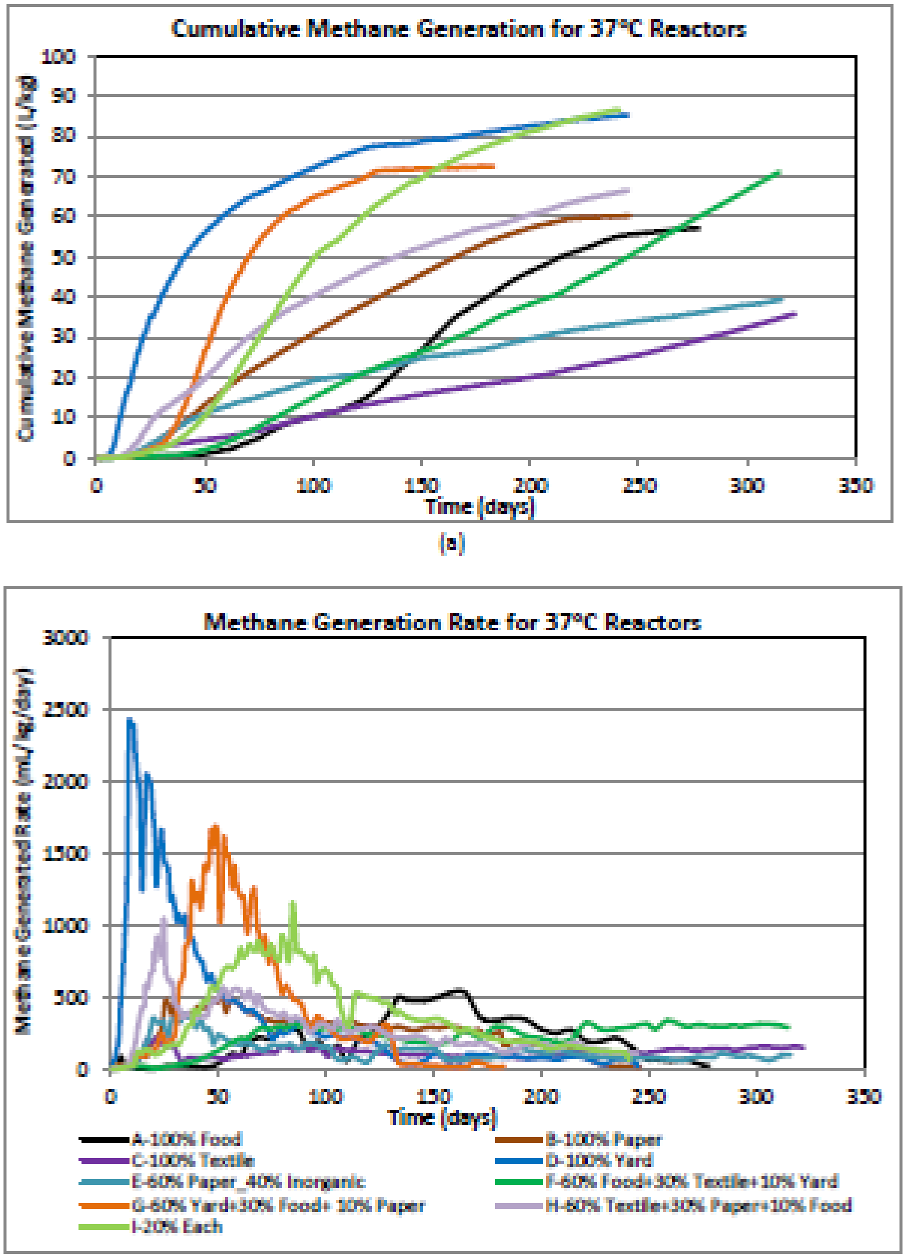


Figure 2-8 (a) Cumulative methane versus time ;(b) Methane generation rate versus time

(Karanjekar, 2013)

2.7.2 Moisture content

Moisture content is considered the most important factor which plays a significant role in waste microbial activity and decomposition. The moisture inside the landfill acts as a medium to transfer the nutrients, microorganism, and to provide the favorable environment for gas production and waste decomposition. The dry waste takes longer for stabilization as compared to the moist waste. Several studies have concluded that methane generation rate increases with an increase in moisture content (Barlaz et al., 1990; Mehta et al., 2002; Rees, 1980; Christensen et. al., 1996; Warith et al., 2005). The increase of moisture content controls the oxygen transport from the atmosphere; facilitates the exchange of substrate, nutrients, buffer, and dilution of inhibitors; and distributes the micro-organisms inside the landfill (Christensen et. al.,1996; Warith et al.,2005). Rees (1980) concluded that with the increase of water content from 25% to 60%, the percentage of methane and the gas production rate are also increased which is shown in Figure 2-9 and Figure 2-10. Rees (1980) also showed the effect of moisture circulation on wastes and reported that maximum methane content is generated by daily circulation of water which resulted in increased gas generation.

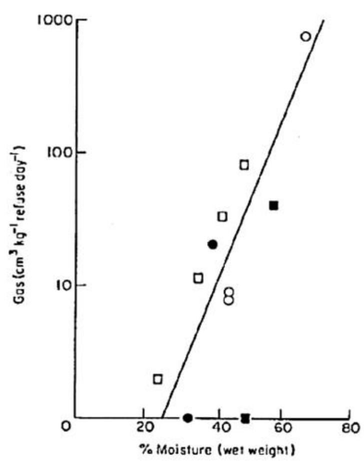


Figure 2-9 Effect of moisture content on gas generation rate (Rees, 1980)

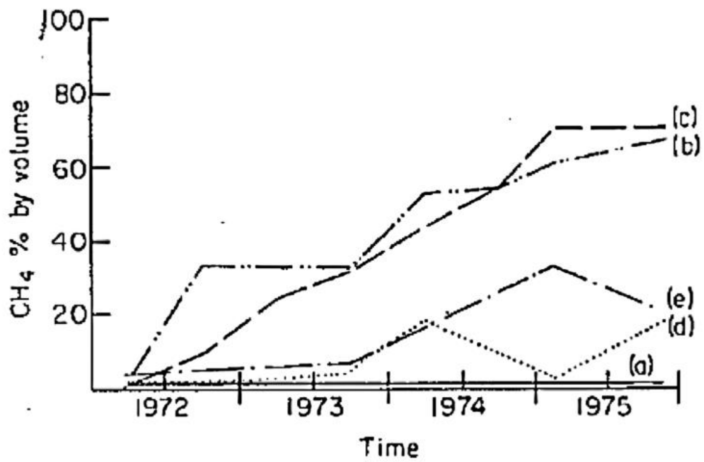


Figure 2-10 Effect of moisture content on methane generation a) Dry condition; b) & c) Every day liquid application; d) & e) Initially saturation (Rees, 1980)

Another study on two cells with and without controlled moisture addition was conducted by Mehta et al (2002) which showed increased gas generation with the increase in moisture content as shown in Figure 2-11.

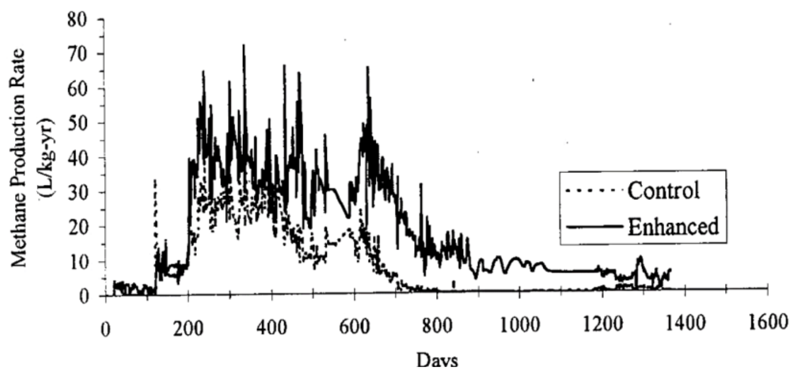


Figure 2-11 Comparison of methane production rate between cells without moisture addition (control) and with moisture addition (Enhanced) (Mehta et al., 2002)

2.7.3 pH

The pH is another crucial influencing factor for biodegradation as well as gas generation from the solid waste. It affects the microbial activity predominantly. Optimum pH for the wastes depends on the phase of the involved bacterial activities. The acidogenic bacteria can survive in comparatively higher pH range than the methanogenic bacteria. The optimum pH range for the gas production is 6.8 – 7.4 and the methanogenic bacteria are most effective at pH value of 7.0. (Warith, 2003; Wraith et al., 2005). The bacterial activity gets slower at pH value below 6.8 or above 7.4. To enhance the biodegradation of MSW, pH of the leachate before recirculation should be neutralized with buffer solution.

2.7.4 Temperature

The temperature of waste plays an important role in the gas generation as temperature controls bacterial activity. With the increase of temperature in mesophilic and thermophilic conditions, the bacterial activities accelerate (El-Fadel et al., 1996). According to the laboratory experiments by Christensen and Kjeldsen (1989), the optimum temperature varies between 30°C to 40°C in mesophilic conditions and between 50ºC to 55ºC in thermophilic conditions. Tchobanoglous et al. (1993) found that below 20°C and above 70°C methane generation decreases abruptly. The methanogenic bacteria become inactive at low temperature as compared to higher temperature. Buivid et al. (1981) conducted experiments on laboratory grade simulated reactors to observe the effect of temperature on waste decomposition by choosing 25°C, 37°C, and 60°C and reported a temperature of 37°C is the most favorable for the enhanced methane generation.

2.7.5 Particle size of waste

The size of waste particles also has the effect on the degradation of waste. The more homogeneous waste can be obtained by reduced particle size than unprocessed waste without shredding. Sponza et al. (2005) concluded that the shredding has a positive impact on the waste degradation. Three types of reactors were constructed among which one was filled with raw waste, the second was filled with shredded waste, and the third was filled with compacted waste. At the end of the experiments after 57 days, it was found that the reactor with shredded waste had the lowest COD and VFA. According to the study conducted by Ham and Bookter (1982) showed that the rate of decomposition and methane production increases with the shredding of waste. The shredded and well-mixed waste provides greater contact between the key refuge particles (moisture, substrate, and microorganisms) required for methane production (Barlaz et al. 1990). Therefore, the shredding of waste could lead to rapid oxygen utilization, increased rate of waste decomposition, and leads to early methane production (Ham and Bookter 1982; Otieno, 1989).

2.7.6 Oxygen content

Soon after the disposal of waste in the landfill, the voids of waste will be filled with atmospheric air which acts as the source of oxygen. Aerobic phase, which is the first phase of degradation, lasts till the oxygen is available for aerobic bacteria. The aerobic phase in the landfill refuse prevents the formation of methane in these layers (Christensen and Kjeldsen (1989). Methanogenic bacteria are sensitive to the presence of oxygen. Therefore, the methane generation started with the end of aerobic phase as anaerobic bacteria becomes prominent. The oxygen content can be reduced by optimum compaction which ensures minimum air intrusion.

2.9 Factors enhancing gas generation process

The gas generation process is directly related to the waste degradation process which depends upon several factors. The re-circulation of produced leachate, addition of nutrients and inoculum are the factors which help in faster degradation of waste as well as accelerate gas generation. (Barlaz et al., 1990; Warith et al., 1999; Alkaabi et al., 2009)

2.9.1 Leachate Recirculation and moisture addition

Leachate recirculation is the process of recirculating the leachate which is produced during the waste degradation process. It is the most effective technique which helps to maintain the suitable environment for microorganism by increasing moisture content inside landfill in a controlled fashion. According to El-Fadel (1999), the primary process in a bioreactor landfill is to recirculate the produced leachate back to the landfill in order to increase moisture content which enhances the biodegradation of organic fraction of MSW. The advantages of leachate recirculation include distribution of nutrients and enzymes, pH buffering, dilution of inhibitory compounds, recycling and distribution of methanogens, liquid storage and evaporation opportunities (Reinhart, 1996). The duration of landfill waste stabilization could be reduced from several decades to 2 to 3 years thereby reducing long term environment effects due to leachate recirculation (Pohland 1975).

San and Onay (2001) conducted a study with two set of reactors, with and without leachate recirculation. They found that waste stabilized faster and removal of COD was faster in the reactors with leachate recirculation shown in Figure 2-12.

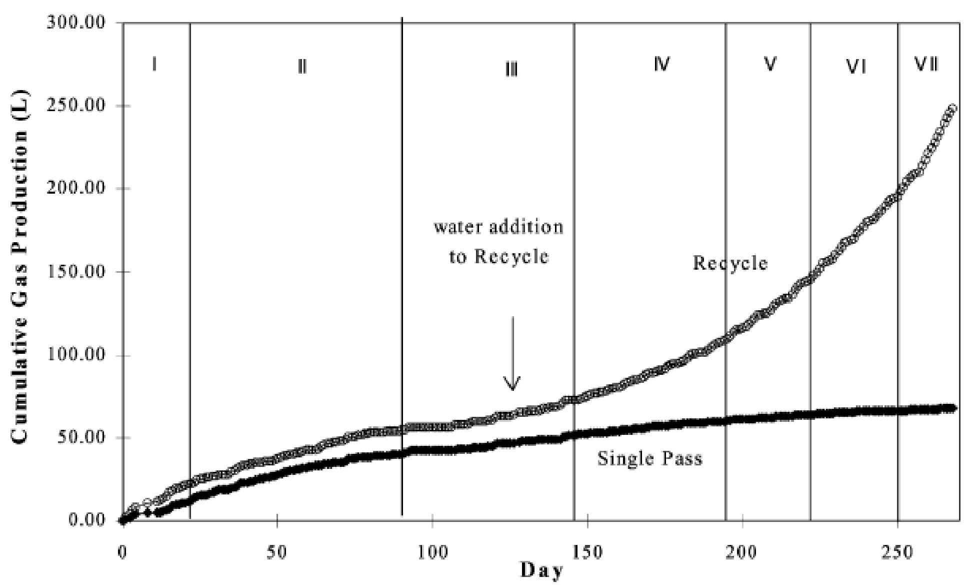


Figure 2-12 Cumulative gas production for the single pass and recycle reactors (i.e. without or with leachate recirculation) San and Onay (2001)

According to the study conducted by Mehta et. al. (2002) at Yolo County, CA results in both higher methane yield and increased settlement due to recirculation of leachate. The comparison between two 8,000 metric ton test cells was performed to investigate the effects of leachate recirculation. Figure 2-13 shows the comparison of methane production rate and settlement with the volume of leachate recirculated and water addition in enhanced and control cells.

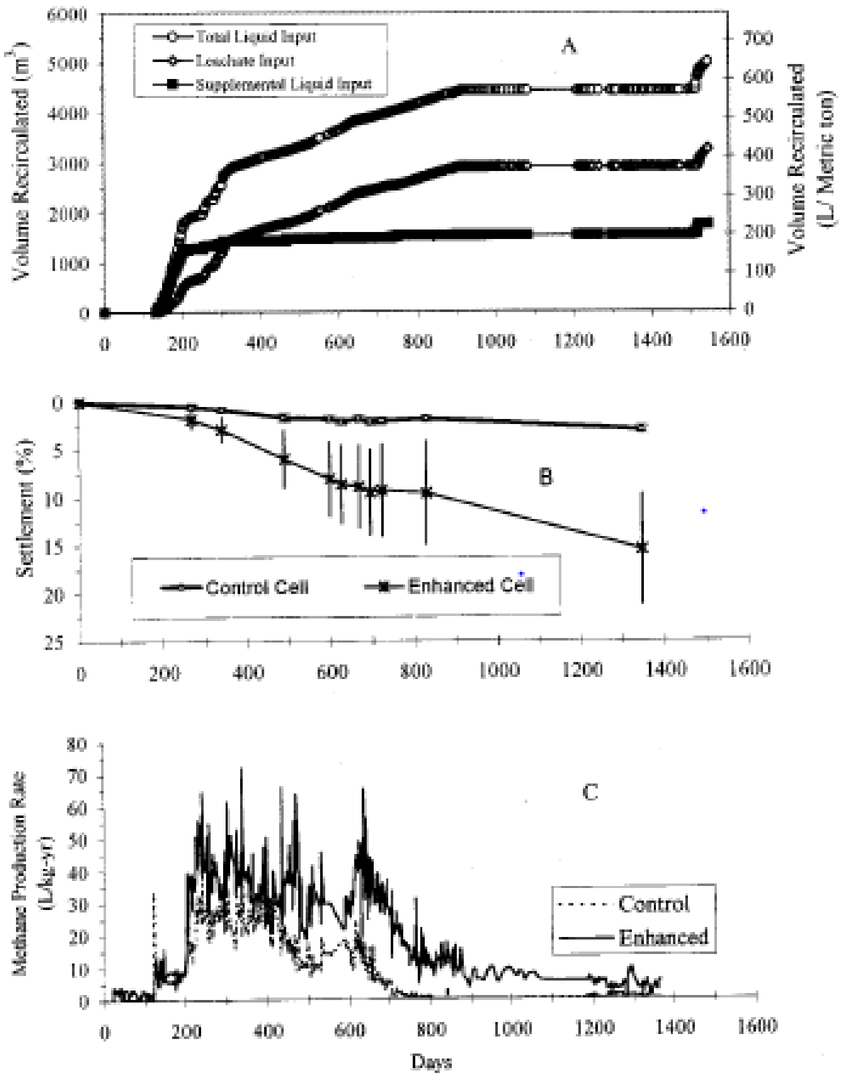


Figure 2-13 a) Cumulative liquid input for recirculation of leachate and supplemental liquids; b) refuse settlement over time; and c) methane production rate in enhanced and control cells (Mehta, 2002)

Leachate recirculation can accelerate methane generation from landfills. (Chan et al., 1998). According to Morris et al. (2003), leachate recirculation has the tremendous impact on subsequent waste stabilization due to degradation in landfills.

A study conducted by Chan et.al. (2002) has shown that leachate recirculation was effective in enhancing the degradation rate of the waste, and gas production which is shown in Figure 2-14.

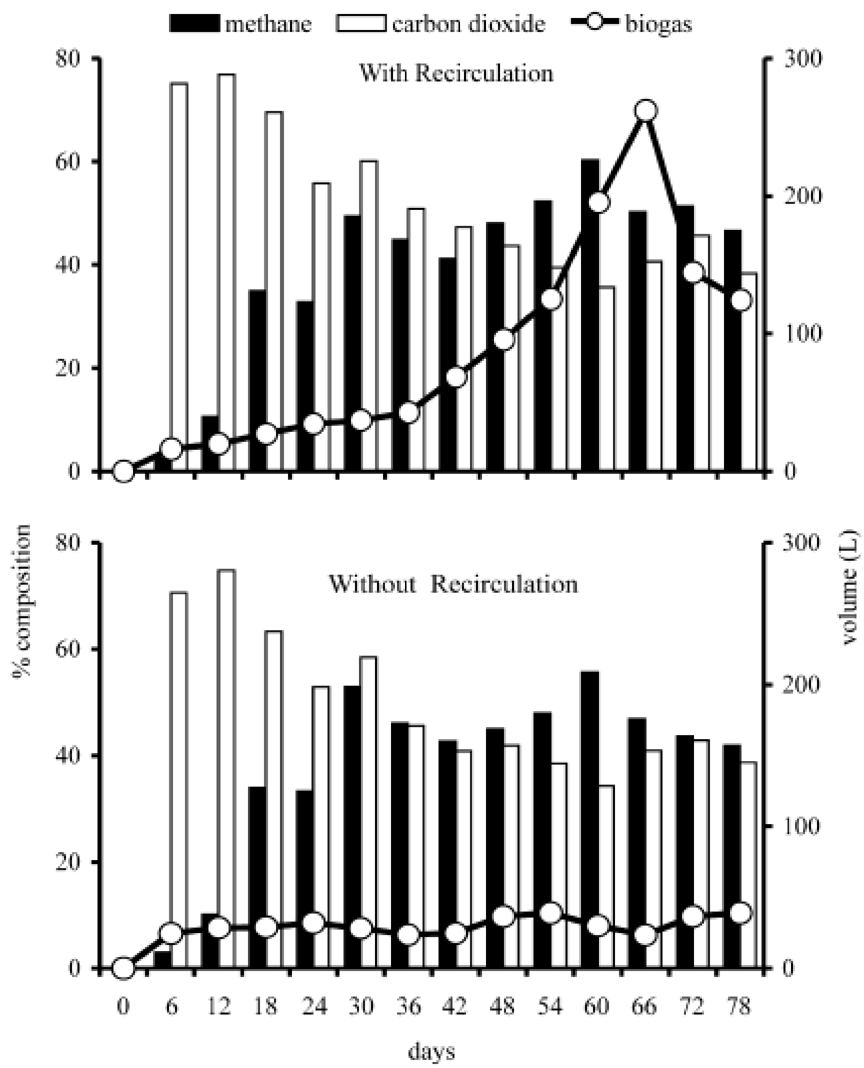


Figure 2-14 Gas production rate and composition with and without leachate recirculation.

Chan et.al. (2002)

2.9.2 Addition of inoculum

The duration of bio-stabilization of organic waste also depends upon the addition of inoculum. The utilization of inoculum decreases the stabilization time of organic waste. The inoculum used in previous studies that shows satisfactory results are sewage treatment sludge or some materials from animal origins, such as bovine manure and others (Craveiro, 1982; Leite, 1997; Lopes, 2000; Callaghan et. al., 2002; Lopes et. al., 2003; Sosnowski et. al, 2003).

The cellulose and lignocellulose enzyme also provided the satisfactory result in enhancing the waste degradation. (Lagerkvist and Chen, 1993; Sah, 2006; Cirne et al., 2008, Jayasinghe et.al., 2013). The old landfill leachate can also be used as inoculum to young landfill. (Erses & Onay, 2003). Among these techniques, the addition of sludge is shown to be the most common and oldest practice.

2.10 Use of sludge as inoculum

The addition of sludge for enhancing the degradation of organic fraction of MSW is the most common and oldest practice. Wastewater treatment sludge or sewage sludge is rich in the microorganism which is responsible for degrading waste. The effect of sludge addition on the MSW biodegradation is covered by Leuschner (1982), Pacey (1989) and Warith (2002). Significant increase in pH and decrease in biological oxygen demand with time was observed in the case of reactors with sludge which indicates the enhancement of waste degradation and gas generation (Warith, 2002). The addition of anaerobically digested sludge has the positive effect on the degradation of waste and can serve as a seed to microorganisms as well as the source of nitrogen, phosphorous, and other nutrients. (Warith 2005). The addition of sludge also increases the moisture content of waste thereby creating suitable environment for microorganism

According to Buivid et. al., (1981) MSW mixed with 10% of anaerobically digested waste water sludge produced three times more methane than with primary sludge after 90 days. A study was conducted by Gulec et al. (2000) by building and operating 10 L laboratory-scale batch digesters filled with 2 year old MSW at ratios of 1:9, 1:6 and 1:4 of anaerobically digested sludge to waste on the wet basis. The result shows that pH of leachate ranged from 7.0 to 8.5 in the reactor with sludge while sharp drop in pH levels to the acidic range occurred in the control reactors without sludge addition. The addition of sludge acts as the buffer and increases alkalinity as well. The higher sludge to waste ratio yielded higher methane recovery rates, due to faster removal of decomposable organic material.

Alkaabi *et al.* (2009) showed that the addition of sludge under saline conditions enhances the biodegradation of MSW. Two sets of reactors with different concentration of salt, with or without sludge, were built to study the effect of sludge and saline water on waste degradation. The methane yield was about 14% more in the bioreactors with sludge addition at different salt concentrations compared to the bioreactors operated without sludge addition. The percentage reductions of oxygen demands were higher in the bioreactors operated with sludge addition and showed a significant decrease with increasing the salt concentrations which indicates the faster degradation of waste. R1, R2, R3, and R4 are the reactors with different concentration of salt 0, 0.5, 1 and 3% (w/v) without sludge respectively shown in Figure 2-15 and R5, R6, R7, and R8 were operated with the addition of sludge and with salt concentrations of 0, 0.5, 1 and 3% (w/v), respectively shown in Figure 2-16.

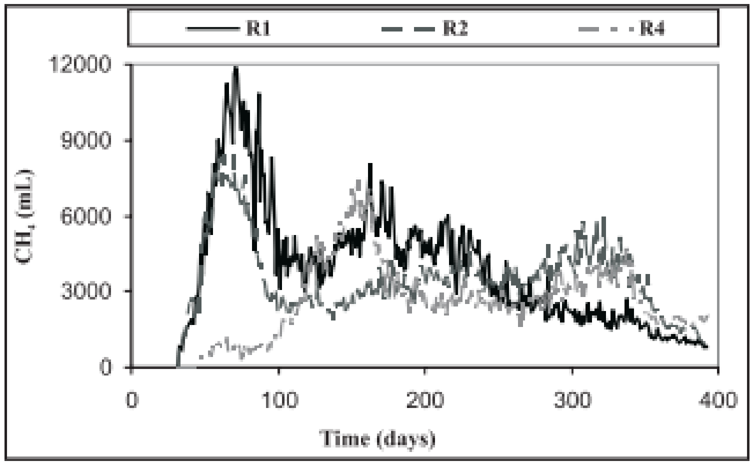


Figure 2-15 Cumulative methane generation with time without sludge addition (Alkaabi *et al.,* 2009)

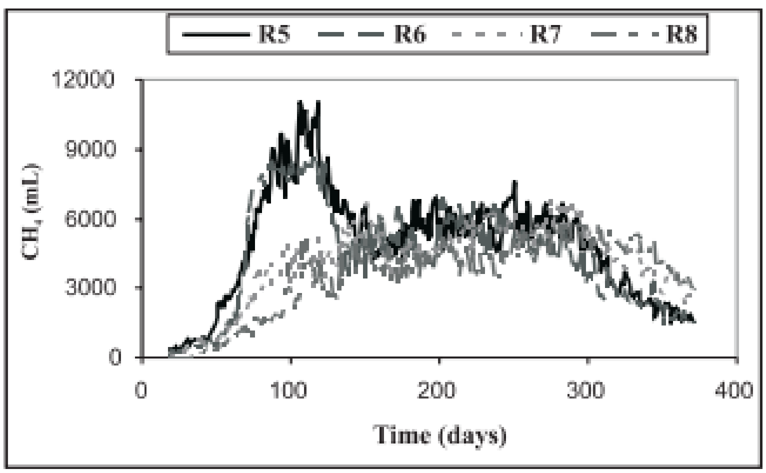


Figure 2-16 Cumulative methane generation with time with sludge addition (Alkaabi *et al.,* 2009)

The study conducted by Bae et. al. (1998) showed that the total amount of produced CH_4 with sludge recycle was 170 and 78 times greater than that from control and from leachate recycle. The leachate recycles began from 45 days after the operation at the rate of 150 ml/d and for digester sludge containing active methanogens were recycled at the rate of 80-150 ml/d. A cumulative gas production and gas production rate with time for control reactors without leachate recirculation and sludge addition (L-control), reactors with leachate recirculation (L-leachate) and reactors with addition of digester sludge only (L-sludge) shown in Figure 2-17 and Figure 2-18 respectively.

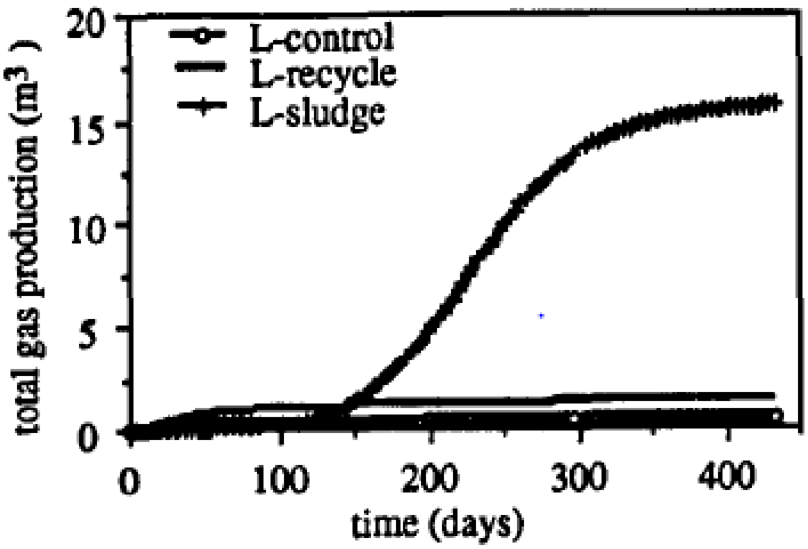


Figure 2-17 Cumulative gas production (Bae et. al., 1998)

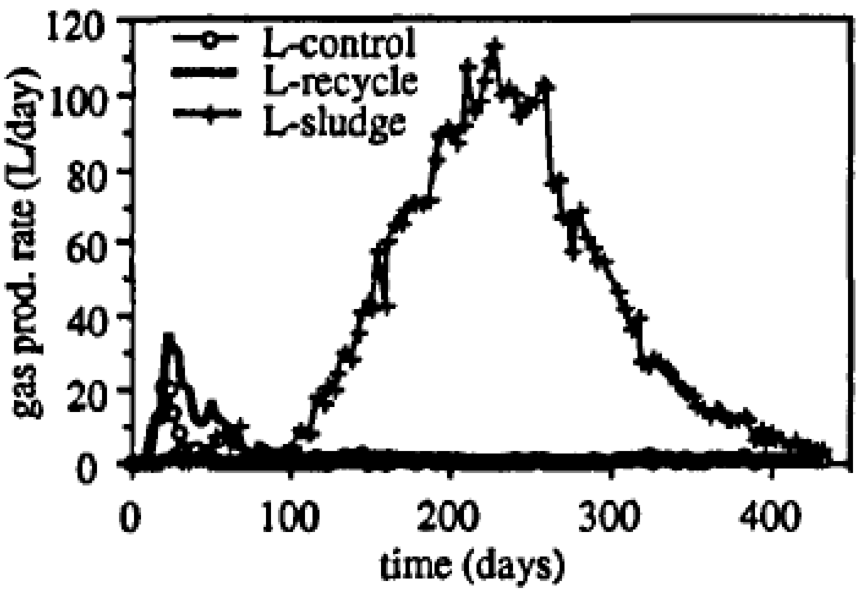


Figure 2-18 Gas production rate (Bae et. al., 1998)

2.11 Use of enzyme as inoculum

Enzymes are macromolecules, mostly protein, that occur in all living organisms. They are biological catalysts or assistants that can either launch a reaction or increase the reaction rate by decreasing the activation energy of the particular reaction. The augmentation of leachate by the addition of enzyme increases the gas generation rate and volume. Studies conducted using cellulose and lignocellulosic enzyme on fresh as well as degraded MSW has proved to increase the gas generation. (Lagerkvist & Chen, 1993; Sah, 2006; Jayasinghe et. al., 2013)

According to the study conducted by (Lagerkvist & Chen, 1993), the use of cellulose enzyme called Econase enhanced degradation of waste as compared to the control cell in laboratory column experiments. The observed conversion of cellulose was found to be 42-70% in cells with enzymes addition while just 29% in cells without enzyme addition and the conversion of VS was found to be 40% to 50% in enzyme added cells.

Sah (2006) studied the effect of Soyabean Peroxidase enzyme addition on waste paper degradation. The columns with enzymes showed faster decomposition than the control column. The results obtained from reactors with enzymes were compared with reactors with biosolids. The COD and BOD reduction efficiency were about 2 times and 1.6 times higher in the reactor with enzyme than with biosolids.

The study conducted by Jayasinghe et. al. (2013) showed that addition of Manganese peroxidase (MnP) enzyme generated more methane as compared to lignin peroxidase (LiP) and soybean peroxidase (SbP). The hydrogen peroxide is required to activate these enzymes. The maximum methane yield from laboratory batch experiment was at enzyme dose of 0.3 mg with the enzyme to hydrogen peroxide ratio of 0.0046 for MnP to 2 gm of degraded waste shown in Figure 2-19.

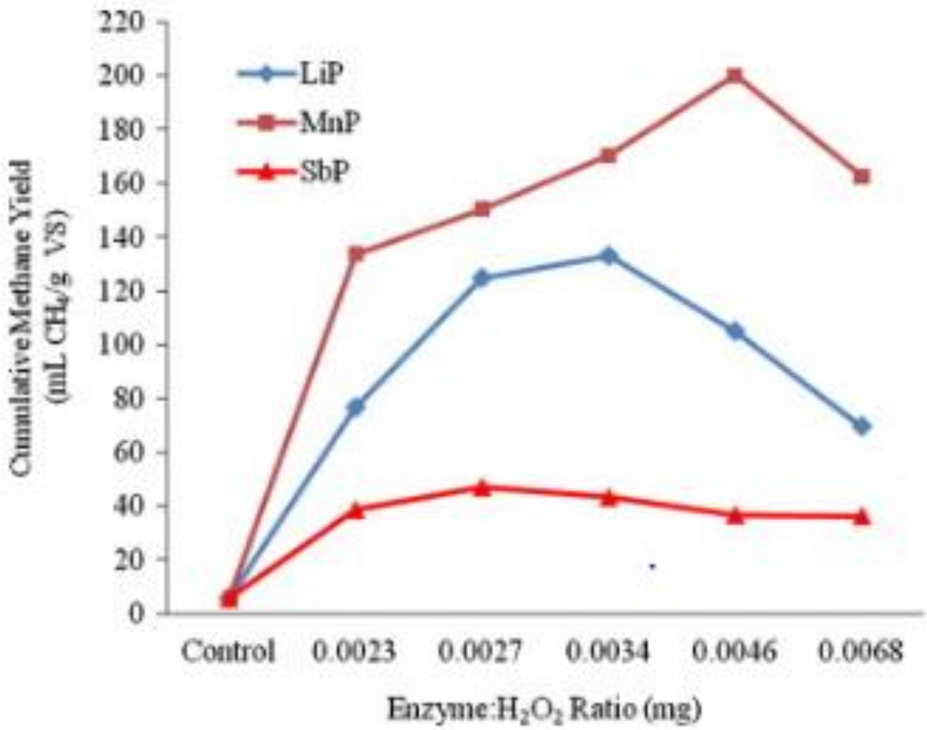


Figure 2-19 Effect of enzyme:H_2O_2 ratio for different enzymes at 0.3 mg enzyme dose. (Jayasinghe et. al.,2013)

For field scale, Calgary Biocell, Jayasinghe (2013) recommended 1024 gm of MnP enzyme for 48000 tons of MSW which are 0.00000214% of waste on the wet weight basis.

2.12 Livestock manure

With the increase in population, the production and concentration of intensive livestock operation also increase. The increment in the production and operation of livestock have resulted in a greater awareness and concern for the proper storage, treatment, and utilization of livestock manure. Livestock manure is also one of the sources of the greenhouse gas emissions. Animal manures, stored indoors in sub-floor pits or outdoors, are also relevant CH_4 sources since conditions usually favor methanogenesis in both slurry and solid manure heaps (Husted, 1994). According to USEPA (2006), manure contributes 4% of all anthropogenic methane generated by various sources. The quantity of manure is expected to rise sharply in Asia, Africa, and America because of increase in the population. The manure can be diverted from the landfill which not only reduces the environmental pollution but also helps to enhance degradation of the organic fraction of MSW. The anaerobic degradation potential of manure depends upon the availability of nutrients and microorganism suitable for waste degradation. Types of animal waste, methane generation potential, and quantity of livestock manure produced are the important factors for selecting different types manure for inoculum.

2.12.1 Types of animal waste in anaerobic digestion

The source of animal waste used in anaerobic digestion is important in ensuring a successful operation of the process because nutrients, microbes and lignin components present in animal manure depends upon the digestive physiology and their feeding habits (Yusuf et. al., 2011). Animals are usually classified into two groups based on their digestive physiology: Ruminants and Monogastric (or non-ruminants). Ruminants animals are always herbivorous in nature whereas Monogastric animals are further classified based on food intake: - Herbivorous, Omnivorous and Carnivorous. Typical classification based on digestive physiology is shown in Figure 2-20.

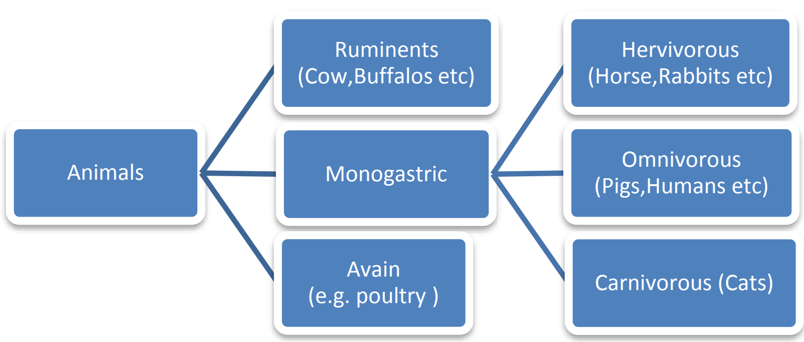


Figure 2-20 Typical classification of animals based on digestive physiology

Monogastric animals are known to produce wastes that contain more nutrients than ruminants. Ruminants are known to excrete more lignocelluloses material due to extensive enzymic exposure in their four-chamber stomach (Wilkie 2005). The volatile solid present in cattle manure is lower as compared to monogastric animals (Wilkie 2005). The volatile organic content of manures directly related to its gas generation potential.

2.12.2 Methane generation potential of manures

The methane generation potential of manure depends upon the digestion of the organic fraction in the feces which is mainly: carbohydrates, proteins, and lipids. The ultimate methane yield in terms of volatile solid content (VS) is considerably higher in pig manures (356 L kg^{-1} VS) and swine manures (275 L kg^{-1} VS) than in dairy cattle manures (148 L kg^{-1} VS) (Moller et. al., 2004). Maximum gas production from different animal waste is shown in Table 2-5.

Table 2-5 Maximum biogas yields from various types of manure (Deublein and Steinhauser, 2008; NAS, 1977; Metcalf & Eddy, 2003; Sattler, 2011)

Substrate	Dry Matter (DM, %)	% of Dry Matter that is organic (oDM)	Biogas Yield (m^3/kg of oDM)	Hydraulic Retention time (days)
Manure from cows	7-20	85-90	0.20-0.50	28-38
Manure from pigs	5-27.5	90	0.56	22-28
Manure from horses	--	--	0.2 – 0.3	--
Manure from poultry	15-75	75	0.31-0.54	17-22
Manure from sheep	--	--	0.37-0.61	20

* % of total that is, organic

2.12.3 Selection of manures

The manure can be diverted to the landfill which not only reduces the environmental pollution but also helps to enhance degradation of the organic fraction of MSW. Different types of manure can be used as inoculum for degrading organic fraction of MSW in Biocell. Based on the type of animal source, availability, quantity of production and methane generation potential, different type of manures were selected for this study. Table 2-6 shows the criteria for selecting different types of manures:

Table 2-6 Selection criteria for manure as inoculum

Type of manures	**Animal Type based on digestive physiology**	**Biogas Yield (m3/kg of dry matter)**	**Availability**
Cow Manure	Ruminants	0.20-0.50	Readily available
Sheep Manure	Ruminants	0.37-0.61	Readily not available
Poultry Manure	Avain	0.31-0.54	Not enough quantity
Horse Manure	Mono-gastric Herbivorous	0.2 – 0.3	Readily Available
Pig Manure	Mono-gastric Omnivorous	0.56	Readily Available
Cat Manure	Mono-gastric Carnivorous	N/A	Not enough quantity

2.12.4 Use of manure as Inoculum

Livestock manure is rich in nutrients nutrient (Carbon, nitrogen, phosphorus etc.) as well as the microorganism, necessary for the growth of the plant. Therefore, from centuries it has been used as a fertilizer for improving agricultural lands. Several studies conducted on anaerobic digestion of waste with manure showed that the use of manure enhanced the waste degradation process and gas generation (Ali et. al., 2016; Yazdani, 2010). Accumulation of volatile fatty acid is common in organic waste which inhibits the gas generation thereby leading to lag phase. The lag phase is the duration in the beginning of waste degradation process in which there is no gas generation. The use of manure inoculum, rich in nutrients as well as the microorganism, enhances the buffer capacity creating an environment to neutralize the pH to some extent and reduces the inhibition time or lag phase (Zhang et al., 2013). Zhang et. al. (2013) showed that methane generation increases significantly with the addition of manure in food waste as shown in Figure 2-21.

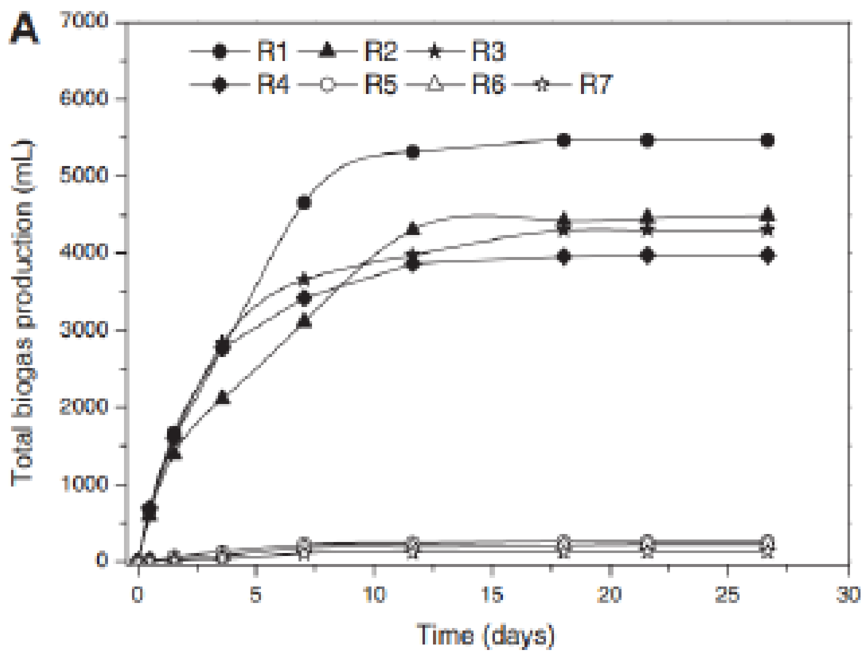


Figure 2-21 Total biogas production with (R1, R2 and R3) or without (R4, R5, R6 and R7) manure addition (Zhang et al., 2013)

Another study conducted by Macias-Corral et. al (2008) showed that manure used as inoculum for degrading organic fraction of MSW enhanced the generation of gas as compared to MSW The percentage of cattle manure used in this study was 9.89% on wet weight basis. The digestion of cattle manure(CM) and Organic Fraction of Municipal Solid Waste (OFMSW) alone resulted in 62 m^3 methane/ton of CM on the dry weight basis and 37 m^3 methane/ton of dry waste respectively. Co-digestion of OFMSW and CM resulted in 172 m^3 methane/ton of dry waste. Co-digestion of Cotton Gin Waste (CGW) and CM produced 87 m^3 methane/ton of dry waste. Comparing the result, the co-digestion of

OFMSW with 9.89% of cattle manure found to be the best combination for enhancing waste degradation process which is shown in Figure 2-22.

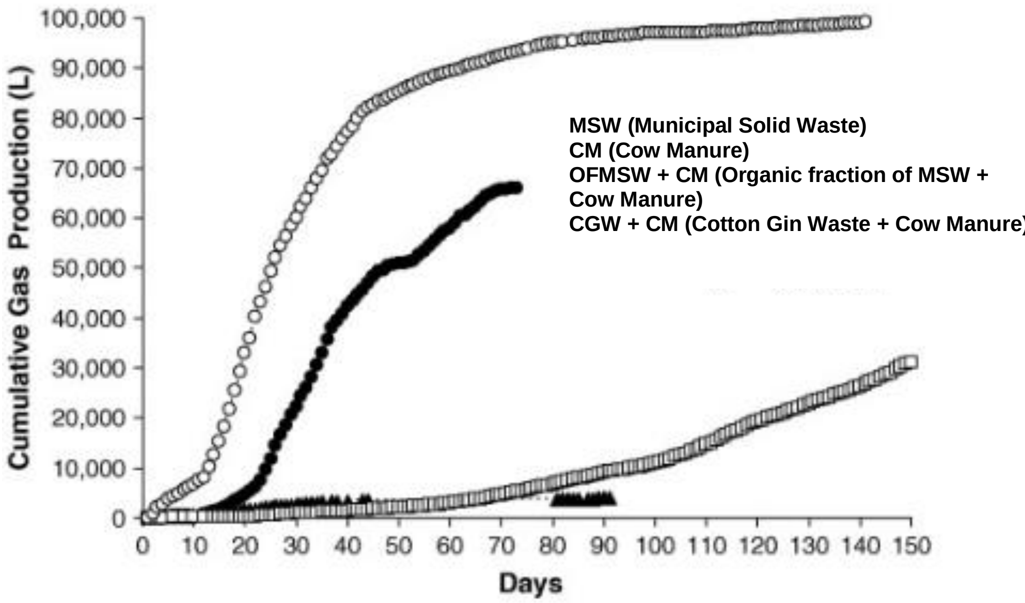


Figure 2-22 Cumulative gas generation with time (Macias-Corral et. al., 2008)

The organic waste in landfill or anaerobic digester goes through a degradation process similar to the digestive tract of ruminant animals like dairy cow (Hungate, 1996; Bryant, 1979) The digestion of cellulose and polysaccharide occurs in rumen where anaerobic and methanogenic microorganisms exists which convert glucose into acetate and acetate into methane and carbon dioxide respectively. (Hungate, 1996; Bryant, 1979; Brock, 2002). Therefore, manure of ruminant or bovine rumen fluid can be used as inoculum for faster degradation of the organic fraction of MSW. According to the study conducted by Lopes et. al. (2004) on the degradation of organic fraction of MSW by using bovine rumen fluid inoculum, the applied mass of feedstock substantially reduced thereby improving the performance of the process. Four reactors: reactor A, reactor B, reactor C, and reactors D were built and operated with organic fraction of MSW to bovine rumen ratio of 0%, 5%, 10% and 15% respectively. Total weight of feedstock in the reactors was always kept constant of 10 kg. The biogas accumulated versus time is shown in Figure 2-23.

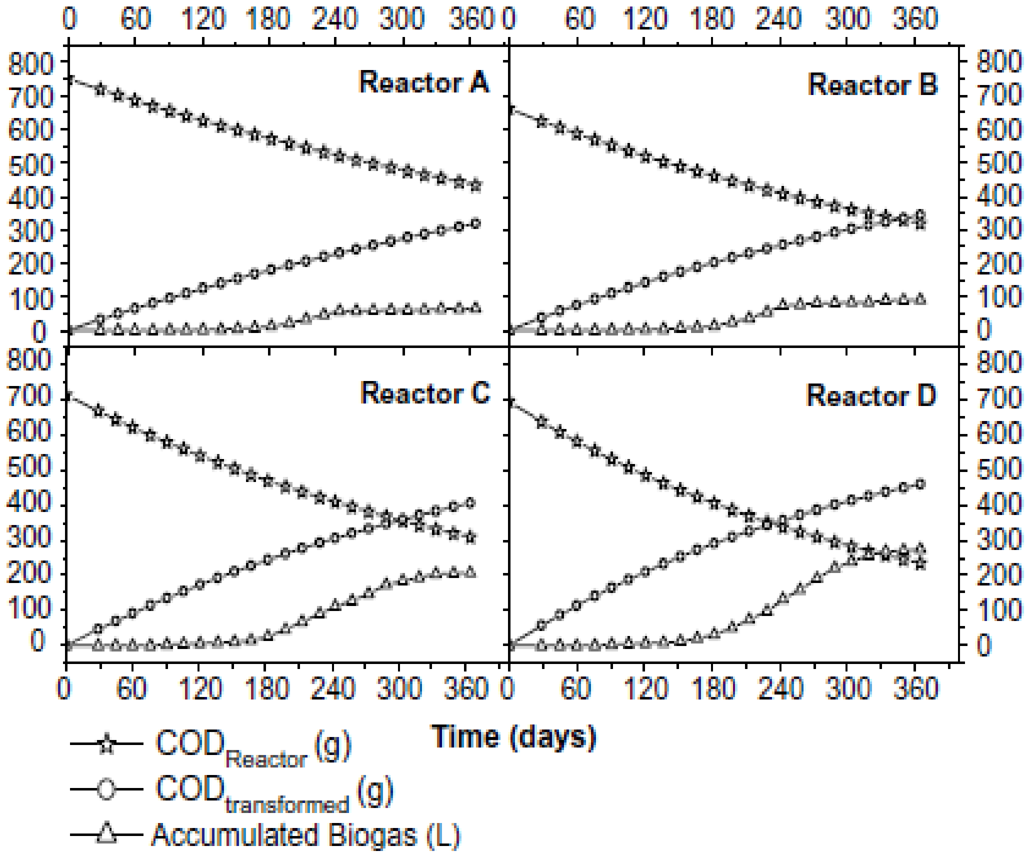


Figure 2-23 Accumulated gas and COD versus time (Lopes et. al., 2004)

In the study conducted by Ali et. al. (2016), cow dung is used as an inoculum for degrading organic municipal waste. The percentage of cow dung used in the reactors Rc, R1, R2 and R3 were 0%, 10%, 20% and 30% respectively. The biogas yields at the end of the digestion from the reactors Rc, R1, R2 and R3 cow dung as inoculum were 173 ml / g VS, 337.365 ml / g VS, 481.95 ml / g VS and 567ml / g VS respectively. Figure 2-24 shows the cumulating biogas generation with time.

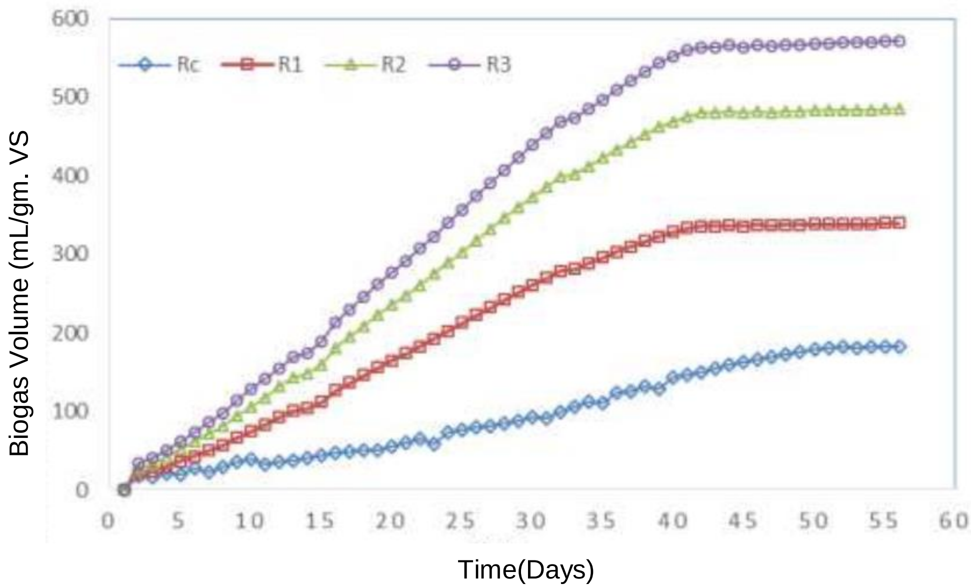


Figure 2-24 Cumulating biogas generation with time. (Ali et. al., 2016)

According to the study conducted by Yazdani (2010) at the Yolo County Central Landfill in California, digester cell was operated in the two stage batch system where the organic matter was degraded under anaerobic conditions followed by aerobic conditions. 1,718 Mg Green waste was mixed with 118 Mg of aged horse manure as an inoculum and 31 Mg of wood chips as part of a base gas collection system, and 23 Mg of limestone as a buffer. During the anaerobic phase of 451 days, the total volume of biogas generated was 1.48 x 10^5 m^3 and the methane yield was 6.07 x 10^4 m^3, or 27.2 m^3 of CH_4 per Mg of solids is shown in Figure 2-25.

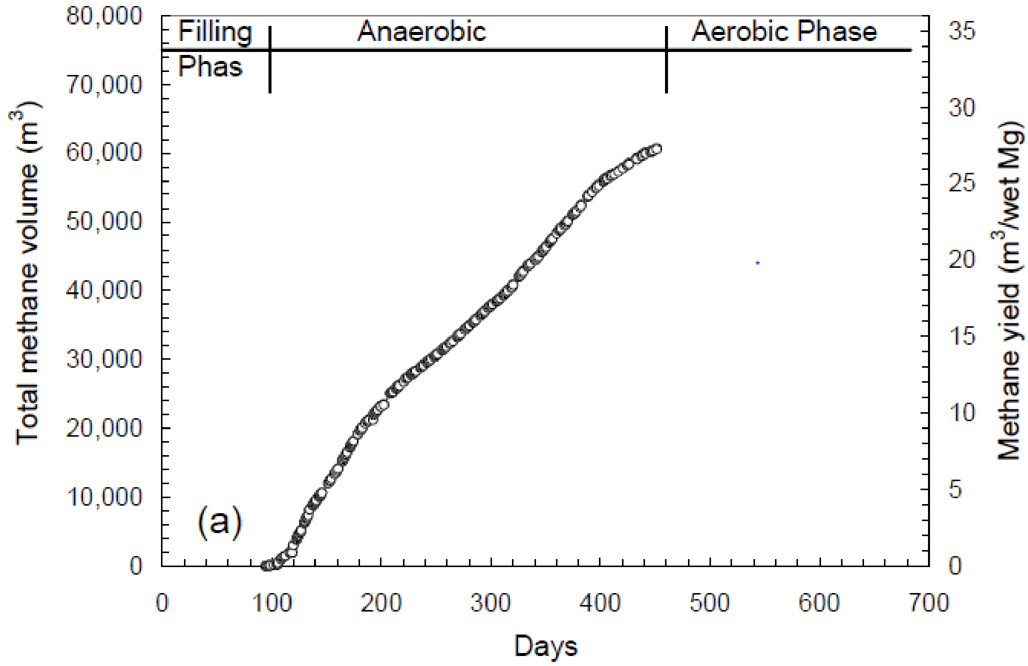


Figure 2-25 Total methane volume and methane yield for digester cell during anaerobic phase (Yazdani, 2010)

Chapter 3

Methodology

3.1 Introduction

The objective of the study was to determine the effects of enzyme and manure on the degradation of organic fraction of Municipal Solid Waste(MSW) in Biocell landfills. Laboratory tests and experiments were conducted for analyzing the effects of additives on degrading organic fraction of MSW thereby enhancing the gas production. Different laboratory tests such as physical characterization, moisture content, volatile organic content test, pH, BOD, and COD tests were conducted along with the measurement of rate, volume, and composition of generated gas.

The overall experiment was divided into following four tasks:

Task 1: Selection of feedstock, inoculum, and its mix ratio for reactors: This included selection of inoculum and determining its percentage in the feedstock. Various combination of manure and waste, enzyme and waste were selected for the reactors' feedstock based on literature review.

Task 2: Collection, storage, and initial characterization of both MSW and inoculum: The physical composition, moisture content and VOC tests were conducted on collected MSW as well as of reactor feedstock. The pH of inoculum was also measured.

Task 3: Construction of laboratory scale simulated landfill Biocell reactors: The reactors were designed and constructed to be air tight. Leak tests were conducted for this purpose.

Task 4: Operating and monitoring laboratory scale simulated landfill reactors: This step involved measuring of generated gas volume and percentages of methane (CH_4), carbon dioxide (CO_2) and oxygen (O_2) in generating gas, and measuring leachate's volume, pH, BOD, and COD.

Task 5: Final laboratory Testing: This included the moisture content and volatile solid test on degraded feedstock waste.

The experimental flowchart showing the experimental procedure that was carried out in the laboratory is shown in Figure 3-1.

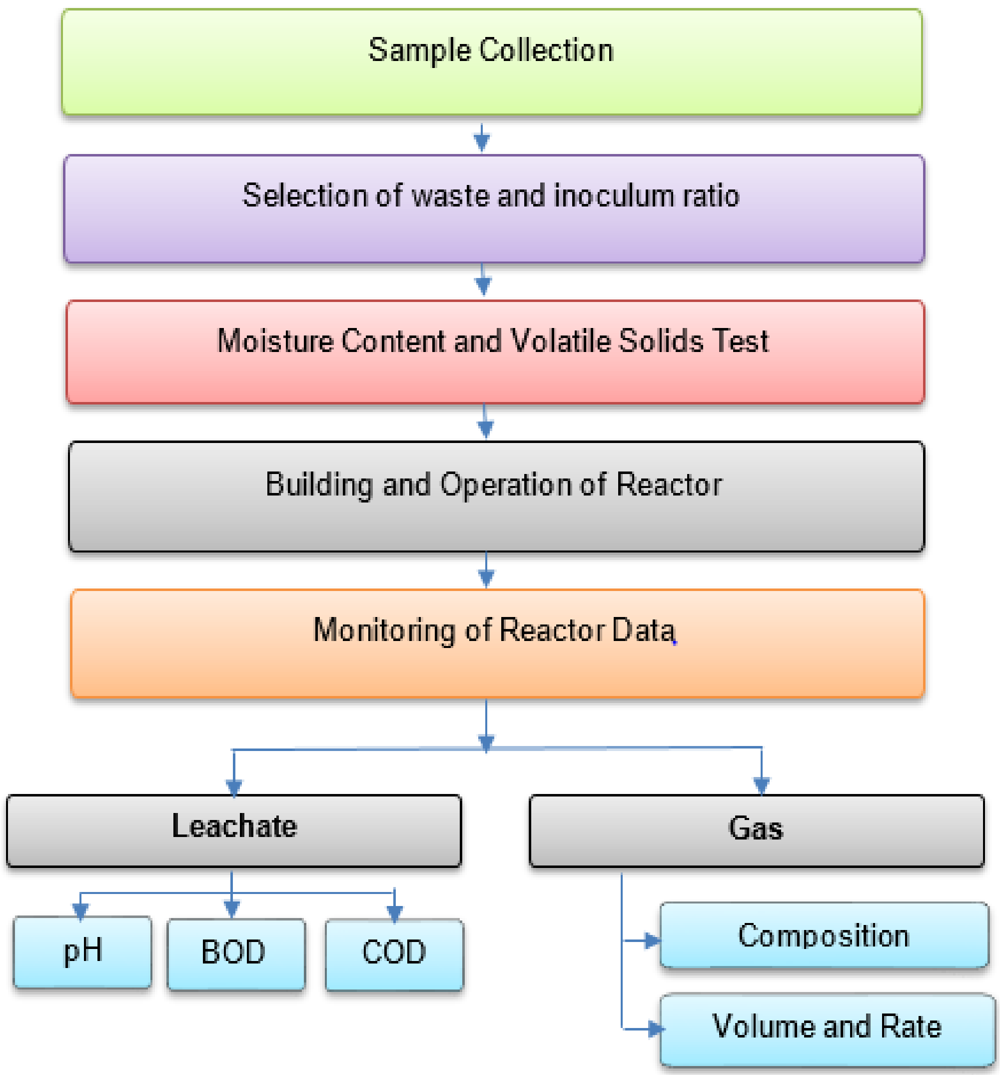


Figure 3-1 Flow chart of Experimental Program

3.2 Inoculum type and its percentage

Three different types of livestock manure were selected for this study based on the digestive physiology of livestock, availability of manure, and manure's inherent gas production efficiency to determine the effect of different types of inoculum on organic waste degradation and consequent gas generation. 10% of sludge was added to all the reactors to introduced microbes to shorten the long lag phase. A mixture of 6% of manure and 10% of sludge was selected for this study based on previous studies found in the literature. The inoculum with 0.00000213% of lignocellulose enzyme, Manganese Peroxidase (MnP), and 10% of sludge was also selected for this study to compare its efficiency in enhancing degradation of organic waste degradation to the reactors using manure. The MnP enzyme was selected because it was proved to degrade waste faster with the good amount of gas generation (Jayasinghe, 2013). The percentage of MnP used in the current study was based on the recommendation of Jayasinghe (2013) for field scale Biocell landfill.

3.3 Sample collection and storage

Sample collection and storage can be divided into four tasks which are described in subsequent sections:

3.3.1 MSW collection and storage.

MSW was collected from the City of Denton Landfill on March 4, 2016. Total 6 bags of MSW samples were collected. The collected bags were tagged A-1 to A-6 in a chronological order of sample collection from the field. Collected samples were brought to the Civil Engineering Laboratory Building in plastic bags and kept inside the environmental growth chamber (cold room) maintained at 4°C (38° F) for the preservation of moisture and other initial properties of collected waste. Figure 3-2 shows the MSW collection from work face of the City of Denton Landfill and storage of sample in environmental growth chambers.

Figure 3-2 (a) Working phase of The City of Denton Landfill; (b) Collection of MSW for working face of The City of Denton Landfill; (c) Environmental control chamber; (d) Sample storage

3.3.2 Collection of manure

The cow, pig, and horse manure needed for the experiment were collected from different sources. Since aged manure is recommended by Yazdani (2010) for green waste decomposition due to its increase nitrogen ratio, reduced acidic bacteria and increased hydraulic retention time, aged manure was preferred for this study. A 20 lb. bag of cow manure was collected from a Calloway's Nursery Dallas which was approximately 9-12 month old. Pig manure was collected from Colvin Clear Creek Farm/ Maypiggen which was approximately 6- 18 months old. Horse manure was collected from Tripe H farm. Collected horse manure was less than 1 week old because it was very difficult to obtained aged horse manure from local nurseries. The manure was stored at room temperature. The collected manure used for this experiment is shown in Figure 3-3.

Figure 3-3 (a) Collected manures; (b) Pig manure; (c) Horse Manure; (d) Cow manure

3.3.3 Collection of sludge

Sludge was collected from the City of Denton Landfill. Two buckets of size 5 gallons were used for collecting the sludge. The collected sludge was stored in air tight bucket to maintain anaerobic condition before it was added to the reactors. Sludge was added to the reactors as a source of microbes and also to act as a buffer which helps to neutralize the acidic environment inside the reactor. Figure 3-4 shows the collected sludge from The City of Denton Landfill.

Figure 3-4 Collected sludge from The City of Denton Landfill.

3.3.4 Manganese Peroxidase and Hydrogen Peroxide

The Manganese Peroxidase (MnP) is described as true lignin degraders because of their high potential redox value (Martinez et al., 2005). The MnP enzyme is soluble in water. MnP used in this experiment was brought from Santa Cruz Biotechnology Company which develops and supplies different products for the biomedical research market. The MnP enzyme was shipped along with the blue ice because it is recommended to store at -20°C. The product number (EC number) for this enzyme is 1.11.1.13. The MnP enzyme thus procured was in the form frozen-dried brown colored amorphous powder. The MnP enzyme, placed between two blue ice packs, was stored in a freezer maintained at -20°C temperature. The MnP enzyme needed to be activated using hydrogen peroxide (H2O2). The 30% H_2O_2 solution was purchased from Hach Company having Product number: 14411. Figure 3-5 shows MnP enzyme and its storage.

(a) (b) (c)

Figure 3-5 a) Manganese Peroxidase (MnP); (b) Received shipment of MnP; (c) Storage of MnP in Freezer

3.4 Physical composition of collected municipal solid waste

The physical composition of MSW samples was determined on the wet weight basis by sorting manually and categorized into food waste, paper, plastics, textile, styrofoam and sponge waste, yard and wood waste, metals, glass, construction debris and others. The MSW samples were also classified based on their degradability. The degradable fraction of MSW is comprised of food waste, paper, yard and wood waste, and textile waste whereas the nondegradable fraction of MSW is comprised of plastic, metals, glass, construction debris, and others.

According to the physical composition of degradable waste, the percentage of paper, food waste, textile & leather, yard and wood waste was fixed. MSW composition thus selected for reactors was 50% paper, 20% food waste, 15% textiles, and 15% yard and wood waste which is shown in Figure 3-6. The details of the determination of this composition are explained in Chapter 4.

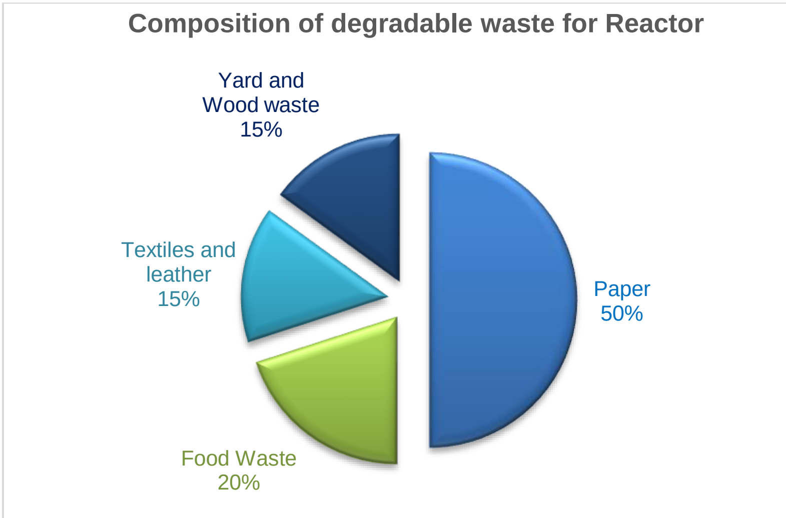


Figure 3-6 Composition of degradable waste used in reactors

3.5 Waste and inoculum combination for reactor feedstock

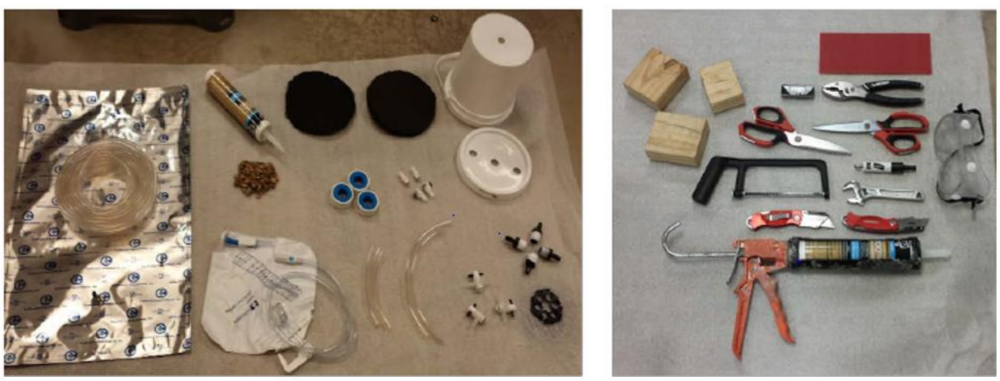
Laboratory simulated Biocell landfill reactors were built in order to analyze the effect of manure and enzyme on the degradation of the organic fraction of MSW and gas generation. Five pairs of reactors were constructed for this study named A1 to A10. The total amount of feedstock in all the reactors was 2 lbs. The composition of waste and inoculum for feedstock for each reactor is shown in Table 3-1.

Table 3-1 Waste and inoculum combination for reactor feedstock

Reactors	**Degradable MSW waste (%)**	**Sludge (%)**	**Manure or Enzyme (%)**
A1 and A2	90	10	control
A3 and A4	84	10	6% (cow manure)
A5 and A6	84	10	6% (pig manure)
A7 and A8	84	10	6%(horse manure)
A9 and A10	90	10	0.00000213%(MnP)

3.6 Construction of laboratory simulated landfill reactor

Five pairs of laboratory simulated landfill reactor were constructed to evaluate the effects of manures and enzyme on the degradation of the organic fraction of MSW. The various combination of waste and inoculum used in the reactors is tabulated in Table 3.1. The reactors were prepared by using smart seal leak tight HDPE buckets of 1 gallon capacity, bought from United States Plastic Corporation. Different size of tubings, connectors, Cali-5-BondTM gas bags of 22 liter capacity, leachate bags of 2 liter capacity, valves, silicon sealants, washer, clamps, geocomposites, and gravels were the material required for building the reactor. Figure 3-7 shows the materials and equipment used to construct the reactors.



(a) (b)

Figure 3-7 (a) Materials and (b) equipment used for reactor building

The bucket was modified for gas collection and leachate collection. For this, four holes were drilled in the bucket; one at the bottom of the bucket and 3 at the bucket's lid. Then, tubes were connected to these holes using the threaded hose. Thread tape was used to make connection air tight. The two way and three-way valves were also connected to tubes for leachate collection and gas collection system respectively. Then, all the joints were sealed using silicon sealant to make connections air tight too. After application, the sealant was left to dry for 24 hours. Figure 3-8 shows fitting, fixing and sealing of reactors.

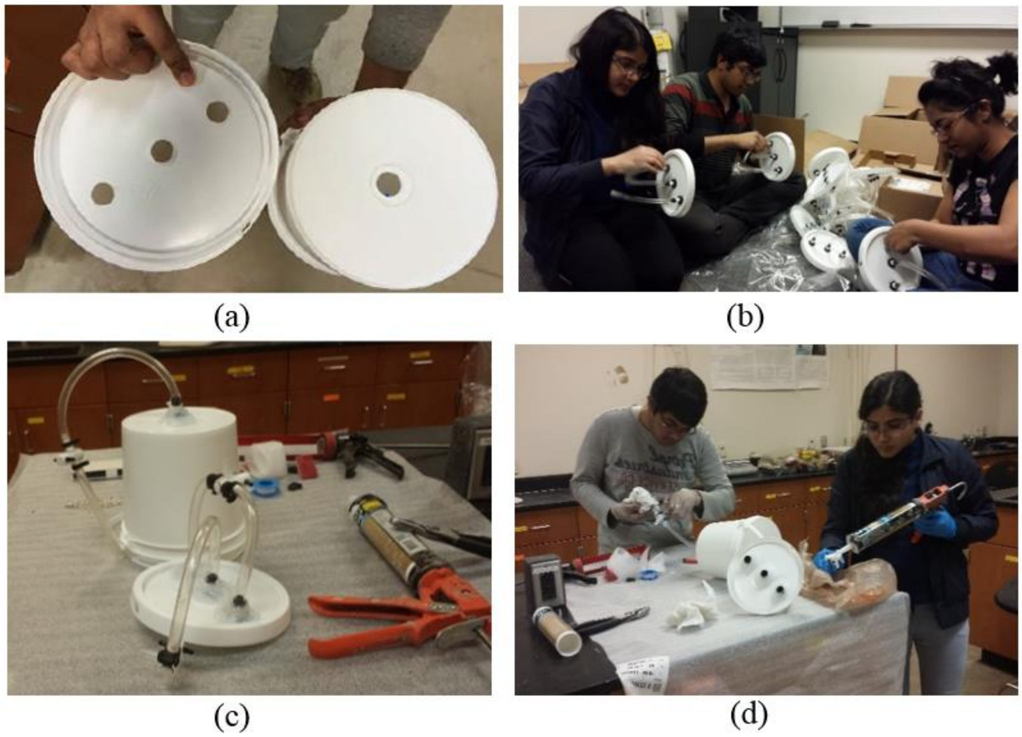


Figure 3-8 (a) Drilling of lid and bucket; (b) Hose and tubing connection; (c) Tubing and Valve connection; (d) Sealing of reactors

The geocomposite layers were provided at bottom and top after filling of the reactors to simulate the landfill liner system. A pea gravel layer was provided up to the level of hose to ensure better drainage of leachate from the geocomposite layer placed at the bottom of the reactor. Figure 3-9 shows the geocomposite and pea gravel installation.

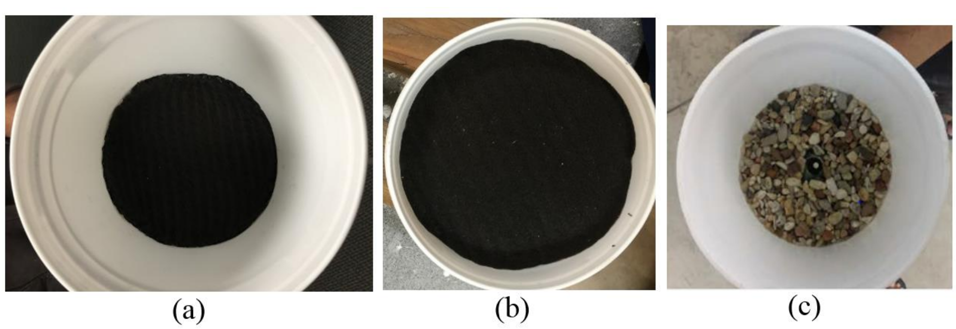


Figure 3-9 (a) Bottom Geo composite; (b) Top Geo-composite; (c) Pea gravel layer

Leak test was conducted to check the reactors for any leaks in valves or sealed connection. Pressure head difference method was used for the test. Leak test was initiated by filling reactors with water from a water tank and maintaining a hydraulic head. The maintained hydraulic level was noted and observed for next 48 hours using transparent tube connected to the reactor. If no drop in the hydraulic head was observed, then the reactor was considered to have passed the leak test. All the reactors passed the leak test following above mentioned procedure. The leak test setup was shown in Figure 3-10.

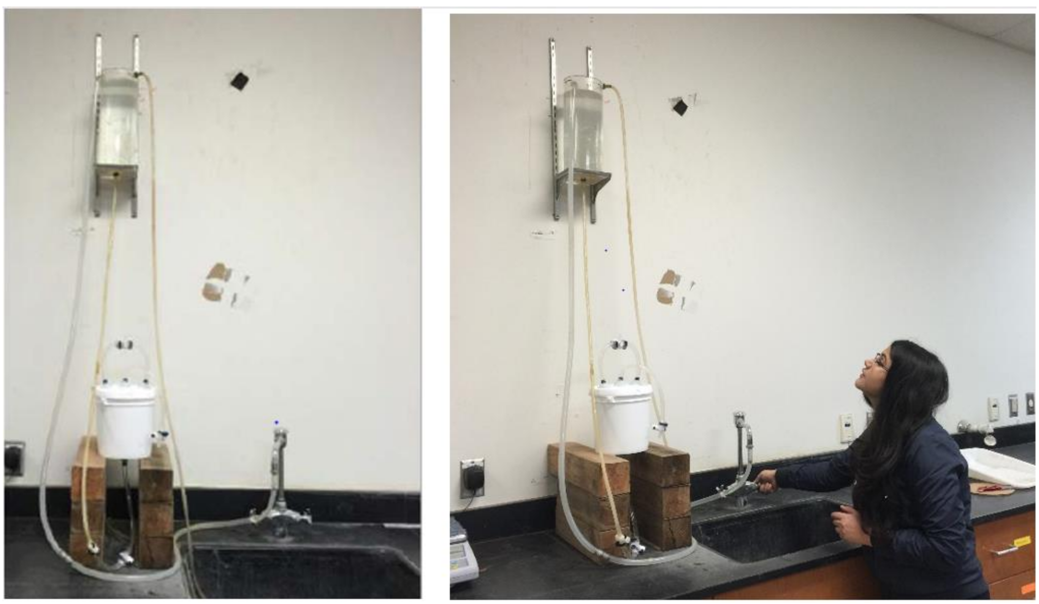
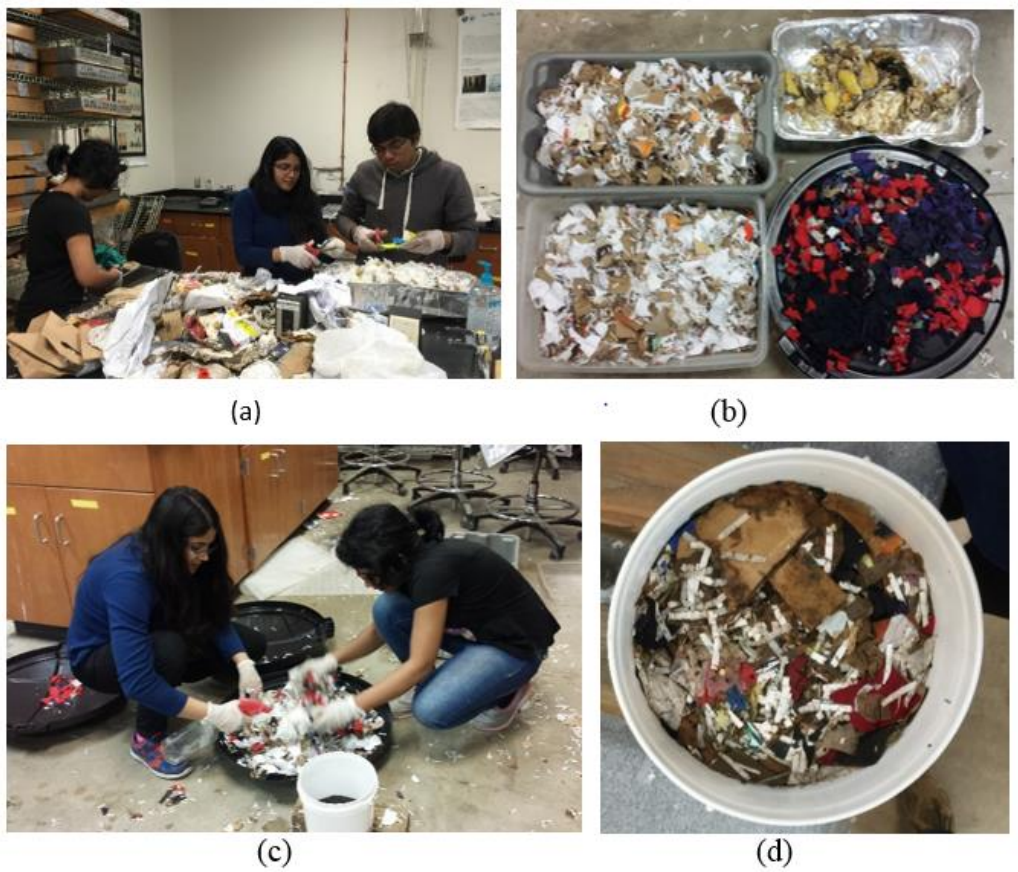


Figure 3-10 Leak test setup

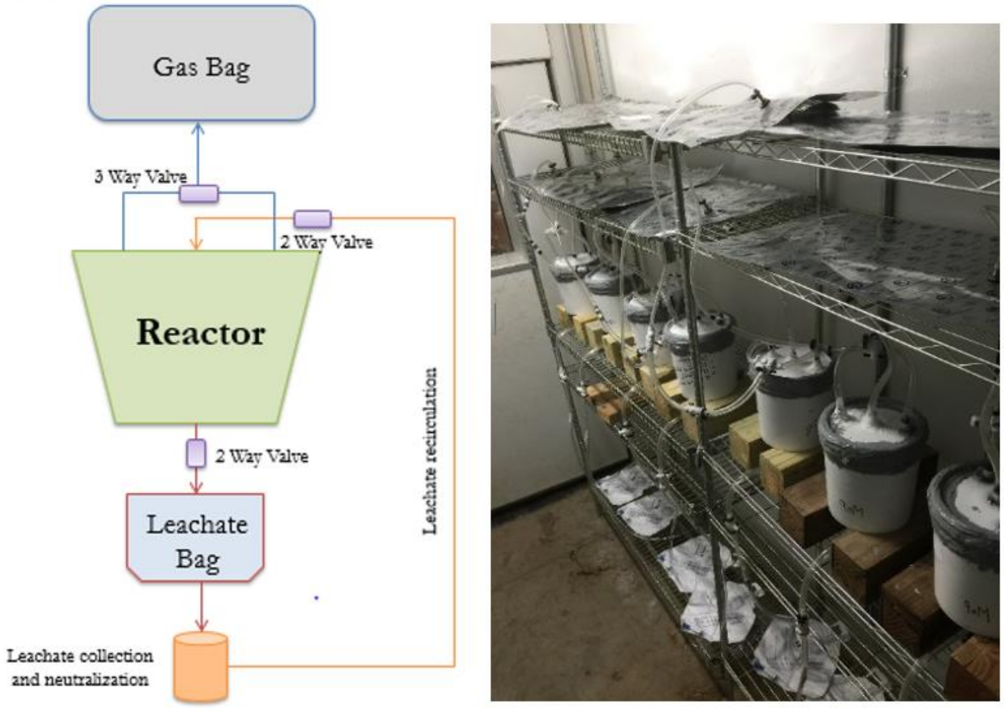
Paper, food waste, textiles and leather, and yard and wood waste were separated from collected MSW. Since shredding of waste improves waste decomposition, MSW samples were shredded to approximately 1.5 inches by 1.5 inches size using scissors before adding to the reactors. It is recommended particle size should be one-fourth to one fifth of the diameter of the bucket for maximum gas production. All the reactors were filled using the organic waste composition provided in Figure 3-6. Degradable waste was mixed homogeneously with inoculum as per combination in Table 3-1. Water was sprayed to each layer of filling in order to ensure proper compaction and to provide sufficient moisture required for microbial activities. Figure 3-11 shows the waste filling procedures.



(a) (b) (c) (d)

Figure 3-11 (a) Shredding of waste; (b) Shredded waste; (c) Mixing of waste and inoculum; (d) Filling of waste

The reactors were filled 1 to 1.5 inch below the top level of the bucket to provide sufficient passage for gas to escape through gas collection outlet. The reactors filled with waste were then sealed around its lid using the double layer of sealant. After sealing, the whole reactor setup was kept in the environmental growth chamber at the temperature of 37˚C. Figure 3-12 shows the schematic diagram for reactor setup and reactors inside the environmental growth chamber.



(a) (b)

Figure 3-12 (a) Schematic diagram for reactor setup; (b) Reactors inside the environmental growth chamber.

3.7 Operation and monitoring of reactors

Leachate and gas generated in the simulated lab scale Biocell reactors were monitored routinely. The leachate monitoring included collection and recirculation of leachate along with measuring generated leachate's volume, pH, BOD, and COD. Gas monitoring included the measurement of composition, rate, and volume of the generated gas. Finally, the difference in moisture and volatile solids before and after degradation was also determined. These activities are discussed in detail in the following sections.

3.7.1 Leachate collection and recirculation

Moisture is one of the important factors affecting degradation of MSW. Research findings suggest that an exponential increase in gas production rate occurs between 35%

and 60% moisture content of waste (Barlaz et al., 1990). Therefore, leachate recirculation is required if moisture content falls below 35%. 400 ml of generated leachate was neutralized and then recirculated daily, for the first few months. The daily recirculation was required to neutralize the acidic environment inside reactors as the pH was dropping significantly below neutral level. The acidic environment inhibits the gas generation causing lag phase. The generated leachate was neutralized using KOH to ensure a basic condition which allows effective microbial action. The volume of leachate was measured in the graduated flask. If the volume of leachate was less than 400 ml then water was added to make the recirculation volume equal to 400 ml. Figure 3-13 shows the leachate collection and recirculation procedure.

(a) (b)

Figure 3-13 (a) Leachate volume measurement; (b) Leachate recirculation through funnel

3.7.2 Gas collection and measurement

The composition and volume of generated gas were measured on a regular basis. The collection of gas was done in Cali-5-BondTM 22 liter gas bags. Landtec GEM 2000 PLUS with infrared analyzer was used to measure the composition of the collected gases.

This Landtec measured the concentration of methane (CH_4), carbon-dioxide (CO_2), oxygen (O_2), and trace gases in the gas bags. The volume of collected gas was measured using standard SKC grab air sampler (SKC Aircheck sampler model 224-44XR) connected to a calibrator (Bios Defender 510). Standard Aircheck sampler was used to pumped out the gas whereas calibrator provided the average rate of gas flow. The time was observed during emptying the gas bags. Then, the volume of gas inside the gas bag was calculated using rate and time noted during emptying the gas. Figure 3-14 shows gas composition and volume measurement.

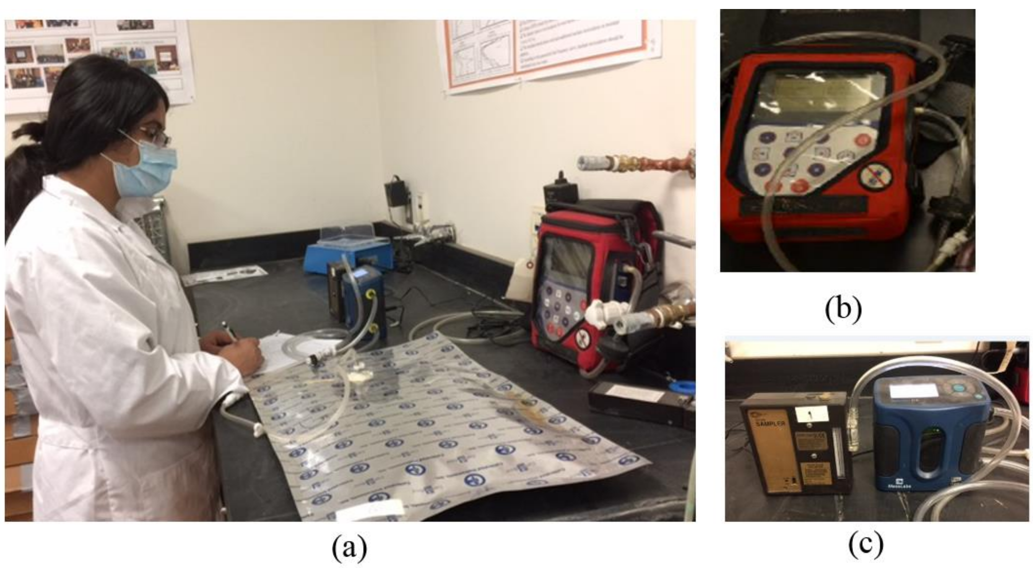


Figure 3-14 (a) Gas composition and volume measurement; (b) Landtec GEM 2000 PLUS; (c) SKC Aircheck sampler model 224-44XR connected to Bios Defender 510

3.7.3 Leachate quality monitoring

Leachate quality monitoring included the measurement of pH, BOD, and COD. The pH, BOD, and COD were monitored periodically.

3.7.3.1 pH

The pH of collected leachate was measured using Bench top Oakton pH meters. The pH meter was calibrated using three point calibration method. The pH buffer of 4.00 ± 0.01, 7.00 ± 0.01 and 10.00 ± 0.01 was used for calibrating the probes. Figure 3-15 shows pH measurement using Oakton pH meters.

(a) (b)

Figure 3-15 (a) pH measurement using Oakton pH meters; (b) KOH solution

3.7.3.2 Chemical Oxygen Demand (COD)

The Chemical Oxygen Demand (COD) of leachate samples was measured using Spectrophotometer (Spectronic 200+). The Spectrophotometer measured the absorbance of light by each sample The absorbance value for each sample was displayed on the screen. Duplicates were prepared for each sample. The dilution factor should be fixed before performing the COD and BOD tests of the leachate. Therefore, to fix the dilution, COD tests were performed with different dilution factor. The dilution factor which falls under the value of calibration curve was selected for this study. The dilution factor of 1: 100 was required as per COD test performed. 1:100 means adding 1 part of leachate to 99 parts of distilled water. The 2.5 ml of diluted leachate samples were added into COD vials. The COD vials with samples were kept in the digester at 150^{o}C temperature for 2 hours. The heated samples were then allowed to cool to room temperature for about 30 minutes and then placed into spectrophotometer to determine the absorbance values. Figure 3-16 shows COD measurement procedure.

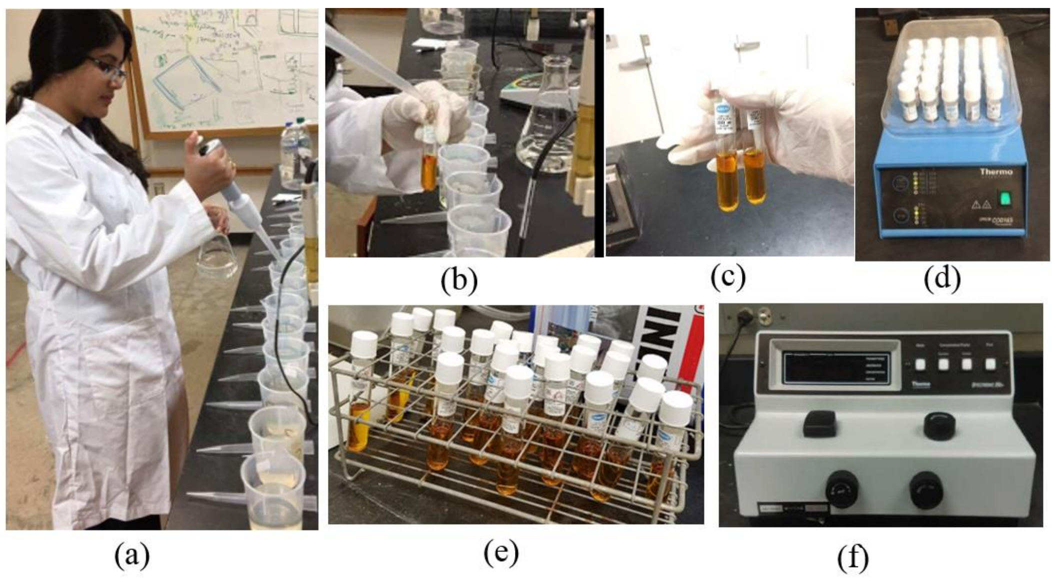


Figure 3-16 COD determination process: (a) Dilution water preparation; (b) Adding 2.5 ml of leachate sample in COD vails; (c) COD vails; (d) Digester; (e) Cooling COD vails in room temperature; (f) Spectrophotometer

A calibration curve was used to obtain the COD values from absorbance percentage. For this purpose, a calibration curve was generated using potassium hydrogen phthalate solution of known COD values. The calibration curve used for determining COD value is shown in Figure 3-17. Using calibration curve, COD values were determined from corresponding absorbance values. Then, the COD values were adjusted as per dilution factor to get actual COD of leachate samples.

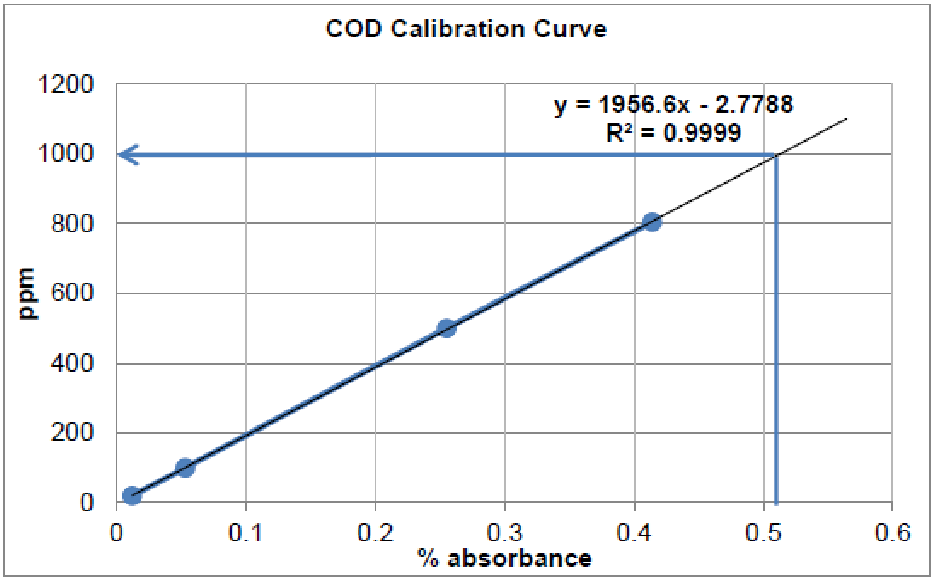


Figure 3-17 COD calibration curve

3.7.3.3 Biochemical Oxygen Demand (BOD)

The Biochemical Oxygen Demand (BOD) was determined by measuring initial and final dissolved oxygen. The dissolved oxygen was measured using HACH HQ 440d benchtop dissolved oxygen measuring instrument. The Standard BOD Procedure 8043 was used to perform BOD test. The dilution factor used in BOD was same as COD test. A dilution factor of 1: 100 was required. Initially, the 75% of 300 ml BOD bottles were filled dilution water. Dilution water was prepared by adding buffer pillow (one pillow for 4 liter deionized water) which was aerated for two hours. Then, 3 ml of seed solution, prepared by adding one seed capsule in 500ml of dilution water, was also added to the BOD bottles. After adding seed solution, leachate samples were added. Each test was performed in triplicates by varying the volume of leachate added. The calibrated BOD probe was used to measure initial dissolved oxygen (DO) for each sample. The samples were then kept at 20^0 C temperature for 5 days and final dissolved oxygen was determined. The five day BOD_5 was calculated for all the samples. Figure 3-18 shows the BOD_5 measuring process.

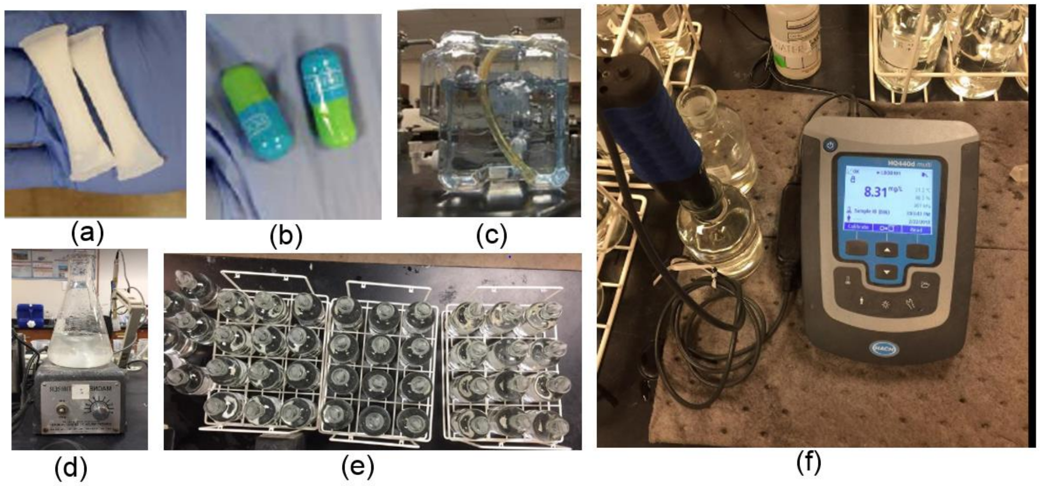


Figure 3-18 BOD measurement process: (a) Buffer pillow; (b) seed capsule; (c) aeriation of dilution water; (d) Preparation of seeding solution using Magnetic stirrer; (e) BOD bottles; (f) Dissolved oxygen (DO) measurement.

3.7.4 Dismantling of reactors

The reactors were dismantled after 241 days of operation. The settlement of waste and weight loss after degradation was measured using the scale and weighing machine respectively. The Figure 3-19 shows the process of dismantling of reactors.

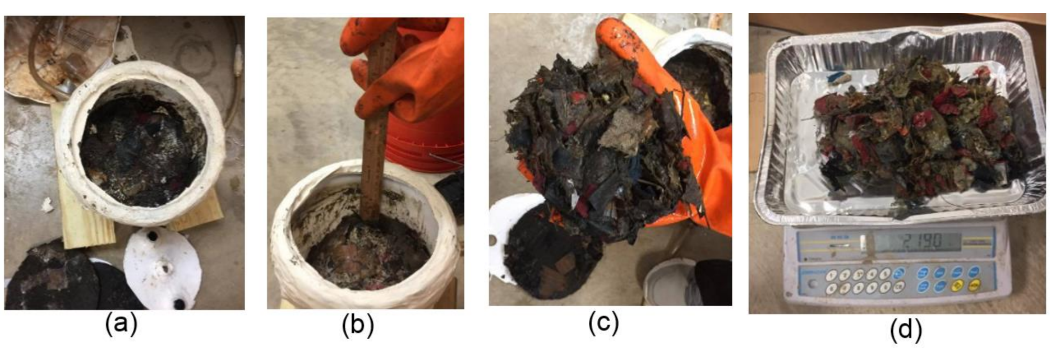


Figure 3-19 (a) Reactor Dismantling; (b) Height settled; (c) Degraded waste; (d) Weight measurement

3.7.5 Determination of change in moisture content

The moisture content test was performed using standard method. The moisture content test was done using approximately 2 lb. of samples. The samples were then dried in the oven at 105°C for 24 hours. The moisture lost after drying was used to the calculated moisture content of samples on both dry weight and wet weight basis. Moisture content on a wet weight basis (w_w) and dry weight basis(w_d) were determined using equation 3.1 and 3.2 respectively.

$$w_w = \frac{Mw}{Mt} \times 100\% \text{..}(3.1)$$

$$w_d = \frac{Mw}{Ms} \times 100\% \text{..}(3.2)$$

Where M_w is the mass of water, M_t is the total wet mass of waste and M_s is the dry mass of waste after drying.

The moisture content of MSW for collected bags (A-1 to A-6) was determined in the initial phase of the experiment. The moisture content of the reactors feedstock before and after degradation was determined hereafter referred as initial and final moisture content of reactors respectively. Figure 3-20 shows the moisture content test.

(a) (b)

Figure 3-20 Moisture content test: (a) Oven set at 105ºC; (b) Dried waste in oven after 24 hours

3.7.6 Determination of change in volatile solids

Volatile solids are an indicator of the organic content in the waste samples. Organic content of the waste is expected to decrease as the waste decomposes. The samples were oven dried at 105ºC and then grinded. 50 gram of ground sample was taken and ignited in a muffle furnace at 550^{o}C (± 10^{o}C) for about 2 hours or until it reached the constant weight. The percent weight lost during ignition is the volatile solids in the waste. Volatile content (VS) was determined using equation 3.3.

$$VS\ (\%) = \frac{W_l}{W_t} \times 100\% \qquad (3.3)$$

Where, w_l is the weight loss after burning and w_t is the dry weight of sample before burning.

The volatile solids before and after degradation were determined for all reactors. Figure 3-21 shows the volatile solids test before and after degradation.

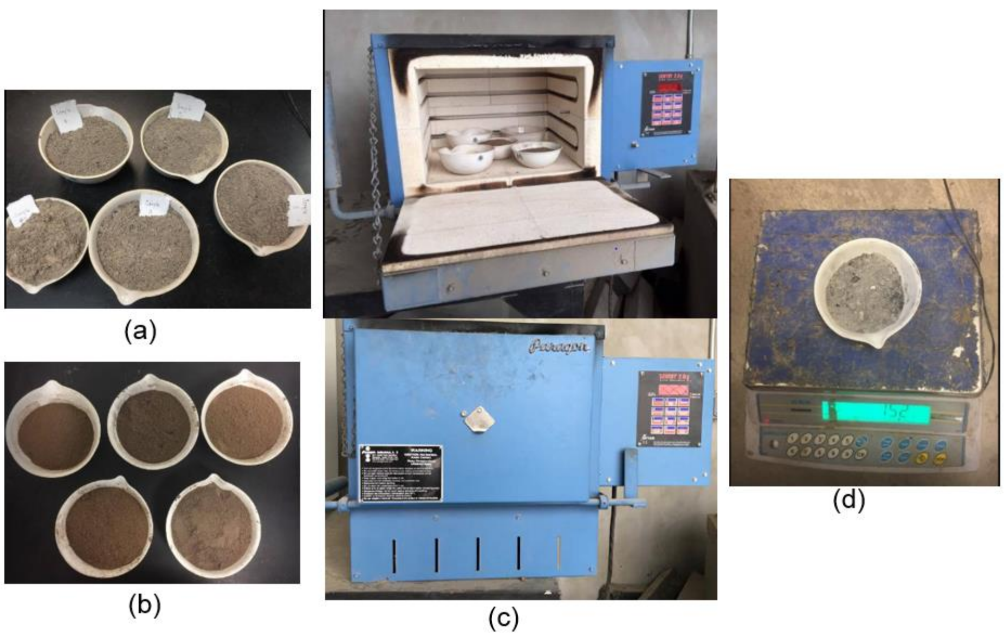


Figure 3-21 Volatile solids test; (a) Grinded fresh feedstock (b) Grinded degraded feedstock;(c) Muffle furnace with samples; (d) Weight after burning

Chapter 4

Results and Discussion

4.1 Introduction

This chapter mainly focuses on the results obtained from the experimental study. Results and its analysis have been presented in four sections. The characterization of collected Municipal Solid Waste(MSW) along with the initial moisture content test and volatile organic content test of the feedstock were discussed in the first section. The second section includes the characterization of collected inoculum. Subsequently, third section includes the measurement of pH, volume and chemical properties such as BOD and COD of generated leachate. This section also includes the measurement related to gas generation such as gas volume, rate of gas generation and composition of generated gas. Finally, the fourth section includes the final moisture content, final volatile organic content, weight loss of feedstock, and its comparison with initial value

4.2 Physical composition of MSW

The Physical composition of MSW samples was determined using wet weight basis. For this, MSW samples were sorted manually and then categorized into different components as discussed in section 2-2 of chapter 3. The physical composition of collected MSW based on the waste type and degradability are described below:

4.2.1 Physical ccomposition of collected MSW

Six bags of sample were collected from the working face of the City of Denton Landfill. All the bags of sample were sorted out manually and physical composition was determined on the wet weight basis. The MSW sample was composed of 34% paper, 19% plastic, 13% food waste, 8% textile, 2% styrofoam and sponge, 9% yard and wood waste, 3% metals, 2% glass, 4% construction debris and 6% others (soils and fines). Results obtained from physical composition test of each bag are presented in the following Table 4-1 and Figure 4-1.

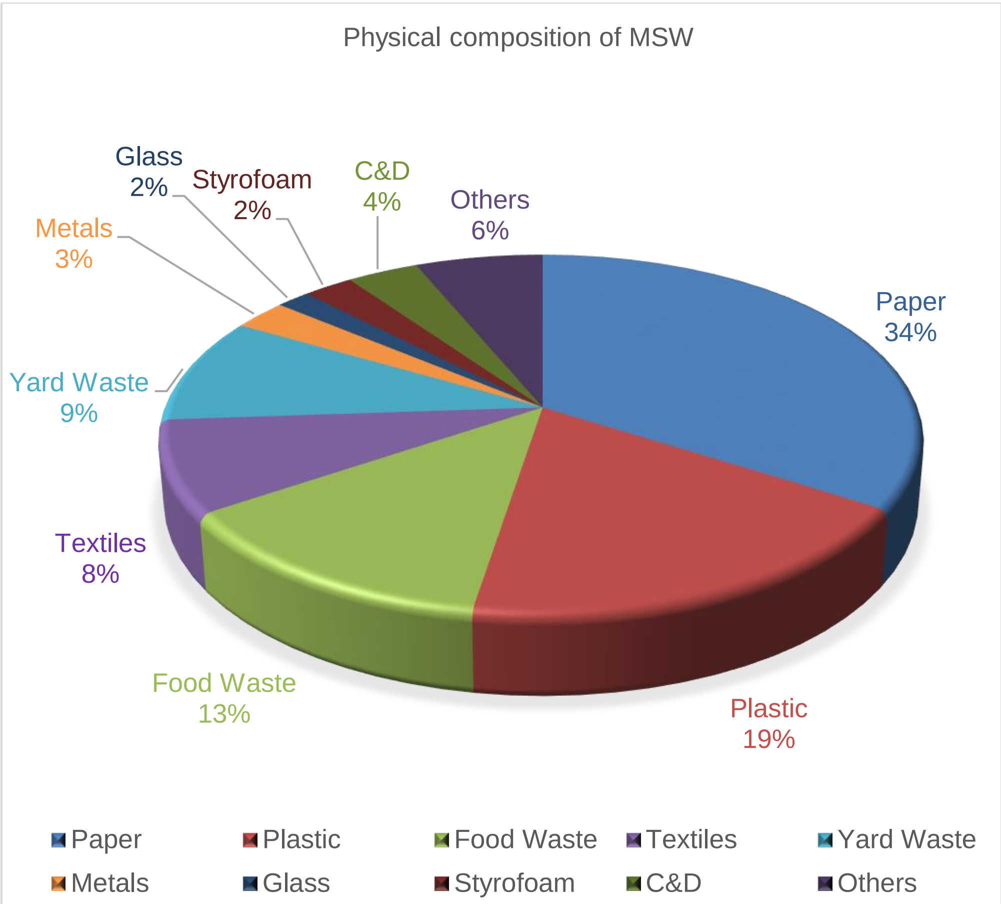


Figure 4-1 Average physical composition of MSW

Table 4-1 Physical composition of MSW

S. No.	Physical Composition (by %)										
	Paper	**Plastic**	**Food waste**	**Textile and Leather**	**Yard Waste and Wood**	**Metals**	**Glass**	**Styrofoam and Sponge**	**C & D Debris**	**Others (Soils and Fines)**	**Total (Weight of waste in lb.)**
1	56.73	8.52	3.49	7.21	16.93	0.33	0.09	2.35	3.40	0.94	22.95
2	20.94	25.18	20.39	1.64	14.85	6.10	3.51	2.57	0.00	4.83	29.94
3	43.98	15.46	5.76	2.49	17.02	1.45	0.15	1.11	8.07	4.49	29.26
4	19.99	13.21	10.95	27.89	6.74	1.95	2.82	5.58	8.19	2.67	32.79
5	25.88	18.68	17.40	10.67	1.25	3.63	1.98	2.00	1.34	17.17	28.77
6	36.29	29.74	19.91	0.06	0.00	3.92	1.46	1.14	0.06	7.40	25.39
Average	**33.97**	**18.47**	**12.98**	**8.33**	**9.46**	**2.69**	**1.67**	**2.46**	**3.51**	**6.25**	

The MSW samples were also classified based on their degradability. The classification showed that 64.74% of MSW was the degradable waste while 35.26% of MSW was the non-degradable waste. The degradable fraction was comprised of food waste, paper, yard and wood waste, and textile whereas non-degradable waste fraction was comprised of plastic, metals, glass, construction debris, and others. The details of this classification are presented in Table 4-2. A bar chart comparing the degradable and non-degradable fraction of sorted sample bags is presented in Figure 4-2.

Table 4-2 Physical composition based on degradability of MSW

Sample No.	**Physical Composition by Degradability (% by Weight)**	
	Degradable	**Non-Degradable**
1	84.36	15.64
2	57.82	42.18
3	69.26	30.74
4	65.57	34.43
5	55.20	44.80
6	56.26	43.74
Average	**64.74**	**35.26**

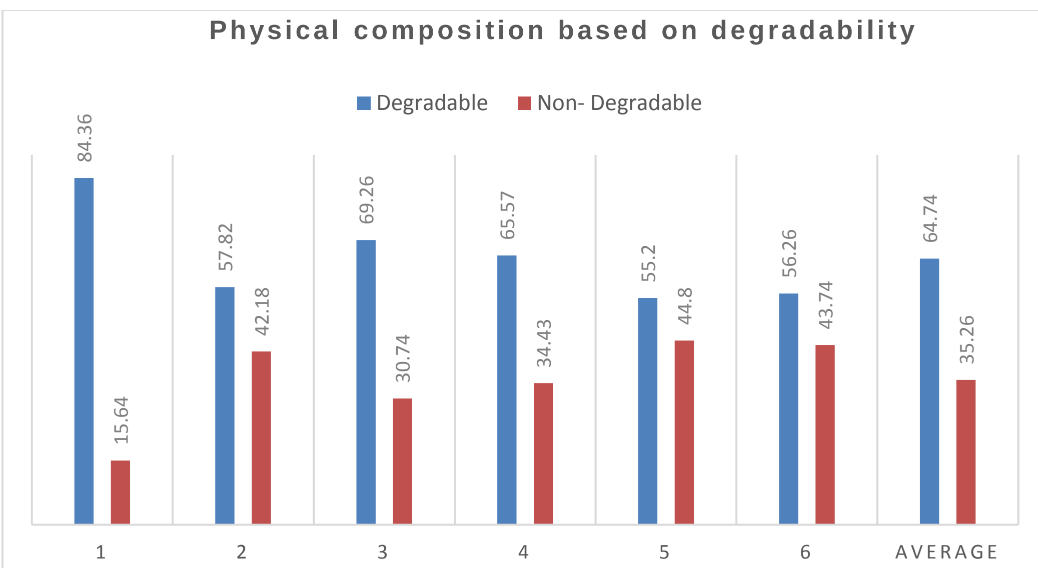


Figure 4-2 Physical composition of MSW based on degradability

Next, physical composition of the degradable fraction of MSW samples was determined which showed that on the average degradable fraction of MSW samples consisted of 51.47% paper, 21.88 % food waste, 12.83% textiles and leather, and 13.8% yard and wood waste. The degradable MSW waste classification is presented in Table 4-3.

Table 4-3 Physical Composition of Degradable component of MSW

Sample No.	Physical Composition (by %)			
	Paper	**Food waste**	**Textile and Leather**	**Yard Waste and Wood**
1	67.25	4.13	8.55	20.07
2	36.22	35.27	2.83	25.68
3	63.49	8.31	3.60	24.57
4	30.49	16.70	42.53	10.28
5	46.88	31.52	19.33	2.27
6	64.49	35.37	0.10	0.00
Average	**51.47**	**21.88**	**12.83**	**13.81**

4.2.2 Comparison with previous literature

Comparison of MSW physical composition determined in the current study was compared with US EPA (2014). The percentage of paper in waste collected from Denton for the current study was more than US EPA National average waste composition and less than Taufiq, T (2010) whereas the percentage of food waste collected was similar to that of national waste composition and more than Taufiq, T (2010). The composition of MSW is highly dependent on regional, cultural, social, and economical aspects. (Denafas et. al., 2014) Therefore, it is possible to get different composition for different countries as well as cities. The average composition of degradable waste was similar to the national average degradable waste composition of 2014 reported in US EPA (2016). The waste composition of the current study and US EPA (2016) is presented in Table 4-4.

Table 4-4 Comparison of physical composition of MSW

Components	MSW composition of 2014 (US EPA, 2016), %	Fresh waste composition (Taufiq, T 2010)	Fresh MSW composition of the current study, %
Paper	26.6	40	33.97
Plastic	12.9	18	18.47
Food waste	14.9	2	12.98
Textile	9.5	4	8.33
Yard trimming	13.3	9	9.46
Wood waste	6.2		
Metal	9	5	2.69
Glass	4.4	1	1.67
Styrofoam	-	1	2.46
C&D debris	-	2	3.51
Others	3.2	18	6.25
Total	100	100	100
%Degradable	70.5	55	64.74
%Non-degradable	29.5	45	35.26

Note: Taufiq's (2010) study was based on MSW from Denton, Texas.

4.2.3 Physical composition of feedstock:

Based on the physical composition of degradable waste collected from the City of Denton Landfill, the percentage of paper, food waste, textile & leather, yard and wood waste was considered a fixed percentage for this study. The composition percentage thus fixed were rounded for ease of sample preparation. MSW composition finally used for this study for preparation of feedstock was 50% paper waste, 20% food waste, 15% textile waste and 15% yard and wood waste which is shown in Table 4-5.

Table 4-5 Comparison of physical composition of MSW

Degradable waste components	Physical Composition (%) (Based on collected waste form City of Denton landfill)	Composition Selected for feedstock (%)
Paper waste	51.47	50
Food waste	21.88	20
Yard and wood waste	12.83	15
Textiles and leathers	13.81	15

Two pounds of organic waste along with the different combination of sludge and manures were fed into the reactors. The sludge and manure were added to provide sufficient nutrients and microorganism to enhance the degradation process. Sludge also act as a buffer which helps to neutralize the acidic environment. A total of 5 pairs of reactors were built and operated for this study. Among these 5 pairs of reactors, one pair acted as control reactor which had MSW with 10% of sludge. Other three pairs were prepared using feedstock composed of MSW with 10% of sludge and 6% of manures. The last pair of reactor was prepared using feedstock composed of MSW with 10% of sludge and 0.00000214% of the enzyme (Manganese Peroxidase) along with the organic fraction of municipal solid. Different combination of organic waste, sludge and manure; and organic waste, sludge, and enzyme used for this study is described and shown in chapter 3.

4.3 Moisture content test

The moisture content tests were also conducted on both collected MSW as well as for the feedstock for Biocell reactors which is described in the subsequent sections:

4.3.1 Moisture content of MSW collected

The moisture content of MSW from sample bags 1 to 6 was determined at the time of physical composition. The collected samples were kept in the cold room at a temperature of 4 degrees centigrade in order to preserve the actual field moisture content. The moisture content was determined in both wet weight and dry weight basis. The average moisture content of fresh MSW was found to be 37.93% on the dry weight basis and 27.22% on wet weight basis. The determined moisture content of MSW samples are presented in Table 4-6.

Table 4-6 Moisture content of MSW

Sample No.	**Moisture Content (%)**	
	Dry weight basis	**Wet weight basis**
1	44.84	30.96
2	46.31	31.65
3	39.67	28.40
4	21.20	17.49
5	37.58	27.31
6	37.99	27.53
Average	**37.93**	**27.22**

4.3.2 Initial moisture content of reactors feedstock

The moisture content of the feedstock depends on the moisture content of MSW waste, the moisture content of inoculum, and water added during the compaction of feedstock while preparing reactors. Feedstocks were prepared with sorted and shredded organic waste and 6% of different types of manures (cow, horse, and pig) for different reactors which is described and shown in Chapter 3. The average moisture content of sorted and shredded organic fraction of MSW used as reactor feedstock was found to be 29.6% and 22.84% based on the dry weight basis and wet weight basis respectively. The moisture content of shredded organic fraction of MSW was found slightly less than the moisture content of collected MSW. The reason for this was the loss of moisture during shredding of waste. The moisture content of cow manure was 2%, pig manure was 5% and horse manure was 36% by dry weight basis. The moisture contents of cow and pig manure were less compared to the moisture content of horse manure because the collected cow and pig manure were 9-12 months old while horse manure was less than a week old and fresh manure has more moisture as compare to the aged one. Extra water was also added to each of the reactors' feedstock during filling of waste. The main purpose of the water addition was to ensure proper compaction and saturate the waste for proper microbial activity. This water addition also influenced initial moisture content of the feedstock. Spraying of water was done in 3-5 layers while filling the reactors as described in Chapter 3.

Before installing each reactor, a sample was taken to determine the initial moisture content of the feedstock. Five samples were taken from each pair of reactors' feedstock before adding water for compaction and amount of water added to each reactor for compaction was recorded. The accurate moisture content of the feedstock mixed with shredded organic waste, manure, and additional water was then calculated considering moisture content of the samples and water added for the compaction. Thus calculated moisture content will be referred as the initial moisture content of the waste hereafter. The initial moisture content data for each reactor is tabulated in Table 4-7.

Table 4-7 Initial moisture content of reactors' feedstock

Sample No.	Weight of moist waste (lb.)	Weight of dry waste (lb.)	Feedstock moisture content without additional water		Initial Moisture content of reactor
			Wet weight basis (%)	Dry weight basis (%)	Wet weight basis (%)
A1 and A2 (Control)	2.20	1.61	26.61	36.26	62.97
A3 and A4 (Cow Manure)	2.32	1.70	26.58	36.20	61.06
A5 and A6 (Pig Manure)	2.14	1.59	25.70	34.59	63.08
A7 and A6 (Horse Manure)	2.05	1.49	27.32	37.58	66.34
A9 and A10 (MnP)	2.18	1.58	27.43	37.80	64.13

4.3.3 Comparison with previous studies

The obtained moisture content of collected MSW samples and the reactors' feedstock in this study were compared to the values reported in literature. The moisture contents of organic fraction of MSW and MSW collected without sorting were found similar to that of previous studies shown in Table 4-8.

Table 4-8 Comparison of moisture content of organic MSW found with previous studies

Source / Author	Moisture content (%) (wet weight Basis)	Remarks
Mannassero et al. (1997)	22.5	Fresh un-compacted MSW
Pichtel (2005)	15-40 (avg. 27.5)	Fresh un-compacted MSW
Han et. Al (2006)	26.5	Household MSW
Current Study	27.22(average)	Unsorted MSW
Current Study	25.70-27.43	Reactors' feedstock without additional water
Current Study	61.06-66.34	Initial Moisture content of reactors

4.4 Volatile organic content

Volatile organic test is the best indicator of organic content present in the waste samples. The result of volatile organic content test was conducted on the each pair of reactor feedstocks and the results were compared with previous literatures. These are described in sections below.

4.4.1 Initial volatile organic content

The volatile organic content tests were conducted on the feedstock sample before sealing of the reactors. Five samples were taken from each pair of reactors for this purpose. The volatile organic content found for these samples were referred as the initial volatile solids content of the waste. The initial volatile solids content was found to be similar for all reactors because the composition of organic waste in the reactors has identical composition. The initial volatile solids content data for each reactor is tabulated in Table 4-9.

Table 4-9 Initial volatile solids results for different set of reactors

Sample No.	Weight of grinded samples (gm)	Weight of burnt grinded samples (gm)	Volatile organic content (%)
A1 and A2 (Control)	51.98	6.91	86.71
A3 and A4 (Cow Manure)	54.00	8.42	84.41
A5 and A6 (Pig Manure)	52.00	6.74	87.04
A7 and A6 (Horse Manure)	52.00	7.07	86.40
A9 and A10 (MnP)	54.00	8.12	84.96

4.4.2 Comparison with previous studies:

Comparison of results of the initial volatile organic content test in this study is compared with various literature which are shown in Table 4-10. The results obtained in this study was found to be comparable to the results from previous studies as represented in the bar chart in Figure 4-3.

Table 4-10 Comparison of volatile solids found in the current study with previous studies

Source	Volatile solids (%)	Author	Volatile solids (%) found in current study
Fresh Waste (MSW & sludge mix) including inorganics	70.4 ~ 74.8	Hossain et al. (2014)	84.41-87.04 (Degradable MSW only)
MSW	84	Sivanesan (2012)	
MSW	84	Al-Kaabi et al. (2009)	
MSW	91.5	Haque (2007)	
Food Waste	90.16	Karanjekar (2013)	
Food Waste	91.66 ~ 92.96	Zaman (2016)	

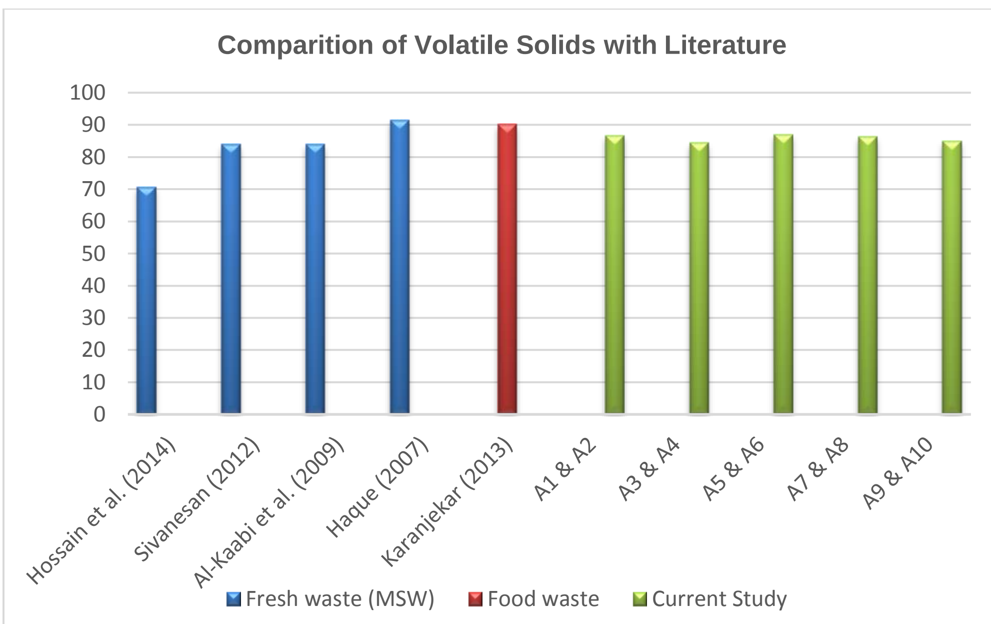


Figure 4-3 Comparison of volatile solid found for organic MSW with previous studies

4.5 Characteristics of collected Inoculum

The characteristic of inoculum is the important property to evaluate its effect on degrading waste. The duration of degradation process was shortened by the addition of inoculum. Inoculum is the source of microorganism which helps to degrade the waste. Degradation of organic fraction of MSW with the use of inoculum was found to be effective in the previous studies (Karanjekar, 2013; Al-Kaabi et al., 2009). Degradation of waste and gas production is directly proportion to the amount of inoculum added. However, for field applicability using the high percentage of inoculum is not feasible. Therefore, low percentages of manures (6%), Manganese Peroxidase (0.00000213 %), and sludge (10%) were used for this study.

The sludge used in this study was collected from the City of Denton landfill, Texas. Anaerobically digested sludge is rich in anaerobic bacteria, acts as the buffer and enhances microbial population to improve the degradation process. The pH of anaerobic sludge is usually above 8 which also helps to neutralize the acidic environment thereby reducing acidogenic phase and transition phase. The pH of the sludge used in this study was found to be 8.37. pH of manures was measured before using them in preparing feedstock. pH for all the manures was found to be above neutral pH (7.0) and shown in Table 4-11. The pH of cow manure was found to be highest about 8.95 and the pH of horse manure was found to be less than other manure (7.69).

Table 4-11 pH test results for sludge and manures

Inoculum Source	Sample 1	Sample 2	Sample 3	Average pH
Sludge	8.42	8.33	8.36	8.37
Cow manure	8.89	9.09	8.86	8.95
Pig manure	7.73	7.43	8.26	7.81
Horse manure	7.68	7.59	7.79	7.69

4.6 Characteristics of generated leachate

The characteristic of generated leachate was determined to understand the level of degradation of the solid waste in the reactors. The pH, volume of generated leachate, BOD (Biological oxygen demand) and COD (Chemical Oxygen demands) was monitored routinely for 241 days and the results thus obtained are discussed in the following sections:

4.6.1 pH of leachate and comparison with previous studies

The pH of the leachate generated from the reactor was measured routinely. The pH of generated leachate provides the best indication of phases of waste degradation. The pH for all the reactors kept on decreasing for first few weeks due to the accumulation of acids. Therefore, Potassium Hydroxide (KOH) was used to neutralize the pH of the generated leachate. When the pH of leachate went below 7, KOH solution was added to neutralize the pH of leachate. Thus neutralized leachate was then recirculated which helped to neutralize the acidic environment inside the reactors as well. The monitoring interval of pH was done frequently for first few months which was gradually increased to 1 week when the reactor entered methanogenic phase. The pH was measured and neutralized in the three day interval for first 20 days and then the leachate recirculation was done daily due to excessive accumulation of acids (Volatile Fatty Acids) which caused pH to decrease below 5 for some reactors. The frequency of pH monitoring was gradually increased to 2, 3, 4, 5 and 7 after 54th, 68th, 113th, 145th and 205th day of operation as the pH started to increase. The increment in the value of pH indicated that the methanogenic bacteria started to convert the organic fatty acids into methane gas and carbon dioxide gas thereby indicating methanogenic phase. The pH of all reactors was monitored till 241 days. The pH variation of generated leachate with time for all the reactors are graphically presented in Figure 4-4. The phases of waste degradation in reactors in relation to the pH level variation is summarized in the following Table 4-12.

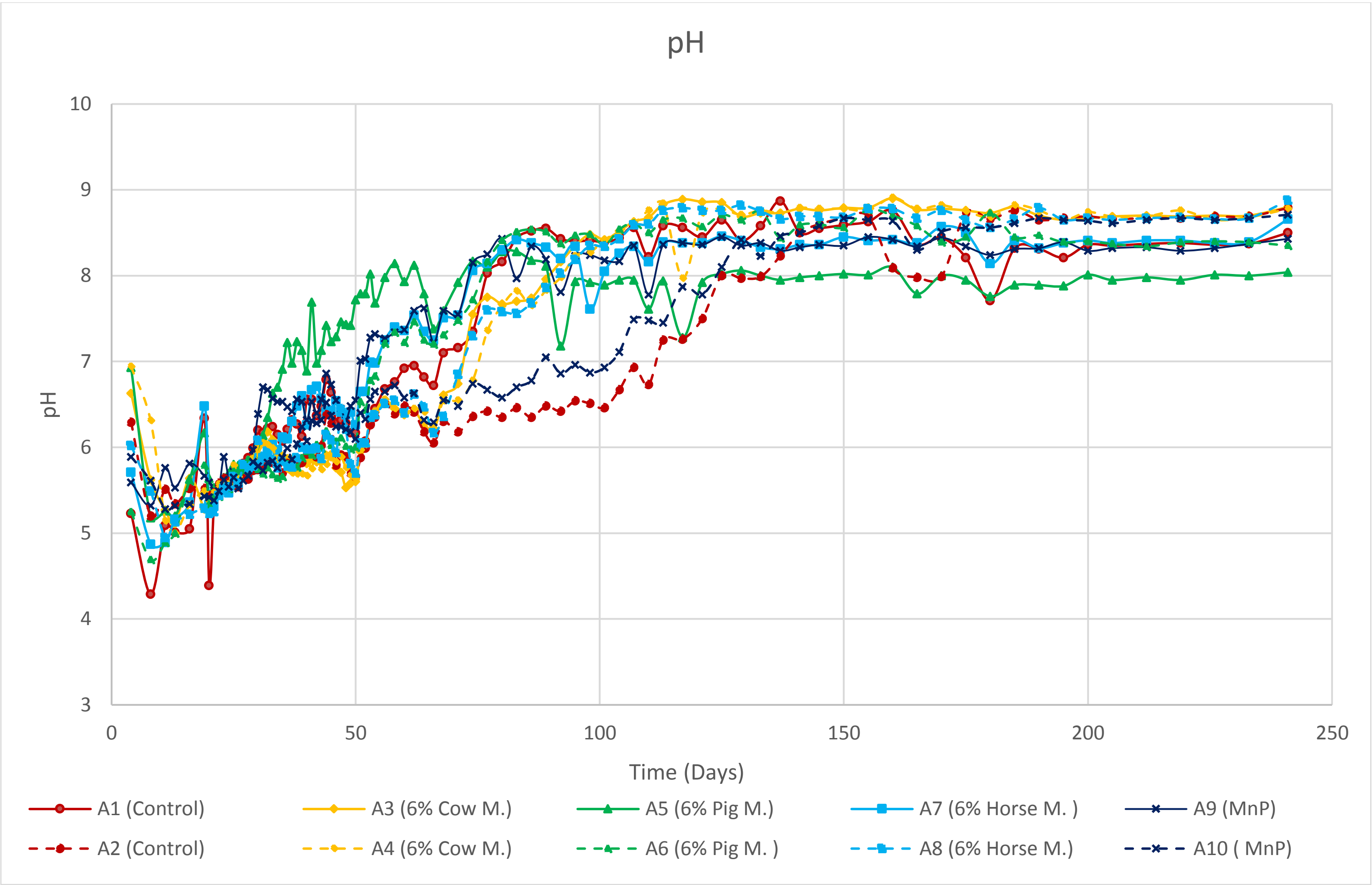


Figure 4-4 Change in pH over time of generated leachate

Table 4-12 Phases of degradation with change in pH levels

Degradation Phase	Phase I		Phase II		Phase III		Phase IV	
Reactor	**pH**	**Time (Days)**	**pH**	**Time (Days)**	**pH**	**Time (Days)**	**pH**	**Time (Days)**
A1 (Control)	5.23 – 7	0 – 4	4.29 – 6.68	4 – 56	6.68 – 7.10	56 – 68	7.10 – 8.87	68 – 241
A2 (Control)	6.29 – 7	0 – 4	5.20 – 6.67	4 – 104	6.67 – 7.25	104 – 113	7.25 – 8.79	113 – 241
A3 (Cow M.)	6.63 – 7	0 – 4	5.11 – 6.56	4 – 56	6.56 – 7.55	56 – 74	7.55 – 8.90	74 – 241
A4 (Cow M.)	6.94 – 7	0 – 4	5.00 – 6.52	4 – 56	6.52 – 7.33	56 – 77	7.33 – 8.91	77 – 241
A5 (Pig M.)	6.93 – 7	0 – 4	5.18 – 6.63	4 – 33	6.63 – 7.22	33 – 36	7.22 – 8.28	36 – 241
A6 (Pig M.)	5.25 – 7	0 – 4	4.69 – 6.53	4 – 51	6.53 – 7.21	51 – 56	7.21 – 8.77	56 – 241
A7 (Horse M.)	5.71 – 7	0 – 4	4.87 – 6.67	4 – 41	6.67 – 7.25	41 – 56	7.25 – 8.66	56 – 241
A8 (Horse M.)	6.02 – 7	0 – 4	4.97 – 6.51	4 – 56	6.51 – 7.30	56 – 74	7.30 – 8.88	74 – 241
A9 (MnP)	5.59 – 7	0 – 4	5.32 – 6.70	4 – 31	6.70 – 7.01	31 – 51	7.01 – 8.45	68 – 241
A10 (MnP)	5.89 – 7	0 – 4	5.28 – 6.56	4 – 53	6.56 – 7.05	53 – 85	7.05 – 8.71	113 – 241

The value of pH indicates the different phases of waste degradation. The initial drop in pH indicates the initial phase of degradation i.e. aerobic phase. Since the size of the reactor was 1 gallon, the aerobic phase of degradation existed for only a few days. Then, the second phase of degradation i.e. acidogenic phase became prominent which was indicated by the decrease in pH value. Acidogenic phase was followed by a transition phase in which pH value started to increase gradually. The transition phase is the phase between acidogenic phase and methanogenic phase. During the transition phase, methanogenic bacteria became active thereby leading to the methanogenic phase. In methanogenic phase, the value of pH increased to 7 and got stabilized with minor fluctuations between 7 and 8. This occurred due to the conversion of the carboxylic acid into methane and carbon dioxide. The variation of pH vs. time for the reactors A1 to A10 are shown in Figure 4-5 and Figure 4-6.

For the reactors A1 and A2 i.e. control reactors with 10% of sludge added as inoculum, initial pH was found to be 5.23 and 6.29 respectively which was measured on the fourth day of operation. The initial pH for reactor A1 was less than reactor A2 which means reactor A1 reached acidogenic phase before reactor A2. The pH of reactor A1 was already low when measured on the 4th day. The lowest value of pH for reactor A1 and A2 was 4.29 and 5.2 respectively observed on the 8th day of operation. The pH of reactor A1 started to increase gradually and reached to 6.68 on 56th day indicating the end of acidogenic phase and beginning of transition phase. Similarly, the pH of reactor A2 started to increase gradually and reached to 6.67 on 104th day. Then, the pH of the reactors A1 and A2 increased to 7.1 on 68th day and 7.25 on 113th day respectively representing methanogenic phase. The reactors pH remained between 7 to 9 for the entire operating period and got settled in the basic state for the rest of their active period. The maximum value of pH for reactors A1 and A2 reached up to 8.87 and 8.79 respectively.

For the reactors A3 and A4 i.e. reactors with 10% of sludge and 6% cow manure added as inoculum, the initial pH was found to be 6.63 and 6.94 respectively which was measured on fourth day of operation. The initial pH for reactor A3 and A4 was comparatively higher than control reactor which might be due to the presence of additional 6% cow manure. The pH cow manure used in reactors was 8.95 whose pH was more than both pig and horse manure. The lowest value of pH for reactor A3 and A4 was 5.11 and 5 respectively observed in 13th day of operation. The pH of reactor A3 started to increase gradually and reached to 6.56 on 56th day indicating the end of acidogenic and beginning of transition phase. Similarly, the pH of reactor A4 started to increase gradually and reached to 6.52 on 56th day. Then the pH of both reactors A3 and A4 increased to 7.55 on 74th day and 7.36 on 77th day respectively representing methanogenic phase. The reactors' pH remained between 7 to 9 for entire operating periods and got settled in the basic state for the rest of their active period. The maximum value of pH for reactors A3 and A4 reached up to 8.9 and 8.91 respectively.

For the reactors A5 and A6, with 10% of sludge and 6% pig manure added as inoculum, the initial pH was found to be 6.93 and 5.25 respectively which was measured on fourth day of operation. The initial pH for reactor A5 was comparatively higher than reactor A6 which means reactor A6 already entered acidogenic phase. The pH of reactor A5 was almost neutral on the fourth day which means reactor A5 was still in the initial phase of degradation. The lowest value of pH for reactor A5 and A6 was 5.18 and 4.69 respectively observed in 8th day of operation. The pH of reactor A5 started to increase gradually and reached to 6.63 on 33rd day indicating the end of acidogenic phase and beginning of transition phase. Similarly, the pH of reactor A6 started to increase gradually and reached to 6.53 on 51st day. Then, the pH of both reactors A5 and A6 increased to 7.22 on 36th day and 7.21 on 56th day respectively representing entry of both the reactors into methanogenic phase. After that, the reactors' pH remained between 6.5 to 8.8 for the entire operating period and got settled in the basic state for the rest of their active period. The maximum value of pH for reactors A5 and A6 reached up to 8.28 and 8.77 respectively.

For the reactors A7 and A8 i.e. reactors with 10% of sludge and 6% horse manure added as inoculum, the initial pH was found to be 5.71 and 6.02 respectively which was measured on fourth day of operation. The lowest value of pH for reactor A7 and A8 was 4.87 and 4.97 respectively observed in 8^{th} and 11^{th} day of operation. The pH decreased rapidly in the reactor A7 as compared to A8 reactor because reactor A7 entered methanogenic phase before reactor A8. This might be because of heterogeneity of solid waste. The pH of reactor A7 and A8 started to increase gradually and reached to 6.67 on 41^{st} day and 6.51 on 56^{th} day respectively indicating the end of acidogenic and beginning of transition phase. Then the pH of reactors A7 and A8 increased to 7.25 on 56^{th} day and 7.3 on 74^{th} day respectively representing methanogenic phase. The reactors' pH remained between 6.5 to 9 for the entire operating period and got settled in the basic state for the rest of their active period. The maximum value of pH for reactors A7 and A8 reached up to 8.66 and 8.88 respectively.

For the reactors A9 and A10 i.e. reactors with 10% of sludge and 0.00000213% Manganese Peroxidase enzyme added as inoculum, the initial pH was found to be 5.59 and 5.89 respectively which was measured on fourth day of operation. The lowest value of pH for reactor A9 and A10 was 5.32 and 5.28 respectively observed in 8^{th} and 11^{th} day of operation. The pH of reactor A9 started to increase gradually and reached to 6.7on 31^{st} day indicating the end of acidogenic and beginning of transition phase. Similarly, the pH of reactor A10 started to increase gradually and reached to 6.56 on 53^{rd} day. Then the pH of reactors A9 and A10 increased to 7.01 on 51^{st} day and 7.05 on 85^{th} day respectively representing methanogenic phase. The reactors pH remained between 6.5 to 8.71 for the entire operating period and got settled in the basic state for the rest of their active period. The maximum value of pH for reactors A9 and A10 reached up to 8.45 and 8.71 respectively.The variation of pH with time for the reactors A1 to A10 is shown in Figure 4-5 and Figure 4-6.

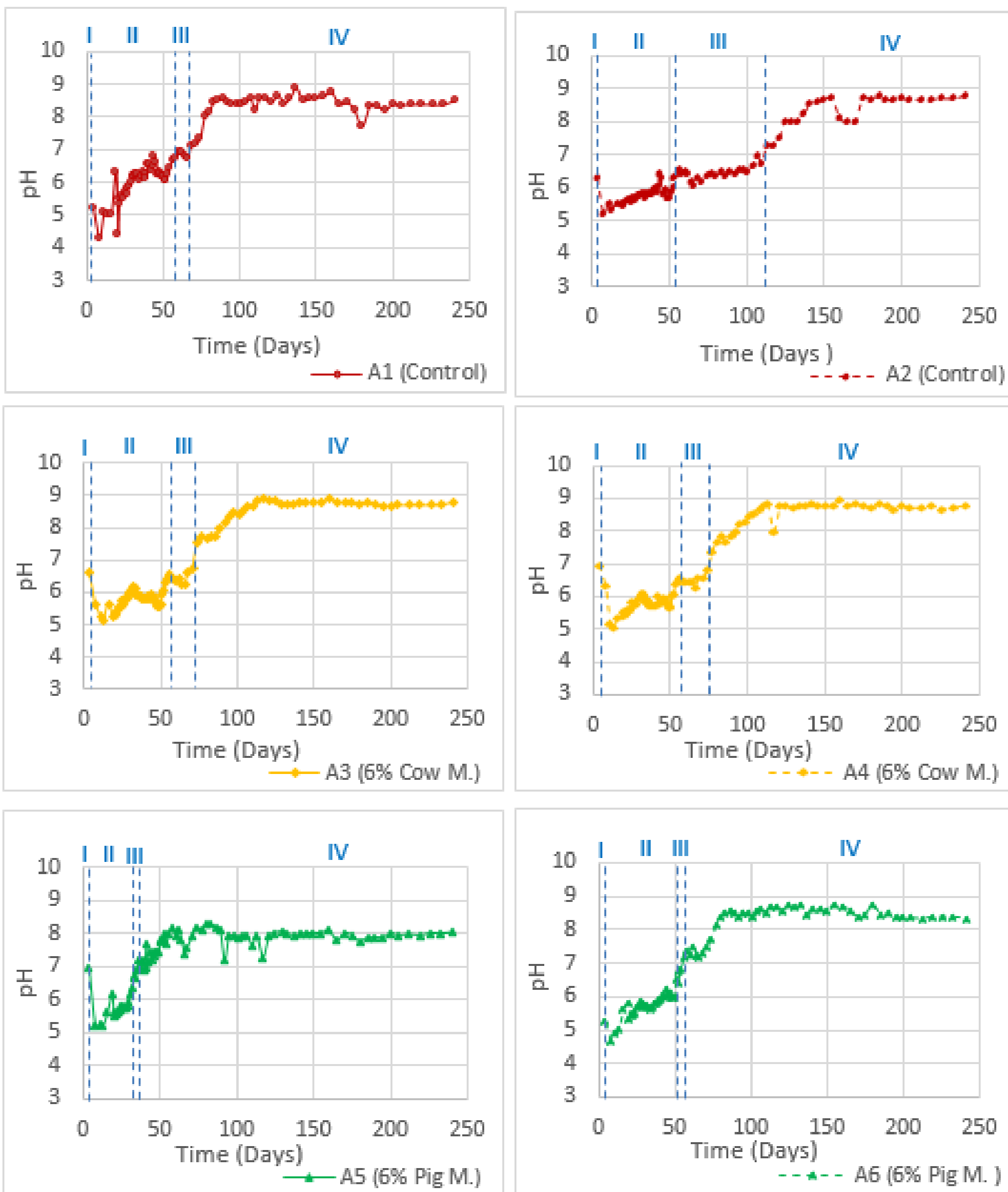


Figure 4-5 Variation of pH versus time for reactors A1 to A6

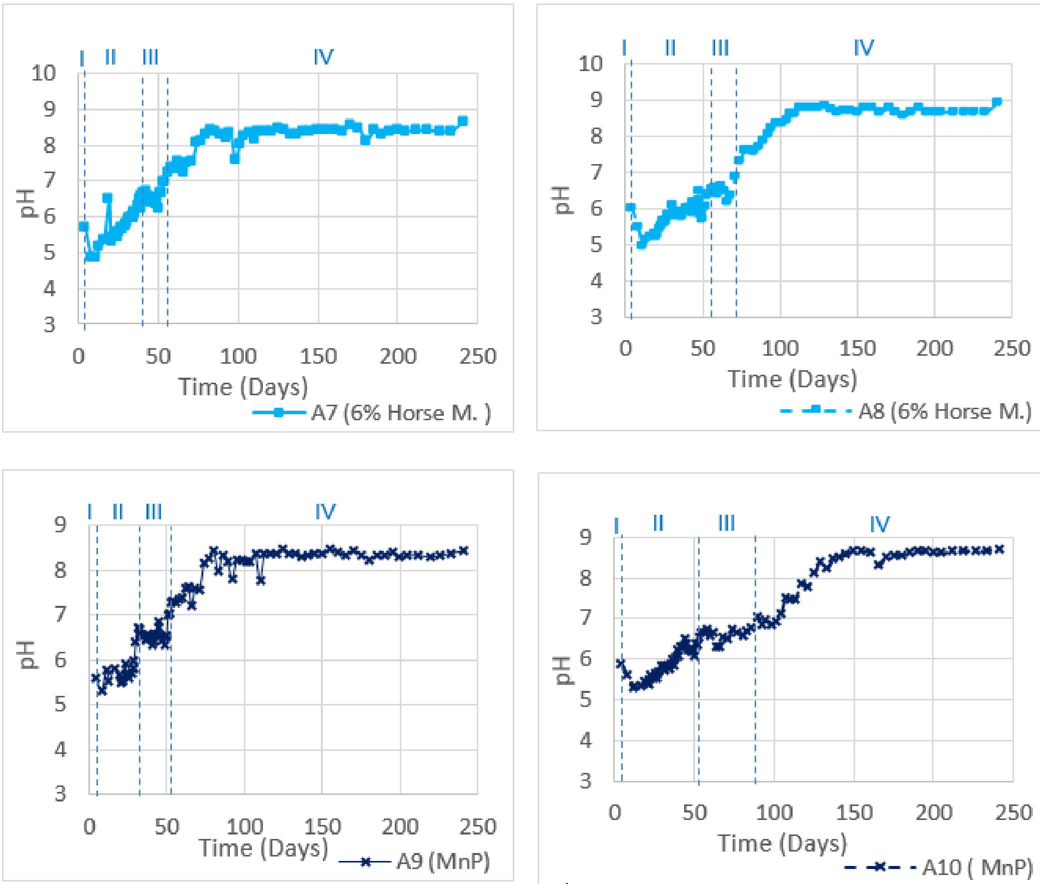


Figure 4-6 Variation of pH versus time for reactors A7 to A10

According to the measured value of pH, the A5 reactor containing 6% of pig manure reached the methanogenic phase earlier than other reactors. It took only 36 days for reactor A5 followed by reactors A9, A6, A7, A1, A3, A8, A4, A10 and A2 which took 51, 56, 56, 68, 74,74, 77, 85 and 113 days respectively. Surprisingly, all the duplicates reactor took longer time to reach the methanogenic phase which might be due to heterogeneity of MSW. Results showed that reactors with pig manure and manganese peroxidase enzyme were effective for faster degradation of organic fraction of MSW.

Warith et al. (2002) conducted the study on three set of reactors with the first set being control reactors where leachate was only recirculated, second sets with nutrients and buffer addition prior to leachate recirculation, and third sets with 5% sludge addition every week of generated total volume of leachate. The change in pH with time (Refer Figure 4-7) was found to be similar to results obtained from the reactors in this study. All the reactors in this study exhibited similar trend but the durations of lag phase were different for different reactors.

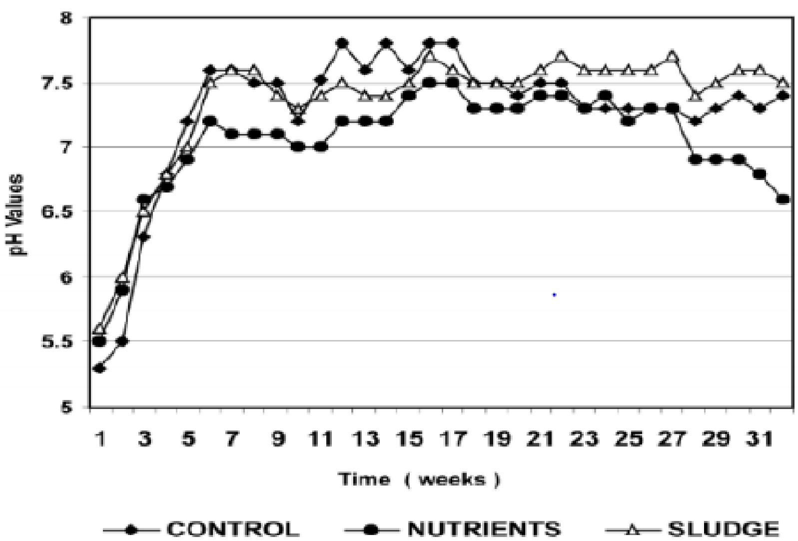


Figure 4-7 Changes in pH of leachate with time (Warith et al. 2002)

4.6.2 Volume of leachate generated

The leachate generation of all the reactors was also monitored for this study. The recirculation of leachate was done very frequently in first few months and then gradually decreased. The recirculation of generated leachate was done in order to maintain the necessary moisture for microbial activity and also to neutralize the acidic environment inside the reactors. The volume of leachate generated was measured and recirculated daily for first 54 days and then frequency of recirculation was gradually reduced to 2, 3, 4, 5 and 7 days interval after 54th, 68th, 113rd, 145th and 205th day of operation as the pH started to increase. 400 ml of leachate was recirculated. The generation of leachate from all the reactors is shown in Figure 4-8.

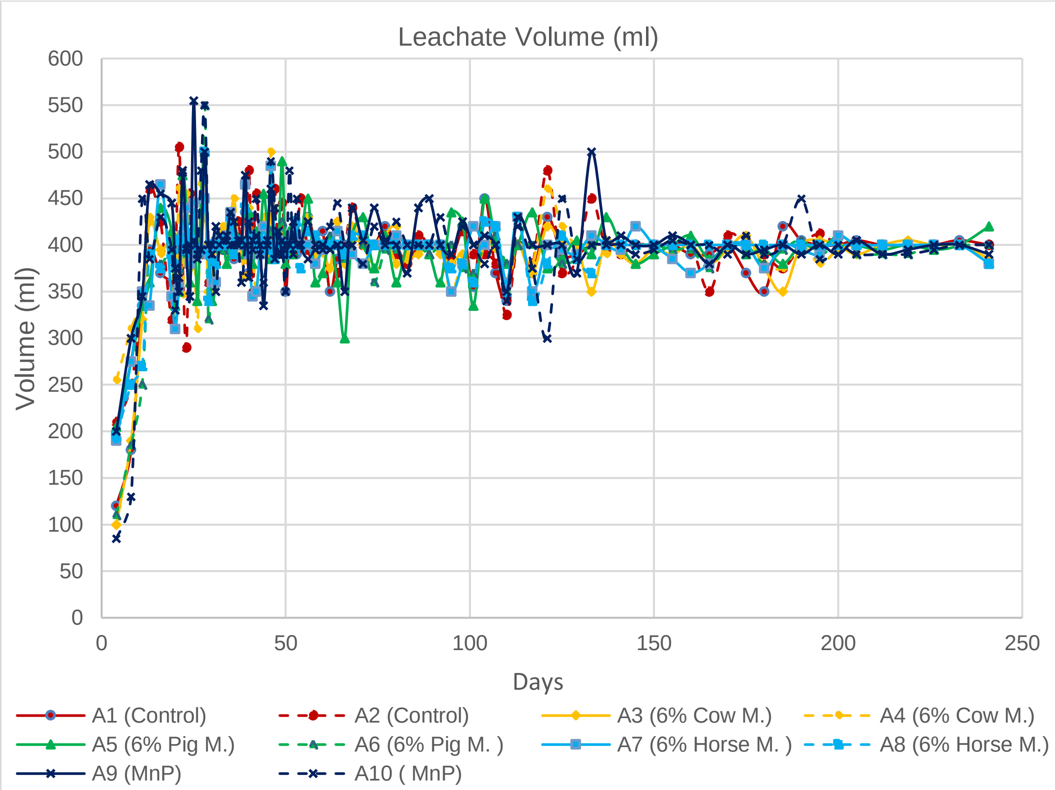


Figure 4-8 Volume of leachate generated from the reactors

For first few days, the amount of leachate generated was less than the amount recirculated because waste inside the reactor was not saturated. The waste absorbed moisture until it reached its field capacity. Despite minor variation in leachate generation, it was observed that almost all the recirculated leachate was collected in the drainage bag on the following day once it reached the field capacity. Minor variation was due to the high frequency of recirculation which did not provide enough time to drain the total amount of leachate that was recirculated. The variation in leachate generation data was reduced after recirculation was less frequent thus providing enough time to drain the circulated leachate.

The increase in the volume of leachate generated was observed during 26th and 27th day. It was due to the addition of additional 10% sludge mixed with 200 ml of water during leachate recirculation. Since pH of the leachate was very low even after 26th day of operation. Therefore, it was necessary to introduce anaerobic bacteria to reduce the extended lag phase and to neutralize the acidic environment. Therefore, the sludge was added in each of the reactors to neutralize the pH and also to reduce the lag phase.

Leachate generation is directly proportional to the gas generation and waste degradation. For reactors A5, A9 and A6 after 36 days, 51 days and 56 days of operation respectively, the volume of generated leachate was found to be less than the amount recirculated. The volume was reduced because methanogenic bacteria utilizes water to produce methane and carbon dioxide. Similar trend was observed for other reactors as well when they reached the methanogenic phase shown in Figure 4-8.

4.3.3 Chemical oxygen demand and comparison with previous studies

The Chemical Oxygen Demand (COD) tests were conducted on the leachate samples every month. Results obtained from COD tests are presented in Table 4-13 and the COD versus time graph for various combination of reactors are shown in Figure 4-9.

During the beginning of anaerobic stage, the value of COD in all the reactors started to increase due to the deficiency of oxygen which indicates the gradual change of phase from the transition phase to the anaerobic phase. The value of COD kept on increasing as the hydrolysis continued. This increase in value was followed by a drop in COD concentration as an outcome of increase in the methanogenic activity and a subsequent rise in the daily methane production. Methane generation rate played a major role in the COD variation.

Table 4-13 Variation of COD with time on monthly basis

Reactor Age (mo.)	Chemical Oxygen Demand (COD) (mg/L)									
	A1	**A2**	**A3**	**A4**	**A5**	**A6**	**A7**	**A8**	**A9**	**A10**
1	37093.18	46387.03	37386.67	33375.64	24668.77	33179.98	38071.48	38071.48	42669.49	39049.78
2	35723.56	63311.62	56659.18	47854.48	6863.71	47267.5	34843.09	56072.2	30734.23	54702.58
3	10981.93	67239.85	36864.48	37645.84	6125.82	13912.03	7856.49	40185.26	14888.73	51612.65
4	6196.1	44189.73	7075.13	9907.56	4340.37	5219.4	11079.6	12642.32	8833.19	13716.69
5	7346.7	18334.57	14281.27	14574.28	11400	13451.07	15355.64	14525.44	12083.69	9983.79
6	6543.54	15549.43	12567.3	12456.34	10984.67	11345.7	14987.45	13432.5	11345.67	9720.45
7	8372.23	13206.9	8909.42	9348.93	6711.84	9788.45	10618.64	11204.66	3293.39	6076.99

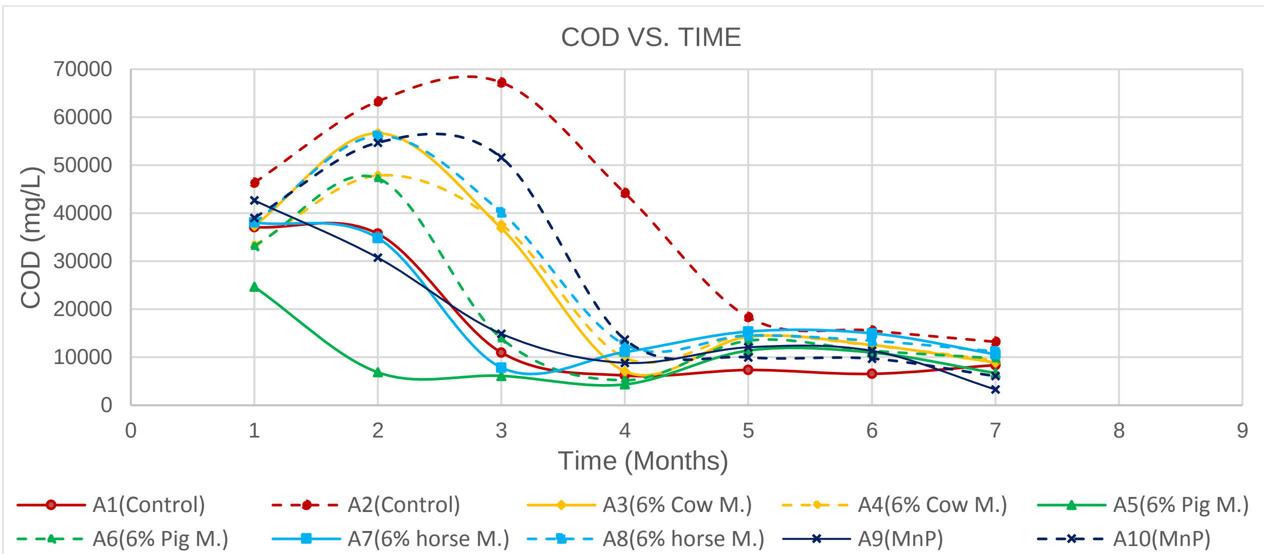


Figure 4-9 Variation of COD versus time

The less value of COD was observed for reactors A5 and A9 from the very beginning. The value of COD decreased sharply for reactors A5 and A6 on the following month. This decreasing trend showed that reactors A5 and A9 were already in methanogenic phase when COD test was conducted after one month. The initial COD values for reactors A5 and A9 are 24668.77 mg/L and 42669.49 mg/L respectively and got reduced up to 6711.84 mg/L and 3293.39 mg/L respectively. The waste degradation process started fasters on reactors A5 and A9 as compared to other reactors.

COD value for other reactors except A2 kept on increasing till the second month due to the absence of oxygen and switching of transition phase to anaerobic phase. Then, the value of COD started to decrease like A5 and A9 reactors. Similar trend was observed for reactor A2 except the COD value increased till third month and then started to decrease.

According to Alkaabi et. al. (2009), the COD value decreases initially and then increases. The initial decrease was due to aerobic phase. The COD value kept on increasing as hydrolysis continued and reached its peak value as shown in Figure 4-10. Then, COD value started to decrease as methanogenic phase started. Similar trend was observed in the current study also. The reactors A5 and A9 in the current study reached methanogenic phase earlier than the reactors with sludge (R5, R6, R7 and R8) in the study conducted by Alkaabi et. al. (2009). Therefore, it can be concluded that using of manure and sludge can help reach methanogenic phase earlier than using sludge alone.

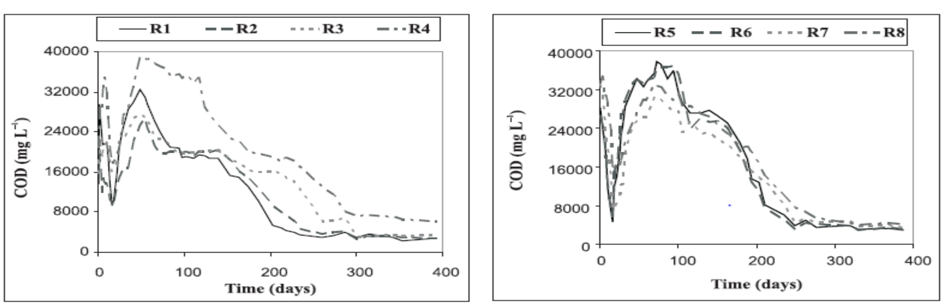


Figure 4-10 COD variation with time Alkaabi et. al. (2009)

4.6.4 Biological oxygen demand and comparison with previous studies

The Biological Oxygen Demand (BOD) tests were also conducted on the leachate samples every month to measure microbial concentration to oxidize carbonaceous and nitrogenous compounds present in the leachate. Results obtained from BOD tests are presented in Table 4-14 and the BOD versus time graph for various reactors are shown in Figure 4-11.

The BOD value for all the reactors showed a similar trend as that of COD value. The sharp decrease in BOD value was observed for reactors A5 and A9 from the very beginning. This decreasing trend showed that reactors A5 and A9 were already in methanogenic phase when BOD test was conducted after one month. The initial BOD values for reactors A5 and A9 are 16612.5 mg/L and 25800 mg/L respectively and while BOD of A5 and A9 at the end of the operation was to 187.55 mg/L and 169.92 mg/L respectively. The waste degradation process started faster for reactors A5 and A9 as compared to other reactors.

The BOD value of second month for the other reactors except A2 and A10 was found to be almost similar to first month reading which means the reactors were still in the transition phase. The sharp decrease in the value of BOD was found after second month except for reactors A2 and A10.

For the reactors A2 and A10, the value of BOD remained similar for second month and then increased rapidly on the third month. The increment on the value of BOD was due to the lack of oxygen and due to the fact that reactors were still in the transition phase. On the third month, the value of reactors A2 and A10 reached the peak value and then started to decrease. The value of BOD decreased sharply after third month indicating the beginning of the methanogenic phase.

Table 4-14 Variation of BOD with Time on monthly basis

Reactor's Age (months)	Biological Oxygen Demand (BOD) (mg/L)									
	A1	A2	A3	A4	A5	A6	A7	A8	A9	A10
1	25309	28858.6	25546.9	22320.85	16612.5	19809.35	21650.2	26500	25800	25975
2	22138.9	28308.25	22681	21817.13	3458.9	18483.57	19911.1	24861.4	18946.07	24252.43
3	5099.43	40077.77	21666.67	24250	1282.82	5297.23	2932.23	23966.67	2472.23	30557.23
4	594.97	18583.97	1925.6	3562.23	465.82	584.97	1103.27	3357.5	1005.63	5050.93
5	270.2	3663.88	614.83	1263.12	310.48	377.08	938.88	564.9	986.22	1454.17
6	305.6	3578.5	512.345	1008.76	315.67	374.678	876.59	543.71	876.9	1235.6
7	216.73	1286.12	297.8	145.62	187.55	210.53	591.67	295.92	169.92	425

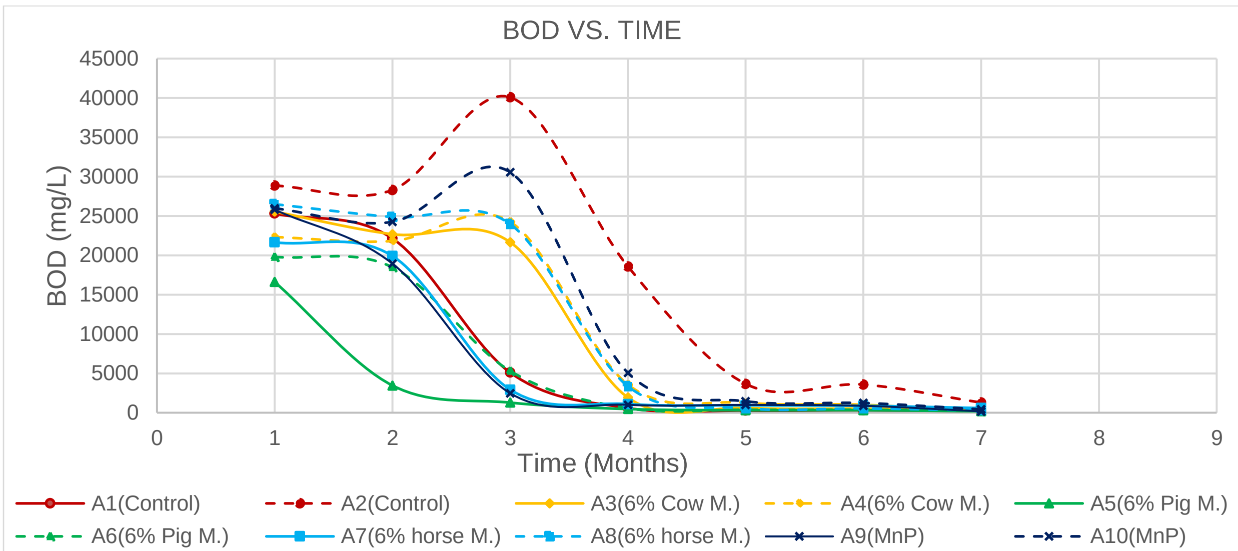


Figure 4-11 Variation of BOD versus time

Al-Kaabi (2007) reported that the variation of BOD value with time from both fresh and saline water reactor's leachate shows the similar trend as the current study. Warith (2002) reported the BOD values of the leachate for the reactors with nutrient and sludge which is shown in Figure 4-12. In that study, the amount of municipal sewage sludge added to the recirculated effluent equaled 5% of the total leachate volume. Sludge was added once in every week prior to recirculation of leachate. The decreasing trend of BOD for that study was similar to the current study except for the initial value of BOD is comparatively less than the current study. The higher concentration BOD lasted for around 5 weeks, followed by a quick decline and got stabilized after 17 weeks. In the current study, higher BOD lasted for reactors A1, A5, A6, A7, and A9 for around 2 months and then got stabilized after 3 months. Similarly, other reactors except for A2 were 1 month behind in every step compared to reactors A1, A5, A6, A7 and A9. Reactor A2 took comparatively more time to get stabilized as compared to all other reactors.

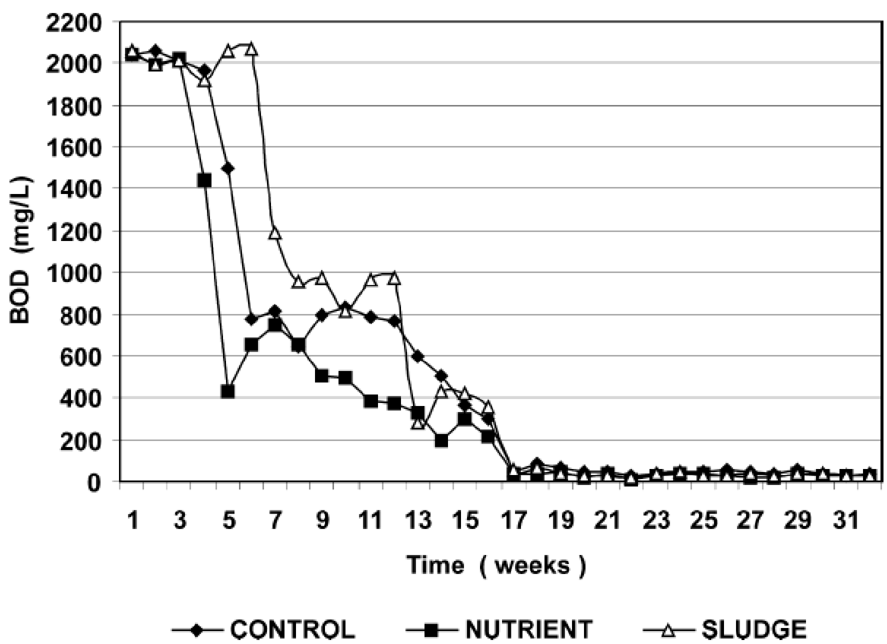


Figure 4-12 Variation of BOD value with time (Warith, 2002)

4.6.5 BOD/COD ratio variation with time

The value BOD/COD ratio provides the clear indication for the biodegradability of organic compounds present in the leachate. The BOD/COD ratios for all the reactors are shown in Table 4-15 and Figure 4-13.

Table 4-15 Variation of BOD5/COD with time

Reactors Age (months)	BOD_5/COD									
	A1	**A2**	**A3**	**A4**	**A5**	**A6**	**A7**	**A8**	**A9**	**A10**
1	0.68	0.62	0.68	0.67	0.67	0.60	0.57	0.70	0.60	0.67
2	0.62	0.45	0.40	0.46	0.50	0.39	0.57	0.44	0.62	0.44
3	0.46	0.60	0.59	0.64	0.21	0.38	0.37	0.60	0.17	0.59
4	0.10	0.42	0.27	0.36	0.11	0.11	0.10	0.27	0.11	0.37
5	0.04	0.20	0.04	0.09	0.03	0.03	0.06	0.04	0.08	0.15
6	0.05	0.23	0.04	0.08	0.03	0.03	0.06	0.04	0.08	0.13
7	0.03	0.10	0.03	0.02	0.03	0.02	0.06	0.03	0.05	0.07

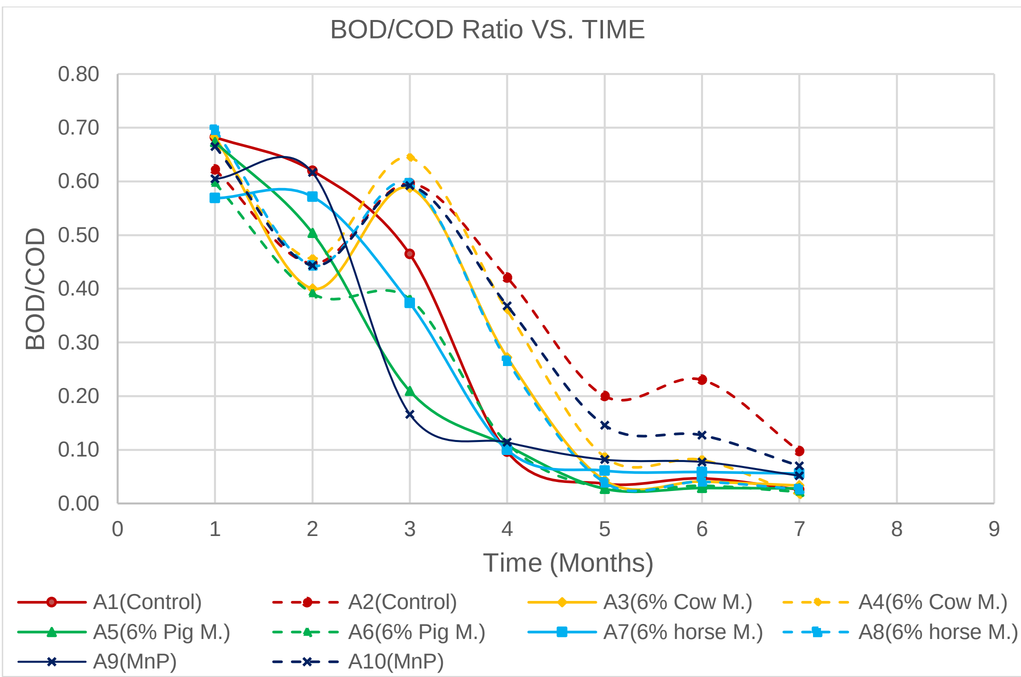


Figure 4-13 Variation of BOD/COD with time

The initial value BOD/COD ratio for all the reactors were between 0.7 to 0. 6 which means biodegradability was quite high initially for all the reactors. The value BOD/COD got reduced eventually and reached to a minimum value. The final value of BOD/COD for all the reactors were around 0.06 except A2. The value of BOD/COD for all the reactors except A2, A4, A9, and A10 were below 0.06 within 5 months of operation which indicated the reactors were in methanogenic phase.

Wraith (2002) reported that if the ratio BOD_5/COD was between 0.4 to 0.8 than the leachate is highly biodegradable. Kjeldsen et al. (2002) concluded that acidic phase will start when the value of BOD_5/COD ratio reached around 0.58 and 0.06 for methanogenic phase. The change in phases of degradation for all the reactors were clearly identified by the data suggested by Wraith (2002) and Kjeldsen et al. (2002).

4.7 Gas generation

The composition and volume of gas generated were measured on regular basis. The composition, volume, and rate of gas generated from each of the reactors are described in the subsection below. The gas generation data for reactors were also compared with the previous studies.

4.7.1 Gas composition

The composition of gas generated was measured using Landtec GEM 2000 in percentage. The composition includes the percentage of methane, carbon dioxide, oxygen and other gases. The other gases is referred as balance here. Figure 4-14 and Figure 4-15 shows gas composition data for all the reactors individually.

The composition of generated gas indicated the presence of methane, carbon dioxide, oxygen and other gases. The percentage of oxygen in all the reactors was less from the very beginning. Since the size of the reactor was very small, the amount of oxygen trapped in the voids was less too. Therefore, aerobic phase did not last for a long time. Due to the lack of available oxygen, the percentage of carbon dioxide and balance (e.g. H_2S, nitrogen ammonia, hydrogen etc.) started to increase in all the reactors. This increase in the percentage of carbon dioxide was due to the acidogenic phase where degradable organic compounds were broken into simpler compounds like carbon dioxide and water vapor. The peak percentage of carbon dioxide was observed on the 8th day when first gas data was measured. The percentage of CO_2 was around 60% for all the reactors. With time, the percentage of carbon dioxide and balance started to decrease and methane percentage increased for all the reactors.

The percentage of methane was below 2 percentage for all the reactors till 20th-25th day which was due to excessive accumulation of Volatile Fatty Acids. Accumulation of Volatile Fatty Acids decreased the leachate pH value causing the prolonged lag phase of around 20-25 days for all the reactors indicating acidogenic phase. The additional 10% of sludge was added to all reactors on 25th day of operation. The addition of sludge neutralized the acidic environment inside the reactors. Immediately after addition of sludge, the percentage of methane started to increase in reactor A5 and A6 with pig manure, and reactor A9 and A10 with Manganese Peroxidase. However, the percentage of methane for other reactors were still less than 2 %. The reactors A3 and A4 with cow manure and reactors A7 and A8 with horse manure started to produce methane from 33rd day while lag phase for A1 and A2 continued to 37th and 41st day respectively. The transition phase existed between acidogenic and methanogenic phase. During this phase, the percentage of carbon dioxide reached 40% of the total gas composition. While in the methanogenic phase, the percentage of methane and carbon dioxide will be more as compared to other gases. In this stage, the methane percentage rises up to 60~65% and the pH of the leachate varies between 6.0 and 8.5 (Karanjekar, 2013). The highest percentage of methane generated for all the reactors in the current study was found in the range of 60~65%.

All the reactors were monitored for 241 days. The gas generation ceased early on reactors A1 and A2 (control) on 110^{th} and 175^{th} respectively, and A7 and A8 with horse manure on 107^{th} and 104^{th} day of operation. The gas production for reactors A3 with cow manure continued until 221 days while its duplicate reactor A4 showed gas for only 107 days. This indicated the possibility of the leak in reactor A4. The gas generation continued for 241 and 233 days on reactors A5 and A6 with pig manure respectively, and 226 and 241 days on reactors A9 and A10 with MnP.

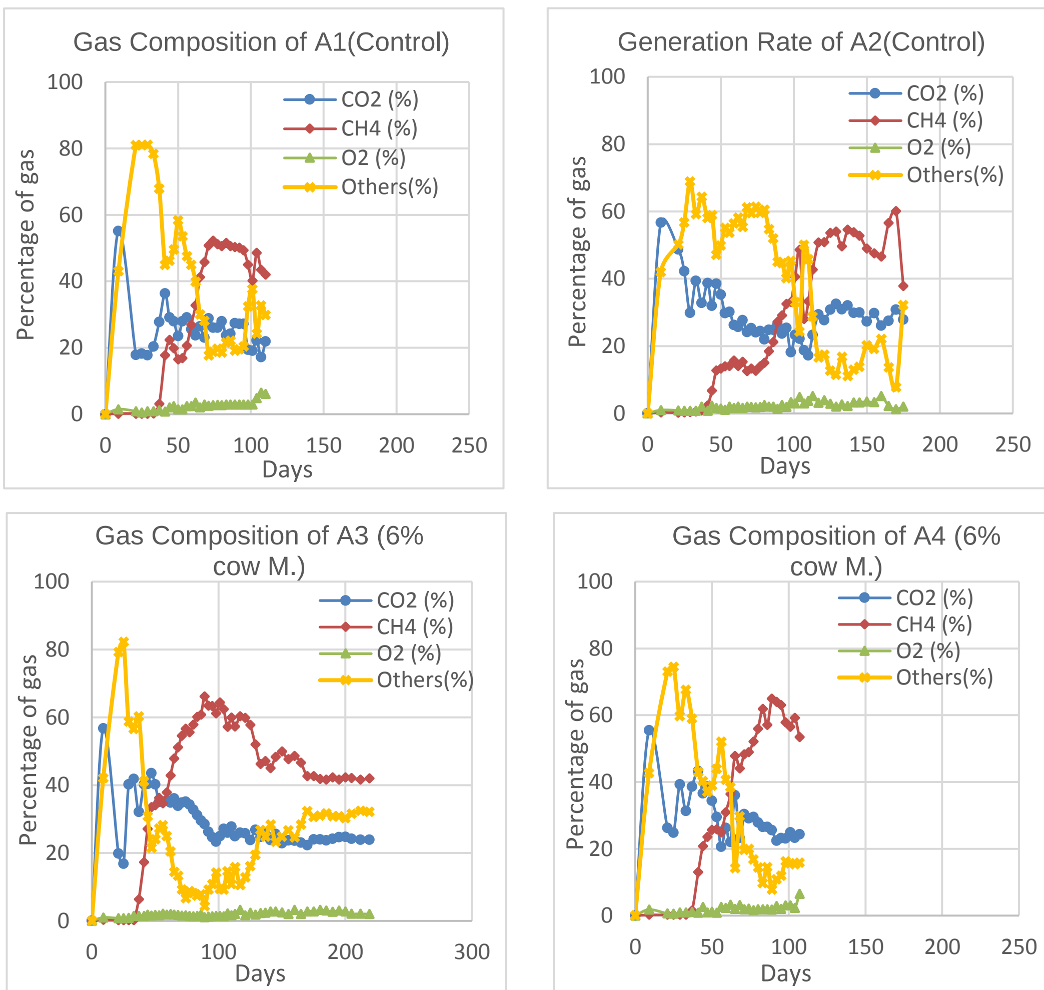


Figure 4-14 Gas composition data for reactors A1 through A4

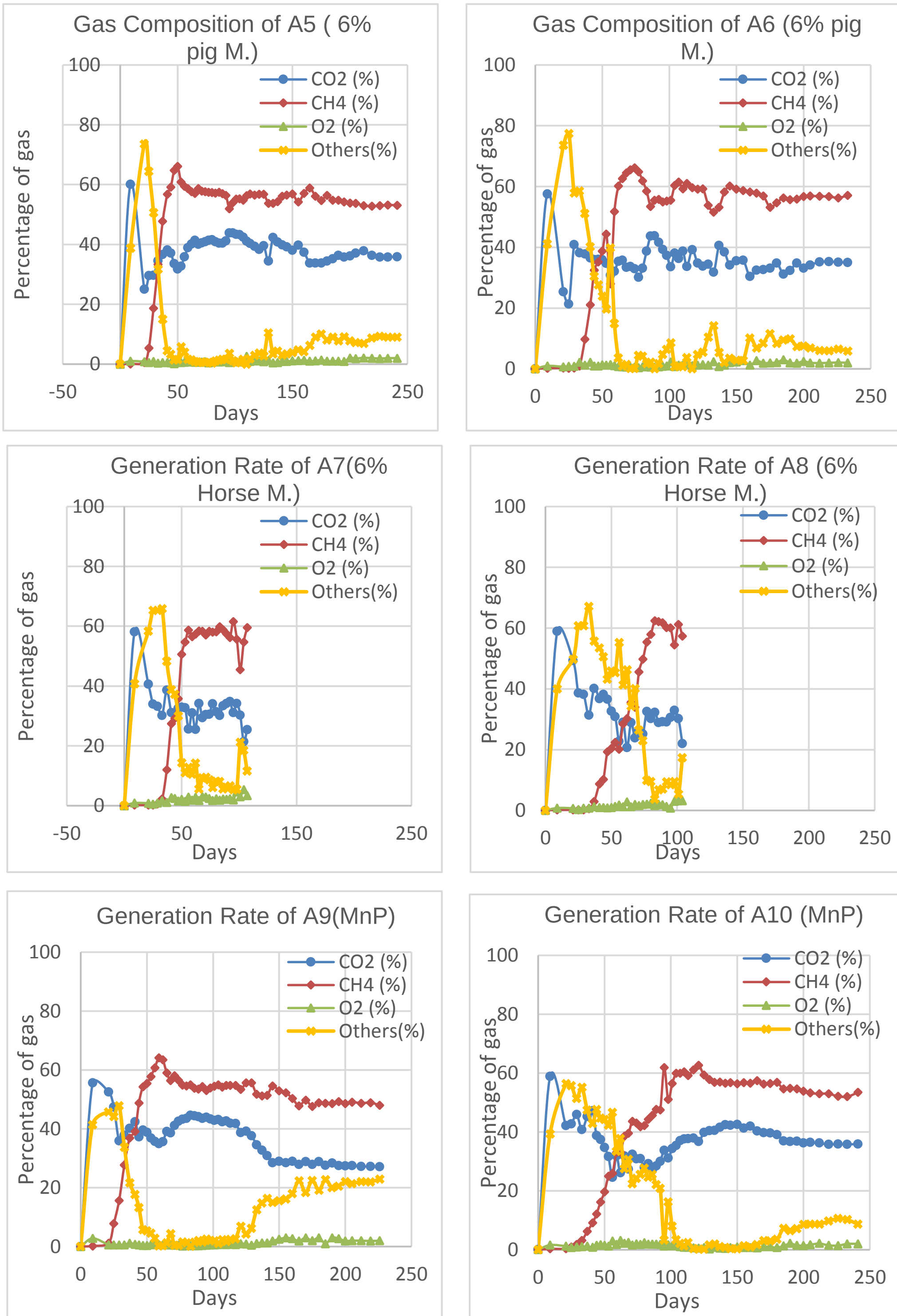


Figure 4-15 Gas composition data for reactors A5 through A10

The variation of methane percentage with time for the reactors A1 through A10 is shown in Figure 4-16. The percentage of methane was very less for all the reactors till first 20 days. The reactors A5 with pig manure and A9 with MnP started producing methane after 20 days of operation. Within 37 days of operation, all the reactors started producing methane. The highest percentage of methane recorded in gas composition was 66.20% and 66.10% for reactor A5 and A6 respectively (with pig manure) followed by reactors with cow manure. The highest percentage of methane observed in reactors A3 and A4 with cow manure was 66.10% and 64.9 %. The percentage went as high as 64.1% for A9 and 62.6% for A10 with MnP. The lowest percentage of methane was observed in the control reactors A1 and A2 which was 52.3 and 60.1% respectively.

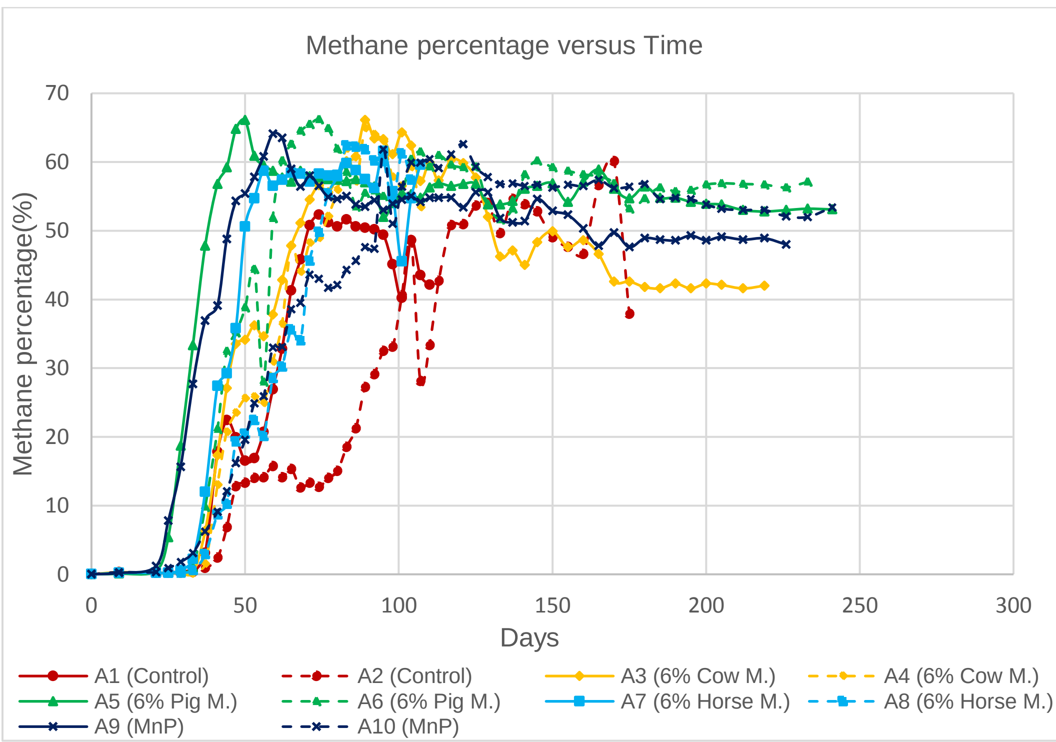


Figure 4-16 Comparison of percent methane with time for the reactors

The percentage of carbon dioxide was about 60% when measured on the 8th day which was due to the breakdown of organics into simpler compounds. Since the percentage of methane was less than 1 for all the reactors, the ratio of CH_4 to CO_2 was nearly zero. The percentage of carbon dioxide started to decrease while the percentage of methane was still less which was due to excessive acid accumulation. After the lag phase, the percentage of methane started to increase. As the percentage of methane increased ratio of CH_4 and CO_2 also increased. The increasing scenario of methane and decreasing carbon dioxide is shown in Figure 4-17. The ratio of CH_4 and CO_2 for all the reactors were with the range of 1.3 to 2.8 during the methanogenic phase.

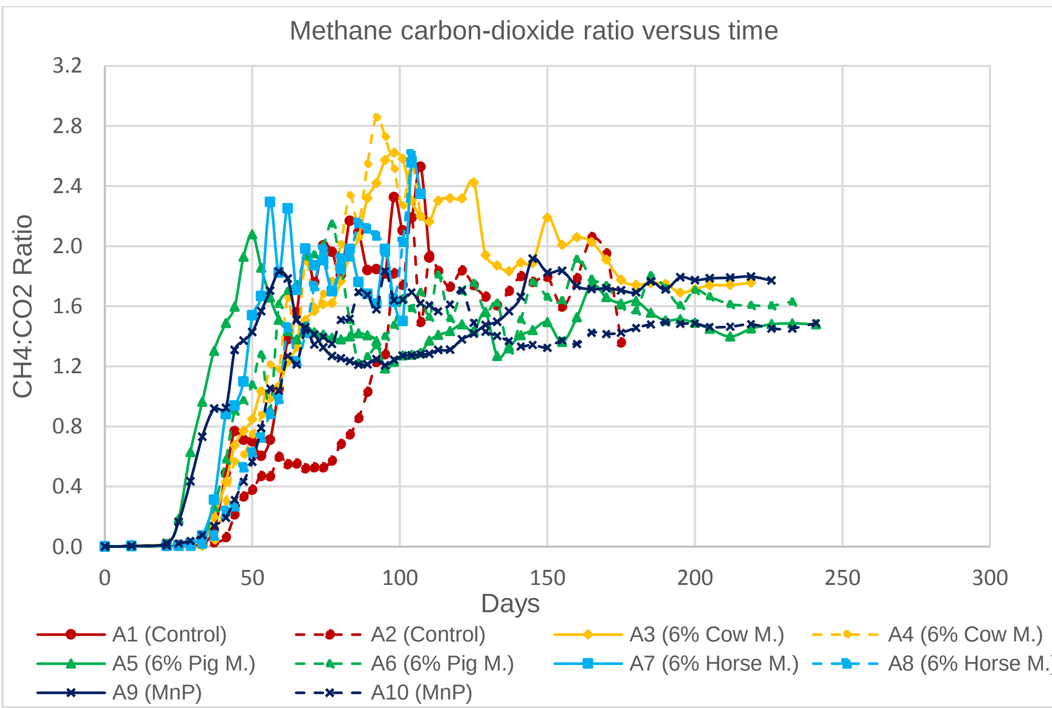


Figure 4-17 Methane to Carbon-di-oxide ratio over time

4.7.2 Volume of generated gas

The total volume of gas generation from all the reactor is shown in Figure 4-18. The gas generation in all the reactors started from the very beginning. First gas volume data for all the reactors were measured on the 8th day after the operation. The volume of gas was very less for all the reactors initially due to the lag phase. The 20 days of lag phase period was observed in the cumulative gas versus time graph in Figure 4-18. The addition of sludge on 26th day eliminated the lag phase simultaneously for all reactors. Once the reactor reached methanogenic phase, gas production bumped up. The cumulative volume of gas remained almost constant and, got stabilized for all reactors except A10 with MnP and A6 with pig manure after 150 days. The volume of gas kept on increasing for reactor A6 and A10 until it reached 200 days and finally got stabilized.

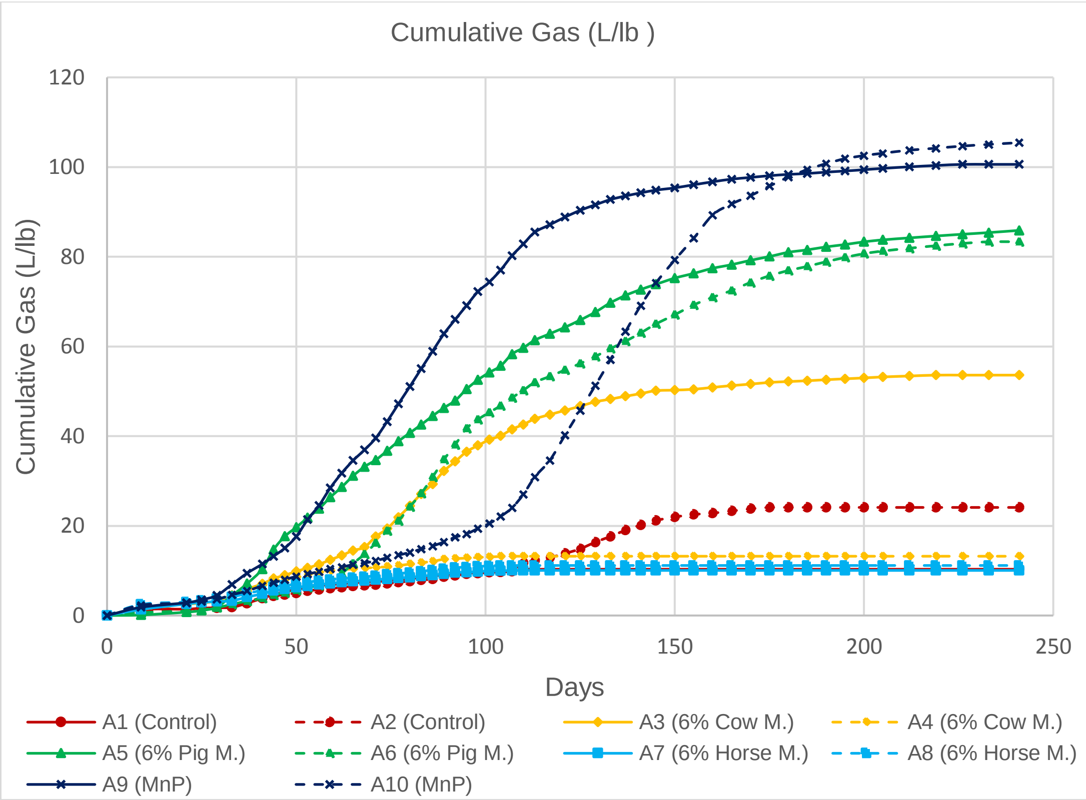


Figure 4-18 Cumulative gas generated versus time for all reactors

The highest amount of gas was generated on the reactors A9 and A10 with MnP followed by reactors A5 and A6 with pig manure and then reactor A3 with cow manure. The lowest amount of gas was generated on the control reactors(A1 and A2) and reactors A7 and A8 with horse manure. The cumulative volume of gas generated by reactors A9 and A10 was 100.60 L/lb. and 105.40 L/lb. respectively, and for reactors, A5 and A6 were 85.88 L/lb. and 83.34 L/lb. respectively. The cumulative volume of gas generated by reactor A3 and A4 was 53.60 L/lb. and 13.21 L/lb. respectively. The volume of gas generation for duplicate reactor A4 with cow manure was less than reactor A3 with the same feedstock which might be due to the leakage in the system. The control reactors (A1 and A2), and reactors with horse manure (A7 and A8) produced less gas as compared to other reactors.

The cumulative methane versus time graph is shown in Figure 4-19. The almost similar trend was observed in the cumulative methane versus time graph except the methane generation started only after lag phase. Only reactor A5 with pig manure and A9 with MnP started producing considerable amount methane after 20 days. The considerable amount of methane was observed in all other reactors only after 50 days. The difference in the duration of lag phase for the same combination of feedstock might be due to the heterogeneity of waste. Even though the composition of paper, food, yard, and textiles was fixed, the heterogeneity still exists among different types of paper, food, yard, and textile waste. The highest volume of methane was generated by reactors A9 and A10 which produced 54.7 L/lb. and 52.03 L/lb. respectively, followed by reactors A 5 and A6 which produced 47.26 L/lb. and 45.91 L/lb. respectively. The cumulative volume of methane generated by reactor A3 was 25.5 L/lb. The volume of methane generated by all other reactors was less than 10 L/lb.

From the cumulative methane generation data, it can be said that enzyme and pig manure in combination with sludge was found to be a better inoculum as compared to other manures for decomposition of organic fraction of MSW.

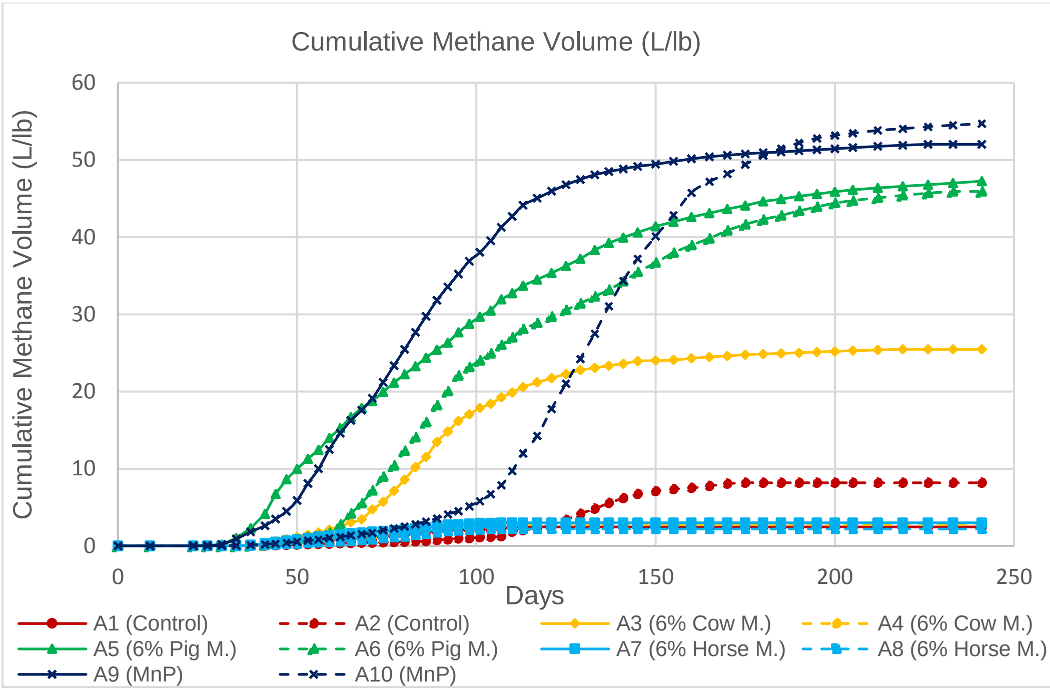


Figure 4-19 Cumulative Methane generated versus time for all reactors

The rate of total gas generation and methane generation are shown graphically in Figure 4-20 and Figure 4-21. All the reactors' gas generation and methane generation rate reached its peak value within first 100 days of operation except for reactors A2 and A10. The reactors A2 and A10 have experienced prolonged acidogenic and transition phase. The peak value of methane yield observed from reactors A9 and A10 was 1.33 and 1.57 L/lb./day respectively followed by reactors A5 and A6 for which the methane yield was 1.45 and 1.30 L/lb./day and then reactor A3 for which methane yield was 0.97 L/lb./day. All other reactors' methane yield was less than 0.4 L/lb./ day except for reactor A2. The peak methane yield of reactor A2 was 0.57 L/lb./day.

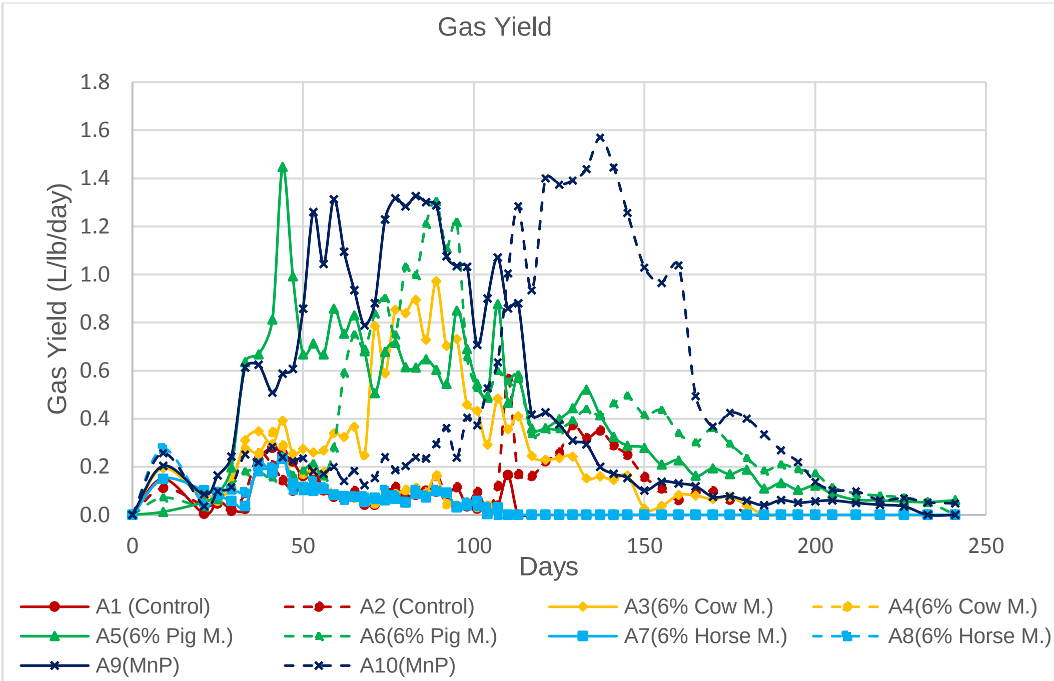


Figure 4-20 Gas yield versus time for all reactors

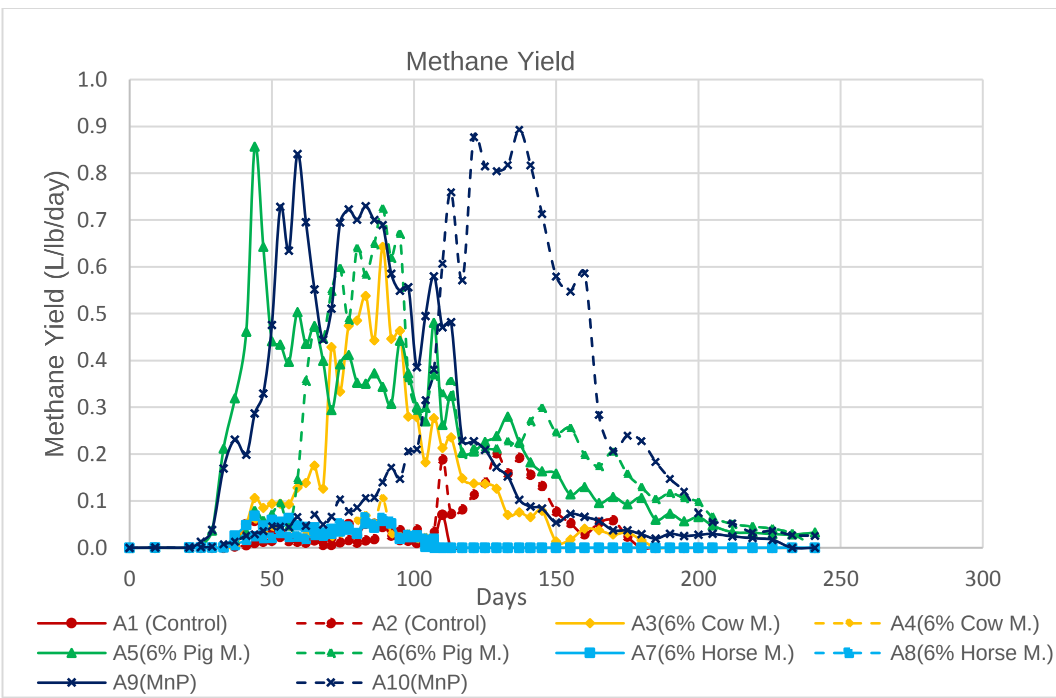


Figure 4-21 Methane yield versus time for all reactors

4.7.3 Comparison with previous studies

According to the study conducted by Ali et. al. (2016), cow dung was used as an inoculum for degrading organic municipal waste. The percentage of Cow Manure (CM) used in the reactors Rc, R1, R2 and R3 were 0%, 10%, 20% and 30% respectively. The biogas yields at the end of the digestion from the reactors Rc, R1, R2 and R3 cow dung as inoculum were 173 ml / g VS, 337.365 ml / g VS, 481.95 ml / g VS and 567ml / g VS respectively. The cumulative methane yield was based on the percentage of volatile organic content reduced. Figure 4-22 shows the cumulating biogas generation with time based on Ali et. al. (2016).

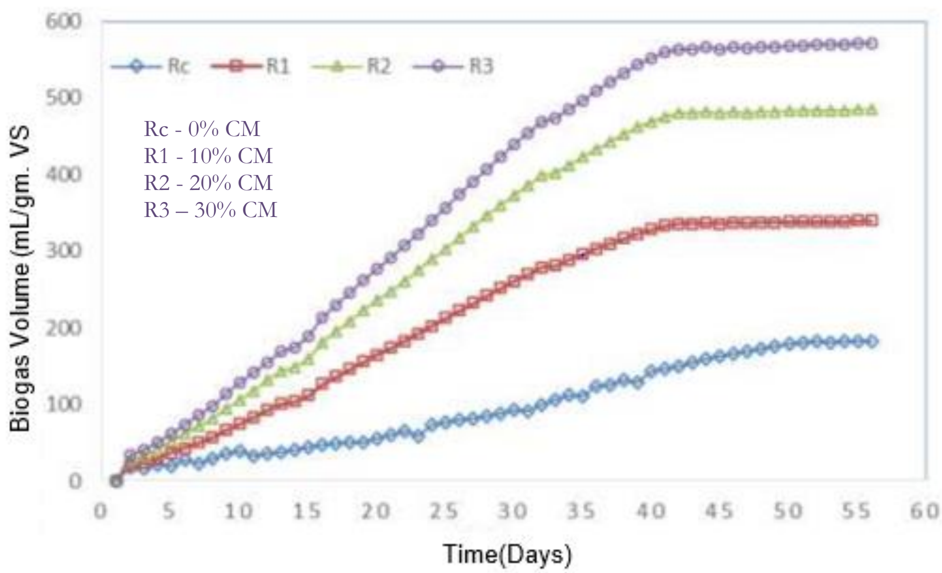


Figure 4-22 Cumulating biogas generation with time (Ali et. al., 2016)

In the current study, the cumulative biogas yield from the reactors with 6% cow manure was 53.60 L/lb. which is equivalent to 279.5 ml/ g of VS when calculated using the percentage of volatile organic content reduced. The overall VS removal percentage for reactor with cow dung was found to be 67.88%.

According to the study conducted by Yazdani (2010) at the Yolo County Central Landfill in California, digester cell was operated in two stage batch system where the organic matter was degraded under anaerobic conditions followed by aerobic conditions. During the anaerobic phase of 451 days, the total volume of biogas generated was 1.48 x 105 m^3 and the methane yield was 6.07 x 10^4 m^3, or 27.2 m^3 of methane per Mg of solid which is shown in Figure 4-23.

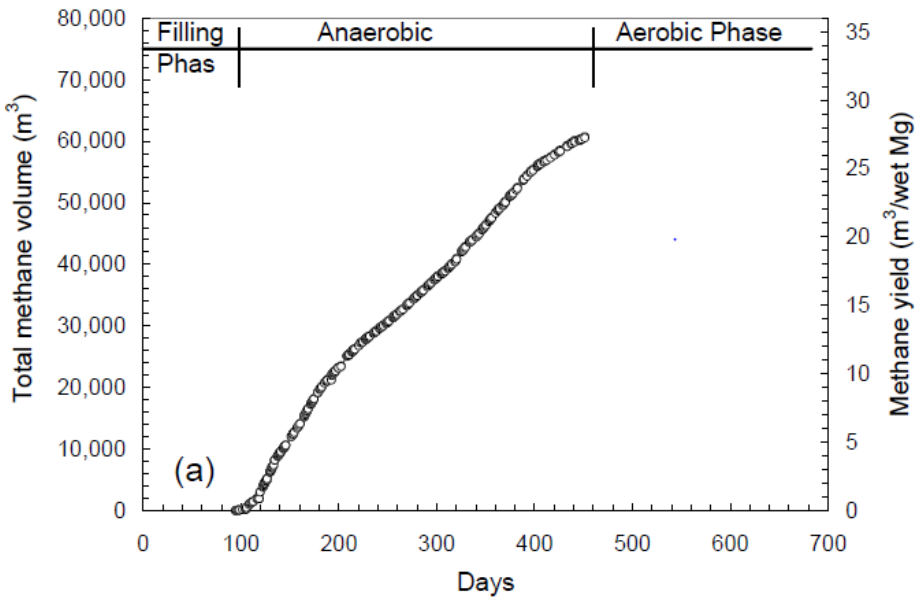


Figure 4-23 Total methane volume and methane yield for digester cell during anaerobic phase (Yazdani, 2010)

In the current study, the methane yield from the reactors with horse manure was very less. It might be due to the age of horse manure. Horse manure used in this study was less than a week old. The cumulative methane yield from reactors with horse manure was 6.59 and 4.81 L/kg which is equivalent to 6.59 m^3/Mg and 4.81 m^3/Mg. But, the methane production from reactors with pig manure and MnP exceed the cumulative methane produced on the study conducted by Yazdani (2010). Therefore, the effect of pig, cow manure and enzyme as inoculum showed the better results on gas generation data.

4.8 Weight loss and height settled of reactor's feedstock after degradation

The percentage weight loss of waste by decomposition is shown in Table 4-16 and Figure 4-24. The highest percentage of weight loss was found in reactors with MnP (A9 and A10), followed by reactors with pig manure (A5 and A6). The percentage of weight loss due to decomposition directly correlates with the gas generation data. The highest gas generation was also found in reactors with MnP. Similarly, the maximum settlement in height of feedstock was 3.5 inch observed for reactors A5, A6, A9 and A10.

Table 4-16 Weight loss due to decomposition

Reactors	Initial Dry weight of waste (lb.)	Final Dry weight of waste (lb.)	Weight loss (lb.)	Weight loss (%)
A1(Control)	1.47	0.9	0.57	39.02
A2(Control)	1.47	0.87	0.6	41.07
A3(Cow M.)	1.47	0.72	0.75	51.31
A4(Cow M.)	1.47	0.78	0.69	47.22
A5(Pig M.)	1.49	0.67	0.82	55.25
A6(Pig M.)	1.49	0.62	0.87	58.28
A7(Horse M.)	1.45	0.85	0.61	41.87
A8(Horse M.)	1.45	0.87	0.58	40.22
A9(MnP)	1.45	0.61	0.85	58.32
A10(MnP)	1.45	0.6	0.86	59.01

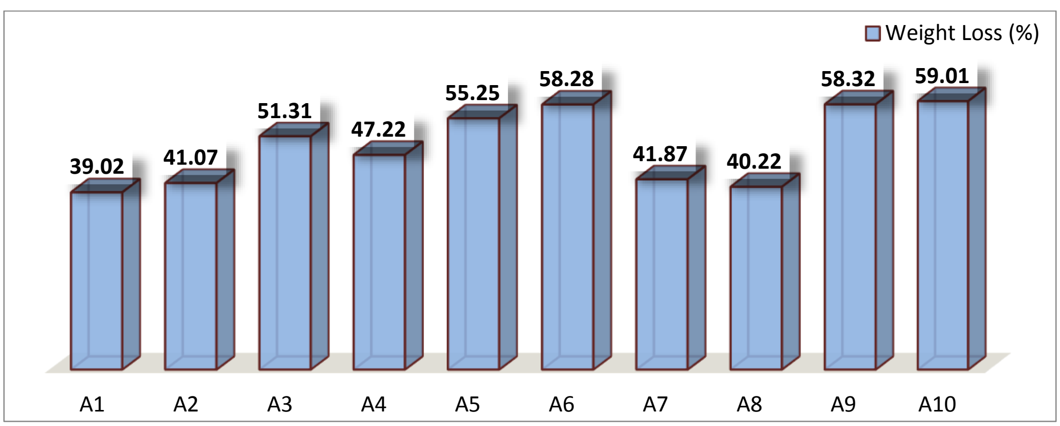


Figure 4-24 Percentage of weight loss after decomposition

The percentage height settled of waste by decomposition is shown in Table 4-17 and Figure 4-25. The highest percentage of settlement was observed in reactors with MnP (A9 and A10) and reactors with pig manure (A5 and A6). The percentage of settlement due to decomposition also directly correlates with the gas generation data. The highest gas generation was also found in reactors with MnP, followed by reactors with pig manure. Similarly, the maximum settlement in height of feedstock was 3.5 inch observed for reactors A5, A6, A9 and A10.

Table 4-17 Height settled after decomposition

Reactors	Initial Height of waste (inch.)	Final Height of waste (inch.)	Height Settled (inch)	Height Settled (%)
A1(Control)	7.5	5.5	2	26.67
A2(Control)	7.5	5	2.5	33.33
A3(Cow M.)	7.5	4.25	3.25	43.33
A4(Cow M.)	7.5	4.5	3	40
A5(Pig M.)	7.5	4	3.5	46.67
A6(Pig M.)	7.5	4	3.5	46.67
A7(Horse M.)	7.5	5	2.5	33.33
A8(Horse M.)	7.5	5	2.5	33.33
A9(MnP)	7.5	4	3.5	46.67
A10(MnP)	7.5	4	3.5	46.67

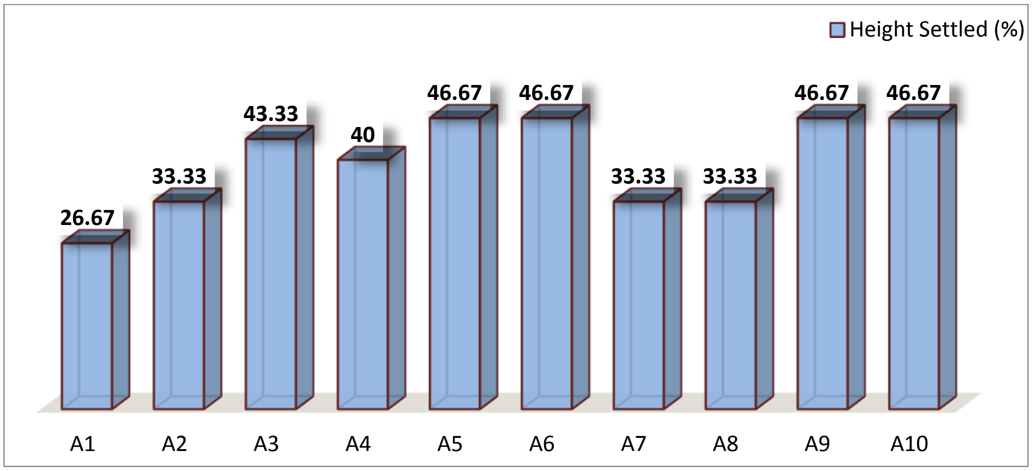


Figure 4-25 Percentage of height settled after decomposition

Therefore, percentage of height settled and weight loss of feedstock after decomposition also showed that MnP and pig manure could be better inoculum for Biocell landfill.

4.9 Moisture variation after degradation

The final moisture contents of all the reactors' feedstock were determined and shown in Table 4-18. The moisture contents of the feedstock of each reactor were determined before putting the waste into the reactor and at the end of the study after dismantling. The moisture contents of degraded samples were found be more when compared to the initial values. The final moisture content of the reactor A1, A2, A7, and A8 were found to be almost similar to that of the initial value. The moisture holding capacity is dependent upon the degree of decomposition.

Table 4-18 Variation of moisture content after degradation

Reactors	Initial Moisture content of reactor (%)	Final Moisture content of reactor (%)
A1(Control)	62.97	63.32
A2(Control)	62.97	62.72
A3(Cow Manure)	61.06	74.00
A4(Cow Manure)	61.06	70.48
A5(Pig Manure)	63.08	75.05
A6(Pig Manure)	63.08	75.97
A7(Horse Manure)	66.34	69.44
A8(Horse Manure)	66.34	69.72
A9(MnP)	64.13	73.64
A10(MnP)	64.13	72.83

4.10 Volatile solids variation after degradation

The volatile organic contents of degraded waste were determined to study the effect of manures on the degradation of organic fraction of MSW. The volatile organic content of degraded waste and fresh waste for all the reactors' feedstock are tabulated in

Table 4-19. The change in volatile organic content for reactors with pig manure and MnP was more than other reactors after 241 days of operation.

Table 4-19 Comparison of volatile solid in fresh and degraded MSW

Sample No.	Initial VS (%)	Final VS (%)
A1(Control)	86.71	63.33
A2(Control)	86.71	62.07
A3(Cow M.)	84.41	55.36
A4(Cow M.)	84.41	57.41
A5(Pig M.)	87.04	48.21
A6(Pig M.)	87.04	48.39
A7(Horse M.)	86.4	63.33
A8(Horse M.)	86.4	62.50
A9(MnP)	84.96	46.43
A10(MnP)	84.96	46.67

The actual removal of volatile solid after 241 days of operation was computed in Table 4-20 and Figure 4-26.

Table 4-20 Volatile solid removal percentage for total weight of feedstock

Reactor	Total Initial VS Dry weight (lb)	Total Final VS Dry weight (lb.)	Overall VS removal (%)
A1(Control)	1.27	0.57	55.28
A2(Control)	1.27	0.54	57.63
A3(Cow Manure)	1.24	0.40	67.88
A4(Cow Manure)	1.24	0.45	63.91
A5(Pig Manure)	1.30	0.32	75.09
A6(Pig Manure)	1.30	0.30	76.87
A7(Horse Manure)	1.25	0.54	57.03
A8(Horse Manure)	1.25	0.54	56.60
A9(MnP)	1.23	0.28	77.01
A10(MnP)	1.23	0.28	77.27

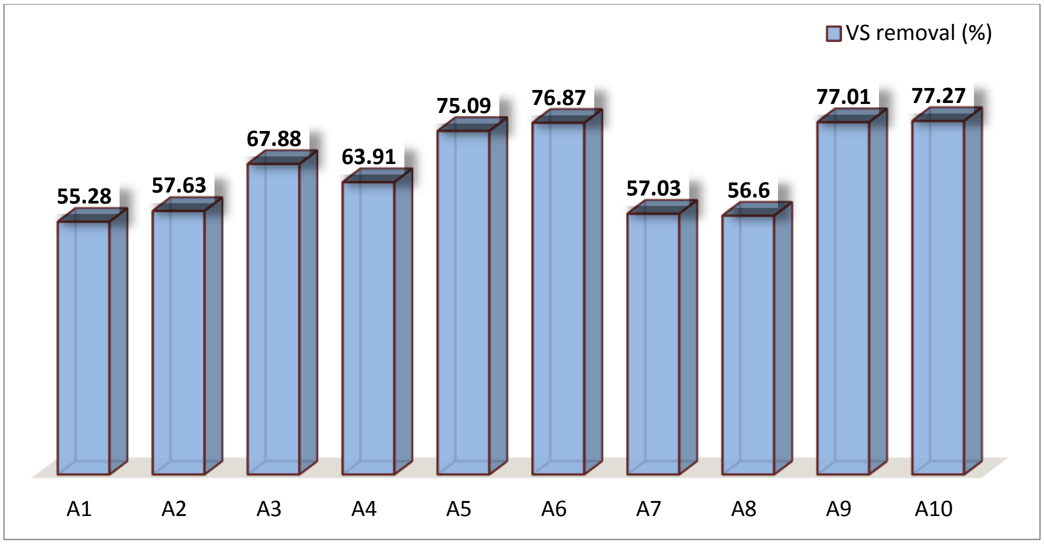


Figure 4-26 Volatile organic content removal after degradation

The reduction in volatile solids in this current study was comparable to previous studies. Several studies have been conducted to determine volatile solids content after degradation. The study conducted by Haque (2007) showed that the initial volatile organic content found to be 91.5 % and then the final volatile organic content percentage was decreased up to 46 %. In this study, also, the volatile solid reduced up to 46.63% for the reactor with MnP and 48.21% for reactors with pig manure. The study conducted by Al-Kaabi et al. (2009) reported that the reduction in volatile organic content were found to be 84%, 78%, 74%, and 66% after anaerobic degradation when leachate was recirculated with the salinity level of 0%, 0.5%, 1.0%, and 3% respectively. Sivanesan (2012) reported that the reduction in volatile solids were 84%, 82%, 77%, and 70% for the reactors with salinity values of 0%, 0.5%, 1.0%, and 3% respectively when the sludge was added. Therefore, the addition of sludge has a positive effect on the removal of volatile organic content.

Chapter 5

Conclusion and Recommendation

5.1 Summary and conclusion

The current study mainly focused on the organic fraction of Municipal Solid Waste(MSW) with the sustainable waste management concept. Handling organic fraction of waste has always been a challenge for the solid waste industry. Evolution of bioreactor landfilling concept solved most of the issues associated with conventional landfill such as ground water contamination, leachate generation, waste stability and gas generation issues except the issue of sustainability. Therefore, a novel concept of Biocell was evolved in which the sustainability is achieved by reusing the same space again and again. Faster degradation of waste is the first step for the landfill to be sustainable. Therefore, enhancement of degradation process is necessary which can be achieved by leachate recirculation, leachate enhancement, and recirculation, nutrient and buffer addition, co-digestion and addition of inoculum such as manure, sludge, degraded waste, old leachate etc. Few studies have been conducted on the laboratory simulated landfill reactors with the addition of sludge only. However, the study on the effect of different types of manure on the degradation of MSW had not been conducted in laboratory simulated landfill. Therefore, the decision to conduct this study was made.

The main objective of the study was to determine the effect of manure and enzyme on the degradation of the organic fraction of MSW. To achieve that goal, characterization of collected MSW and inoculum was done. The organic waste was sorted manually from the MSW collected from city of Denton landfill. The composition of organic waste used in the reactors was based on the composition of degradable waste from the City of Denton landfill. A total of five pair of reactors were built with one pair acting as the control and other four pairs with varying inoculum type (cow manure, pig manure, horse manure, MnP enzyme). Recirculation and monitoring of generated leachate along with the measurement of gas generated was done periodically. The monitoring of leachate included pH measurement, volume measurement, COD and BOD tests whereas composition, rate, and volume of gas generated were measured for monitoring of gas. The results obtained from the current study is summarized as follows:

1. A total of six bags of sample were collected from the working face of the City of Denton Landfill. The sample composition was determined by manual sorting. On average, the samples were composed of –34% paper, 19% plastic, 13% food waste, 8% textile, 2% styrofoam and sponge, 9% yard and wood waste, 3% metals, 2% glass, 4% construction debris and 6% others (soils and fines).
2. The average degradable fraction of fresh waste was 64.74% while non-degradable was 35.26%.Degradable fraction was further classified into 51.47% of paper, 21.88 % of food waste, 12.83% of textiles and leather and 13.81% of yard and wood waste.
3. Only degradable waste was fed into the reactors. Based on the degradable waste composition of collected MSW from City of Denton Landfill, the composition for reactors' feedstock was fixed. The reactors' feedstock contained of 50% paper,20% food waste, 15% textiles, and 15% yard and wood waste
4. The average moisture content of fresh MSW was found to be 37.93% on dry weight basis and 27.22% on wet weight basis. The moisture content of sludge , cow manure, pig manure and horse manure was found to be 75.67%, 2%, 5% and 36% respectively.
5. Initial moisture content of the feedstock was found to be 61.06 to 66.34% and Initial volatile organic content was found to be 84.41% to 87.04 %.The moisture content and volatile content for all reactors was found to be similar because the composition of waste was identical in all reactors except the type of manure used.

6. The pH of all the inoculum used was determined before using it. The pH of sludge, cow manure, pig manure and horse manure was found to be 8.37, 8.95,7.81 and 7.69 respectively. All the manures were aged (6 months to a year) except for horse manure which was only a week old.
7. A total of five pair of reactors were built with one pair for control and other four pair with varying inoculum type (cow manure, pig manure, horse manure, MnP enzyme). 10% sludge was common for all the reactors. The 6% manures and 0.00000213% enzyme was added on wet weight basis. Inoculum was added for accelerated degradation of organic fraction of MSW in Biocell operation.
8. The initial pH levels for all the reactors were less than 7 due to the accumulation of acid in acidogenic phase. Then, pH values for all the reactors started to rise and got stabilized in between 7 and 8 due to the conversion of fatty acids into methane and carbon dioxide.
9. The pH of reactor A5 containing 6% of pig manure reached the methanogenic phase earlier than other reactors. It took only 36 days for reactor A5 followed by reactors A9, A6, A7, A1, A3, A8, A4, A10 and A2 which took 51, 56, 56, 68, 74,74, 77, 85 and 113 days respectively. Surprisingly, all the duplicates reactor took longer time to reach the methanogenic phase which might be due to heterogeneity of MSW. The pH results showed that reactors with pig manure and manganese peroxidase enzyme were more effective for faster degradation of organic fraction of MSW.
10. The volume of leachate generation is related to the gas generation and waste degradation process. When the reactor reached the methanogenic phase, the volume of generated leachate was found to be less than the amount recirculated. The volume was reduced because methanogenic bacteria started utilizing water to produce methane and carbon dioxide. The volume of generated leachate started to decrease

early for reactor A5, A9 and A6 after 36 days, 51 days and 56 days of operation respectively indicating methanogenic phase.

11. The value of COD started to decrease sharply for reactors A5 and A6 from the first month. While COD value for other reactors kept on increasing till third month due to the absence of oxygen and switching of transition phase to anaerobic phase. The decreasing value of COD indicated the beginning of methanogenic phase. The BOD value of leachate samples also showed a similar trend as the COD concentration in all reactors.
12. The initial value of BOD/COD ratio for all the reactors were between 0.7 to 0.6 which means biodegradability was quite high initially for all the reactors. The value BOD/COD got reduced eventually and reached to a minimum value. . The final value of BOD/COD for all the reactors were around 0.06 except A2. The value of BOD/COD for all the reactors except A2, A4, A9 and A10 were below 0.06 within 5 months of operation which indicated the reactors were in methanogenic phase.
13. The highest amount of gas was generated in the reactors A9 and A10 with MnP, followed by reactors A5 and A6 with pig manure and then reactor A3 with cow manure. The lowest amount of gas was generated on the control reactors A1 and A2 and reactors A7 and A8 with horse manure.
14. The cumulative methane versus time graph showed the similar trend as cumulative gas versus time except the methane generation started only after lag phase. The highest volume of methane was generated by reactors A9 and A10 which produced 54.7 L/lb. and 52.03 L/lb. respectively, followed by reactors A 5 and A6 which produced 47.26 L/lb. and 45.91 L/lb. respectively. The cumulative volume of methane generated by reactor A3 was 25.5 L/lb. The volume of methane generated by all other reactors was less than 10 L/lb.

15. Gas generation and methane generation rate versus showed the similar trend. Both gas and methane generation rate reached its peak value within first 100 days of operation except for reactors A2 and A10. The reactors A2 and A10 have experienced prolonged acidogenic and transition phase. The peak value of methane yield observed from reactors A9 and A10 was 1.33 and 1.57 L/lb./day respectively followed by reactors A5 and A6 for which the methane yield was 1.45 and 1.30 L/lb./day and then reactor A3 for which methane yield was 0.97 L/lb./day. All other reactors' methane yield was less than 0.4 L/lb./ day except for reactor A2. The peak methane yield of reactor A2 was 0.57 L/lb./day
16. From all the results of gas generation rate and volume, and methane generation rate and volume, the reactors with MnP (A9 and A10) and reactors with pig manure showed more generation rate and volume as compared to other reactors. Therefore, MnP and pig manure were the most effective inoculum for generating more gas as compared to other inoculums used in this experiment.
17. The highest percentage of weight loss was found in reactors with MnP (A9 and A10), followed by reactors with pig manure (A5 and A6). The percentage of weight lost during decomposition directly correlates with gas generation. Therefore, pig manure and MnP were the most effective inoculum in degrading organic fraction of MSWs as compared to other inoculums.
18. The final moisture contents of the reactor A1 , A2, A7 and A8 was found almost similar to that of initial values. However, the increment in moisture content was observed in other reactors. The moisture holding capacity for the reactors with MnP (A9 and A10) and with pig manure (A5 and A6) was found to be more after degradation. The moisture holding capacity increase with the degradation of waste. Therefore, result showed that reactors A5, A6, A9 and A10 degraded more as compared to other reactors.

19. The overall volatile solid removal percentage after degradation for reactors with pig manure and MnP was found to be more as compared to others. The percentage removal of VS for reactors A5 and A6 with pig manures was 75.09 and 76.87% respectively and for reactors A9 and A10 with MnP was 77.01 and 77.2% respectively. The VS removal efficiency of pig manure and MnP enzyme as inoculum was found to be more effective than others.
20. Based on the experimental results, it can be summarized that the addition of inoculum increased the waste degradation potential. The rate and volume of gas production was found to be more in the reactors with pig manure and MnP as inoculum. Therefore, pig manure and MnP enzyme can be utilized as inoculum for improving the degradation potential of waste.
21. The cost for MnP enzyme is comparatively higher. 10 mg of MnP enzyme cost $134. Even though MnP enzyme helped to degrade more waste and produced highest volume of methane gas, it is not considered an economical solution for field application. Therefore, pig manure is suggested as a better inoculum for sustainable bio cell landfill.

5.2 Recommendations for future studies

Based on the findings of this study and literature review following recommendations are suggested for further research:

1. Further research needs to be done to find out the optimum percentage of manure as inoculum which can further enhance the gas generation.
2. Further research should be done by using different cheaper cellulose enzyme as inoculum for degrading organic fraction of MSW and to compare it’s the gas generation potential with pig manure as inoculum.

3. Further study is necessary to find out the effect of age of manures on the waste decomposition and to find out the optimal age which can yield more biogas.
4. Further studies should be conducted to reduce the lag phase by using anaerobically digested manure.
5. Further studies should be conducted by varying the composition of degradable waste. Since, this study is conducted as per the waste composition of the City of Denton, landfill where paper constitutes half of the fraction of feedstock.

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