TECHNICAL FEASIBILITY OF ANAEROBIC DIGESTION FOR UTILIZING MUNICIPAL SOLID WASTE FROM TALAAD THAI MARKET, THAILAND

BY

TA TUAN ANH

A THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE
REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE
(ENGINEERING AND TECHNOLOGY)
SIRIDHORN INTERNATIONAL INSTITUTE OF TECHNOLOGY
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A Thesis Presented

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Submitted to
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Abstract

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TA TUAN ANH

Bachelor of Engineering (Environmental Technology and Management), Van Lang University, 2013
Master of Science in Engineering and Technology, Sirindhorn International Institute of Technology, Thammasat University, 2018

Agriculture is the most important sector in the economy of Thailand. The data reported by Food and Agriculture Organization (FAO) states that Thailand produces over 10 million tonnes of fruit and vegetables annually. As a result, large amounts of fruit and vegetable waste rich in biodegradable organics are generated. Anaerobic digestion (AD) is a potential technology from an environmental point of view and also beneficial to society as it provides a clean fuel and fertilizer.

In this study, the mixed wastes from the vegetable market in Talaad Thai was used as the feedstock. The granular sludge from up-flow sludge blanket reactor, Pathum Thani Brewery Co., Ltd., Thailand, was used as the inoculum. To determine the influence of feedstock to inoculum ratio (F/I) and temperature on methane generation, biochemical methane potential (BMP) experiments were conducted at different F/I ratios at 37°C and ambient temperature (30±4°C). The effects of external alkalinity were also studied by adding $NaHCO_3$ concentrations ranging from 0 to 1200 mg/g VS_{added}. To confirm again the optimum temperature, BMP assays were conducted at optimum $NaHCO_3$ concentration and two temperature conditions of 37 and 50°C. The reuse of digestate from anaerobic digesters as sources of alkalinity and inoculum was also investigated in the study. The optimum conditions found out in the BMP tests were applied for the lab-scale BIOCEL reactor.

Results and findings obtained in this study are summarized as follows:

- After 53 days of experiment time, without adding $NaHCO_3$, the optimum conditions of F/I ratio and temperature were at 0.5 and 37°C, respectively. The results depicted that the higher the F/I ratio, the lower biogas and methane yield. The F/I of 0.5 at the temperature of 37°C gave the highest biogas and methane yield at 851 mL/g VS and 306 mL/g VS, respectively.
- At the F/I ratios of 2.0, 3.0, 4.0, and 5.0 g VS/g VS, higher hydrogen production was observed for both temperature conditions.
- The results also showed that adding buffer improved the biogas production and methane contents at F/I of 1.0 and 2.0, significantly.
- When the optimum condition of $NaHCO_3$ was operated at temperature of 37 and 50°C, the achieved results depict a better performance at reactors of 37°C. The thermophilic reactors shorten the digestion time compared to the mesophilic reactors. However, the final biogas and methane yields at thermophilic reactors were lower than that of mesophilic reactors.
- The study found an economic option to reduce bicarbonate adding into AD reactors by recycling digestate as an inoculum source for new processes. When the digestate is recycled, the sodium bicarbonate can be reduced at the concentration of 150 mg per gram VS added.
- At the optimum conditions obtained from BMP test, AD performance of the lab-scale reactors was higher than that of BMP reactors. When using UASB sludge as inoculum, the methane yield was 404 and 756 mL/g VS in BMP and lab-scale reactor, respectively. When using digestate as inoculum, the methane yield was 395 and 672 mL/g VS in BMP and lab-scale reactor, respectively.

In conclusion, this study successfully demonstrated the potential of utilizing the mixed vegetable waste from Talaad Thai market as a renewable energy source through anaerobic digestion.

Keywords: vegetable waste, biomethane potential, anaerobic digestion, biogas, renewable energy

Acknowledgments

This work would not have been possible without the support of many well-wishers at Sirindhorn International Institute of Technology (SIIT), Thammasat University.

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Chapter 1

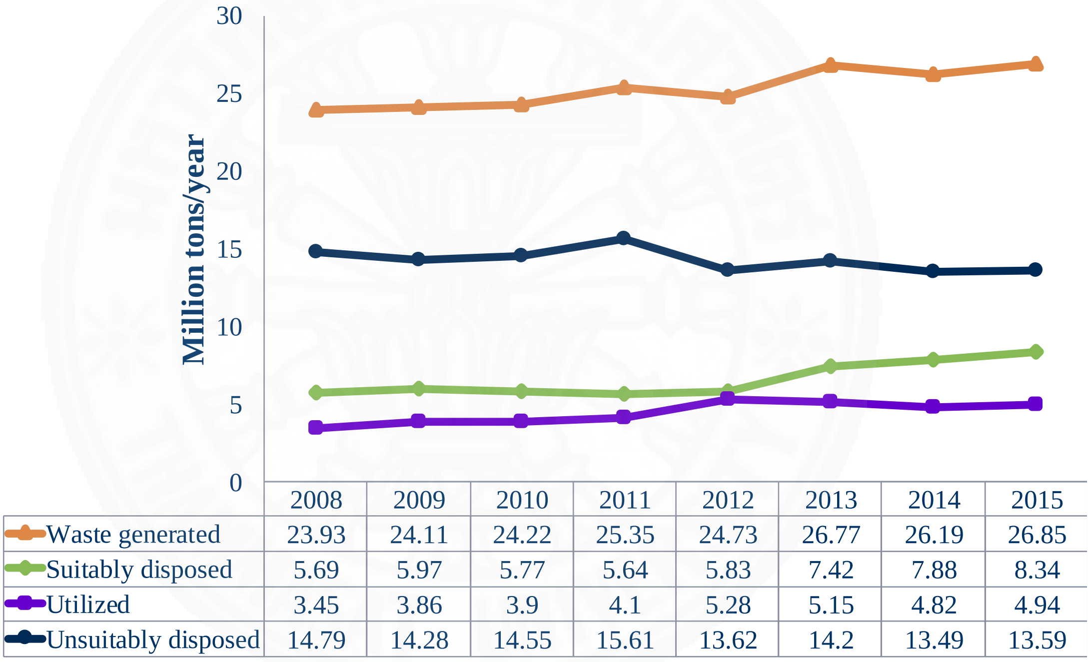
Introduction

1.1 World population prospects and solid waste generation

According to the results of United Nations (2015), the world population reached 7.3 billion in 2015, implying that the world has added approximately one billion people in the span of the last twelve years. The world population is projected to increase by more than one billion people within the next 15 years, reaching 8.5 billion in 2030, and to increase further to 9.7 billion in 2050 and 11.2 billion by 2100. In 2010, more than half world's population, 3.3 billion people, lived in urban areas. By 2030, the number is expected to increase to almost 5 billion. Already in the year 2000, there were at least 23 megacities with the population of more than 10 million. Most of these cities were located in developing countries.

As a result of the increasing number of population and the improvement of living quality since the past three decades, the total amount of municipal solid waste (MSW) is continuously rising. The annual rise of the solid waste amount can be estimated about 2 - 3% (Salhofer et al., 2008). As a report of World Bank, current global MSW generation levels are approximately 1.3 billion tonnes per year, and are expected to increase to approximately 2.2 billion tonnes per year by 2025 (Hoornweg and Bhada-Tata, 2012). The annual waste generation in East Asia and the Pacific Region is approximately 270 million tonnes per year.

In Thailand, the total amount of municipal solid waste generated in 2015 was 26.85 million tonnes or 73,560 tonnes per day. Out of this number, 4.19 million tonnes were generated in Bangkok (16%) and 22.66 million tonnes were generated in the other provinces (84%) (PCD, 2016). Fig. 1.1 shows the trend of MSW generation in Thailand from 2008 to 2015. As shown in the Fig, high amount of MSW in Thailand is unsuitably disposed or dumped into landfills. Improper handling and disposal of the huge amount of solid waste seriously affect the air, land and water environments and human health. The problem associated with direct landfills or open dumping are (1) the possibility of water table contamination by the leachate, (2) the low potentiality of biogas utilization, and (3) the difficulty of finding suitable areas near the sites where the waste is being generated. It is also contributing to climate change by releasing methane and carbon dioxide. As reported by Pollution Control Department, Ministry of Natural Resources and Environment Thailand, 64% of the waste generation in Thailand are recyclable organic wastes (PCD, 2016). Therefore, it is possible to reduce the amount of waste dumped into landfills by applying treatment technology for the organic wastes. Concerning the treatment of solid waste, the anaerobic digestion of solid waste has been studied in recent decades, trying to develop a technology that sums up advantages for volume and mass reduction as well as for energy and resources recovering.



	2008	2009	2010	2011	2012	2013	2014	2015
Waste generated	23.93	24.11	24.22	25.35	24.73	26.77	26.19	26.85
Suitably disposed	5.69	5.97	5.77	5.64	5.83	7.42	7.88	8.34
Utilized	3.45	3.86	3.9	4.1	5.28	5.15	4.82	4.94
Unsuitably disposed	14.79	14.28	14.55	15.61	13.62	14.2	13.49	13.59

Figure 1.1: Trend of MSW generation in Thailand (PCD, 2016)

1.2 State of solid waste management in Talaad Thai Market

In this study, solid waste generated from Talaad Thai market was chosen as a case study for applying anaerobic digestion. The market is the biggest and the most modern integrated center for agricultural and industrial goods in Thailand. It was found in 1997 and administrated by Thai Agro Products Company Limited. Talaad Thai is located in Klong Luang district in the Pathumthani province of Thailand, within the boundary of the Bangkok Metropolitan Region.

This market is approximately 80 ha and is home to the distribution center for domestic and international agricultural products, especially fruits and vegetables. Talaad Thai serves as a collection and distribution channel of agricultural products not only in Thailand but also for South East Asian countries. There are more than 15,000 tons of produce passing through the market every day with a daily trade value of 400 – 600 million Baht.

According to Ghaffar et al. (2012), Talaad Thai generates 120 tonnes of waste per day, of which almost 85% is organic and hence suitable for use in renewable energy, especially biogas production. The current waste management system and processes are displayed in Fig 1.1.

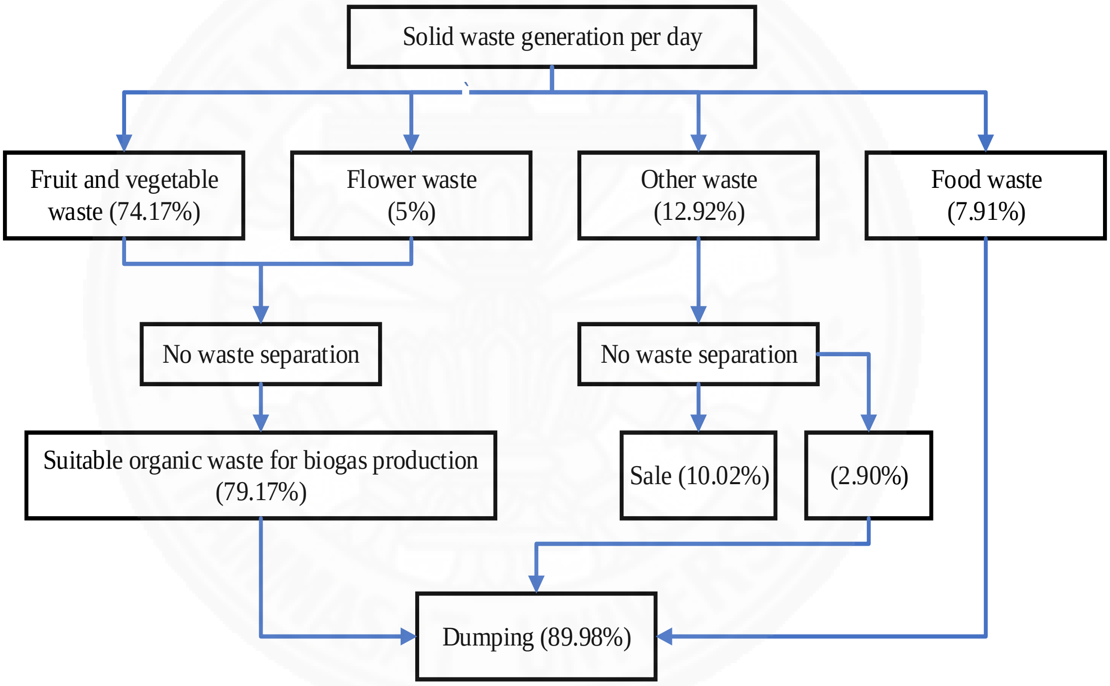


Figure 1.2: Current solid waste management system in Talaad Thai (Ghaffar et al. 2012)

1.3 Problem statement

As shown in Fig 1.1, 90% the organic waste from Talaad Thai is dumped into landfills, except for a few tons of vegetable leaves are sold to the local people as animal feed (Ali et al., 2012). Talaad Thai does not own any landfill sites, the management contracts private landfill sites situated almost 60 km away from the market. A total of 12 trucks are appointed for waste handling from the market to the Bangsai disposal site. From the 12 trucks, 6 are used for the collection and transfer of waste within the market itself, while 6 are used for outside transfer, that is, for transporting solid waste from the market to a disposal site in Ayutthaya province. As a result, almost Thai baht 10 million is spent for the cleaning and disposal of the waste from this market. If anaerobic digestion (AD) system is installed for the organic waste treatment, it would increase the profits and reduce environmental impacts through converting these wastes into biogas for energy production and a compost product from the digestate.

The AD is a biological conversion process in the absence of external electron acceptor like oxygen. In the process, organic carbon is converted to its final form of oxidation and reduction state (CO_2, CH_4) (Angelidaki and Sanders, 2004). The AD process, not only offers benefits by treating the solid waste, but also yields the biogas for power generation, and when possible the digestate can be used as fertilizer in agriculture. However, even though many studies were done on the field of the AD of solid waste, still the application of this process is limitedly practiced, especially, in developing countries due to the lack of appropriate treatment system configurations. Before starting a full-scale AD reactor, biochemical methane potential (BMP) assays should be investigated. BMP assays help analyzing types of substrates, having the highest biomethane potential among various substrates. The assays also determine the optimum ratios between substrates and inoculums; and evaluate the anaerobic biodegradation rate of substrates or retention time required for complete the digestion (Elbeshbishy et al., 2012). This research was conducted to evaluate the feasibility to develop an AD system of mixed vegetable wastes from the largest agricultural wholesale market in Thailand – Talaad Thai.

1.4 Objectives

The main goal of the study is to determine the optimum conditions of AD for utilizing vegetable waste from the Talaad Thai market. In order to reach the goal, this study comprises several objectives as follows:

- To determine the composition and characteristics of MSW generated from vegetable market of Talaad Thai;
- To investigate the effects of feedstock to inoculum ratio, temperature, and alkalinity on the AD process through the biochemical methane potential technique;

- To investigate biogas production in lab scale BIOCEL reactor using optimum condition from BMP tests.

1.5 Scope of study

- The study focuses on utilization of the organic fraction of vegetable market waste from Talaad Thai market for biogas production;
- The study is implemented in two phases: the first phase is the BMP test, and the second phase is a lab scale reactor;
- The BMP test were conducted to find the optimum conditions such as F/I ratio (0.5 - 5.0 g VS/g VS), temperature, and alkalinity (0 - 1200 mg/g VS) to apply in the lab-scale reactor;
- Lab-scale BIOCEL reactors are operated at optimum conditions obtained from BMP test with two sources of inoculum UASB and digestate;
- Biogas and methane generation was analysis using GC Perkin Elmer (equipped with thermal conductivity detector). VFA was analysis using GC Perkin Elmer (equipped with flame ionization detector).

Chapter 2
Literature Review

2.1 Potential problems associated with municipal solid waste

As shown in Chapter 1, the annual waste generation increases in proportion to the rise in population and urbanization, and issues related to disposal have become challenging as more lands are needed for the disposal of increased solid wastes. As reported by United Nation (2004), most of the MSW in Southeast Asia countries is disposal by open dumping and landfill (Table 2.1). However, it has many problems associated with landfills, even with those that are clay-lined, includes high water table, groundwater contamination, and greenhouse gases migration. The problems with landfill disposal are summarized as: (1) Need for a site; (2) Risk of land and water pollution; (3) Noise pollution, visual pollution and the attraction of vermin; (4) Contribution to global warming; (5) Increased demand on natural resources and energy; (6) Economically expensive.

Table 2.1: State of MSW disposal methods in Southeast Asia

Country	Disposal method				
	Composting	Open dumping	Landfill	Incineration	Others
Indonesia	15	60	10	2	13
Malaysia	10	50	30	5	5
Myanmar	5	80	10	-	5
Philippines	10	75	10	-	5
Singapore	-	-	10	90	-
Thailand	10	65	5	5	15
Vietnam	10	70	-	-	20

Landfill problems can be solved by controlled biological decomposition of the rapidly biodegradable fraction through aerobic or anaerobic composting. These techniques are subject to pretreatment prior to landfill. As shown in Fig 2.1, various methods can be used to reduce the amount of waste to be disposed. Currently, mechanical pre-treatment, composting and anaerobic digestion plays the major role in pre-treatment of

MSW prior to landfill.

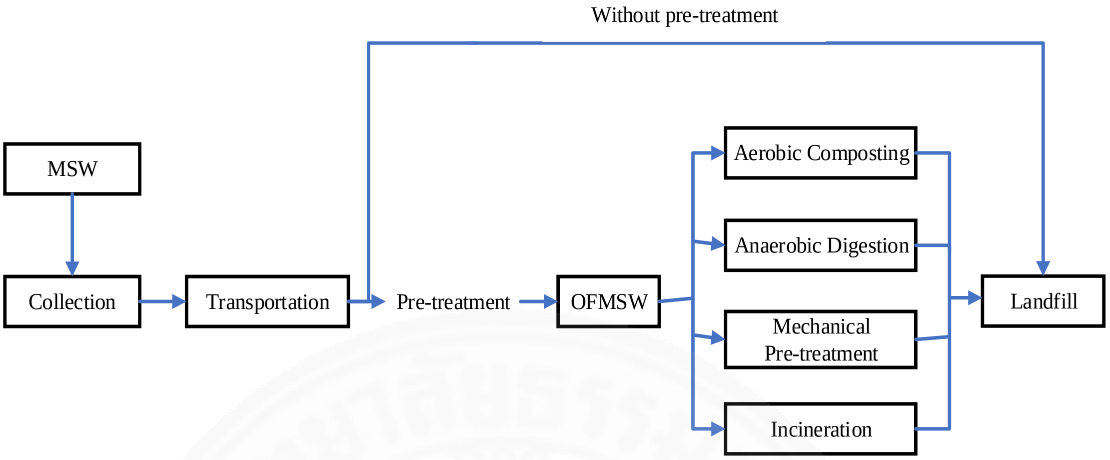


Figure 2.1: Pretreatment of organic fraction of MSW prior to landfill

2.1.1 Aerobic composting

Biological treatment of solid waste by simple composting is one of the applicable methods among the available technologies for pre-treating of MSW. Aerobic processes offer the advantage of relatively simple operation Aerobic composting needs proper aeration to provide sufficient oxygen for the aerobic microbes to stabilize the organic waste. Generally aerobic composting requires a significant area of land, 1.5 to 2 acres, for a plant with 50 ton/day capacity (Tchobanoglous, 1993). Odor problems in composting cannot be neglected. Recently, combining an anaerobic process with composting is getting acceptance as to make a positive energy balance by capturing methane from organic decomposition.

2.1.2 Anaerobic digestion

Anaerobic digestion is considered as an alternative option to manage and treat the organic fraction of municipal solid waste (OFMSW). This process not only treats the organic waste but also produces clean energy (biogas). The digestion residues (digestate) obtain from the process can be used as soil amendment or even nutrient-rich organic fertilizer depending on its final quality.

Anecdotal evidence indicated that biogas was used for heating bath water in Assyria during the l0th century BC and in Persia during the 16th century. In the 17th century, Van Helmont determined that flammable gases could evolve from decaying organic matter. In 1776, Volta reported that there was a relation between the amount of decaying organic matter and the amount of flammable gas produced. During 1804-1808, independent experiments by John Dalton and Humphrey Davy was resolved that the combustible gas is methane (Abbasi et al., 2012). However, it took until the end of the 19th century until anaerobic digestion was applied for the treatment of wastewater and solid waste. The first digestion plant was built at a leper colony in Bombay, India in 1859. AD reached England in 1895 when biogas was recovered from a "carefully designed" sewage treatment facility and used to fuel street lamps in Exeter (Nayono, 2010). The application of AD with the main purpose to reduce and stabilize solid waste gained its popularity after the large-scale introduction of activated sludge systems in the middle of the 20th century.

In general, AD technology of organic fraction of MSW as a pretreatment prior to landfill especially includes three main phases as a combined process: pretreatment, anaerobic digestion process and post-treatment.

Eriksson et al. (2005) used a calculation model, ORWARE (ORganic WAste REsearch), based on life cycle assessment of the material and energy flow with the various combination of waste treatment options. Based on the case study on three Swedish municipalities, it was revealed that composting was comparable to the anaerobic digestion but gave higher energy use and environmental impact. It was recommended that combination of anaerobic digestion; material recycling and incineration provide the best solution to reduce landfilling in terms of both environmental impact and cost. Edelmann et al. (2000) compared six different technologies to treat 1000 tons of biogenic waste per year using life cycle assessment tool. From an ecological point of view, anaerobic digestion with an aerobic post-treatment showed the best performance over composting, incineration or combination of digestion and composting. Lifecycle-based assessment of the major environmental impact of MSW has shown benefits from MSW energy recovery by reducing greenhouse emissions, reducing acid gas emissions, reduce depletion of natural resources (fossil fuel and materials) and reduced impact on water and land.

2.2 Principles of anaerobic digestion

2.2.1 Pretreatment of substrates

Pretreatment of substrates is considered as the first phase of the whole AD system. The main purpose of pretreatment is to alter or remove structural/compositional impediments to increase the yields of fermentable simple sugars/intended products in order to further process utilization. Varieties of physicochemical, mechanical, thermal and biological pretreatment technologies are being used for different organic substrates to improve their digestion. In which, the particle size is an important consideration when treating any substrates by anaerobic processes. According to Palmowski and Muller (2000), size reduction of the particles affects the overall bioconversion efficiency and the digestate quality. The pretreatment improves digester gas production and reduction of technical digestion time because this can improve the active surface area for faster enzymatic reactions.

2.2.2 Anaerobic digestion process

The AD is described as a series of processes involving microorganisms to break down complex organic material into simpler constituents in the absence of oxygen. Normally, 40% and 60% of the organic matter present in the feedstock is converted to biogas. The remaining waste consists of a residue with the appearance similar to peat which has some value as a soil conditioner and also, with some systems, the liquid residue has a potential to be used as liquid fertilizer. The overall result of AD is a nearly complete conversion of the biodegradable organic material into methane, carbon dioxide, hydrogen sulfide, ammonia and new bacterial biomass (Gerardi, 2003; Veeken et al., 2000). According to Gallert et al. (2005), and Gerardi (2003), a general formula describing the overall chemical reaction of the anaerobic fermentation process of organic material can be presented as follows:

$$\text{Organic matter} + H_2O + \text{nutrients} \xrightarrow{\text{Bacteria}} \text{new cells} + \text{resistant organic matter} + CO_2 + CH_4 + NH_3 + H_2S$$

AD process mostly involved the prokaryotic process, wherein the bacteria and archaea play an important role in waste decomposition. Generally, anaerobic digestion process consists of four stages with four different types of microorganisms: hydrolysis, acidogenesis, acetogenesis, and methanogenesis which are schematically illustrated in Figure 2.2 (Appels et al., 2008; Gerardi, 2003).

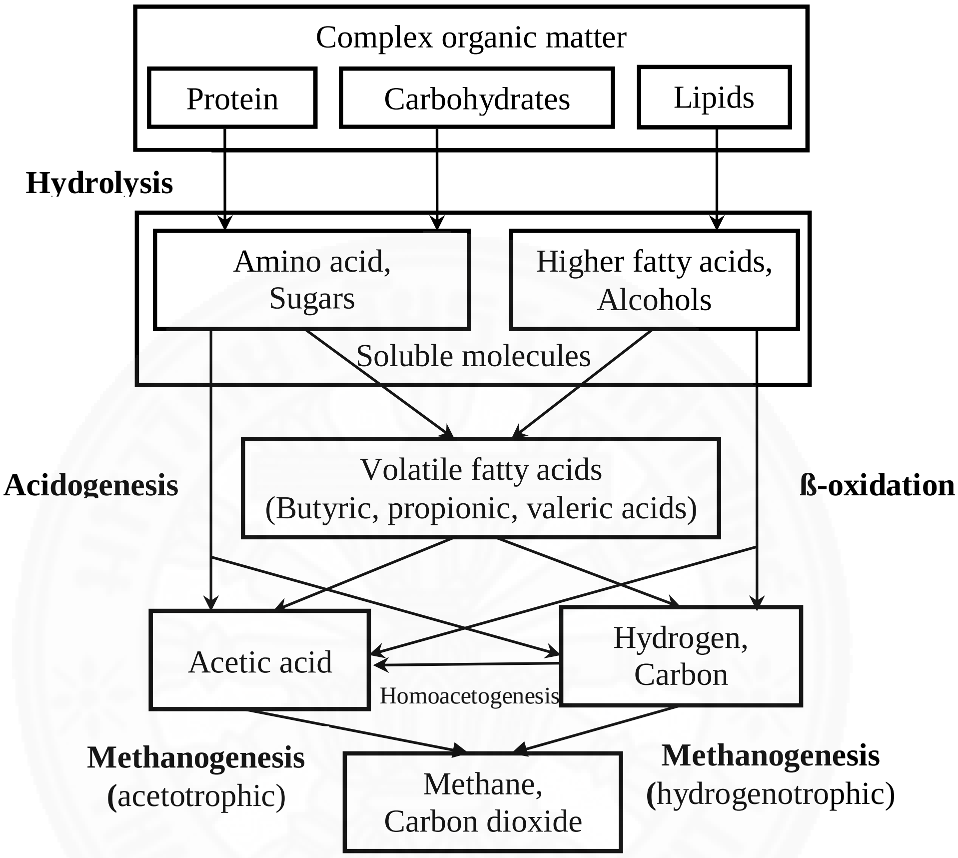


Figure 2.2: Anaerobic digestion biochemical conversion pathways

Hydrolysis

The first step of AD of solid waste is the depolymerization of complex organic polymers such as polysaccharides, proteins, and lipids (fat and grease) into simpler molecules. Hydrolysis reactions are executed by extracellular enzymes called hydrolases which catalyze reactions. In the chemical viewpoint, hydrolysis means the breakdown of long-chain biomolecules by the reaction with water. Thus, water is essential for the enhancement of process. Hydrolysis is a rather slow and energy consuming process and is normally considered as the overall rate limiting step for the complete anaerobic digestion of solid substrate (McCarty and Mosey, 1991; Gerardi, 2003; Pavlostathis and Giraldo-Gomez, 1991). In a nutshell, hydrolysis reaction in this stage converts (1) protein into amino acids, (2) carbohydrate into simple sugars, and (3) lipids into long-chain fatty acids. These simple products are organic monomers, which is further fermented, in the next stage of the process. The hydrolysis rate depends on substrates and bacterial concentrations, as well as on environmental factors such as pH and temperature.

Acidogenesis

The soluble organic components produced from the hydrolysis process are then degraded by a large variety of facultative anaerobic bacteria through many fermentative pathways. The degradation of these compounds results in the production of carbon dioxide, hydrogen gas, alcohols, organic acids, some organic nitrogen compounds, and some organic sulfur compounds. The most important of the organic acids is acetate since it can be used directly as a substrate by methanogenic bacteria (Pavlostathis and Giraldo-Gomez, 1991; Gerardi, 2003)

Acetate can be produced not only from the acidogenic of soluble organic compounds, but also through acidogenesis by degradation of intermediary products - volatile fatty acids (VFAs). In the process, VFAs are converted into acetate, hydrogen gas, and carbon dioxide. This conversion proceeds with the action of obligate hydrogen producing acetogenic bacteria, which are considered as acetogens and some are part of acidogens under syntropy phenomena (Gerardi, 2003; Pavlostathis and Giraldo-Gomez, 1991).

Methanogenesis

Methanogenesis as the final stage of AD, generates methane gas by methane producing bacteria. Methane is formed around 66% from acetate by means acetate decarboxylation proceeded by acetolactic methanogenic bacteria and 34% from carbon dioxide reduction by hydrogen, catalyzed by hydrogen utilizing (hydrocephalic) methanogenic bacteria (Gerardi, 2003; Pavlostathis and Giraldo-Gomez, 1991). The pathways along with the stoichiometry of the overall chemical reactions are:

Acetotrophic methanogenesis: $4CH_3COOH \longrightarrow 4CO_2 + 4CH_4$

Hydrogenotrophic methanogenesis: $CO_2 + 4H_2 \longrightarrow CH_2 + 2H_2O$

2.2.3 Post-treatment

When the AD process is completed, the digestate or biodegradable solid residues is subjected to post-treatment. Such treatment contains dewatering, aeration, and leachate treatment. The purpose of aeration as post-treatment is to remove remaining organics, through aerating the compounds are reduced and produce valuable products such as fertilizer and soil conditioner. Also, the possibility of terminating and optimizing the anaerobic fermentation at the organic-acid stage as part of the pre-stage of anaerobic digestion has been seriously considered. According to Mata-Alvarez and Sans (1995), the organic acids produced in the leachate could be recovered and process further. These acids could be used to produce methyl or ethyl esters wherein considering their elevated octane numbers (between 103 and 118) could be advantageously used as an additive for gasoline. Moreover, the VFA would be extracted from the liquid phase and would be converted to products such as methyl or ethyl esters, for commercial purposes (D'Addario et al., 1993). Moreover, the amount and quality of these products depend entirely upon the quality of the MSW feedstock, the method of digestion, and the extent of any post-treatment refining processes.

2.3 Factors affecting AD process for biogas production

In AD process, the production of methane is influenced by a number of parameters, such as temperature, type of feedstock, pH level, retention time, and C/N ratio, etc. The maximum production takes place when these parameters are chosen in the optimum range. The optimized range of some of these parameters is discussed in this section.

2.3.1 Temperature

Temperature is one of the most significant parameters influencing AD, because it not only affects the activity of enzymes and co-enzymes but also affects the methane yield and digestate (effluent) quality (Gerardi, 2003; Karthikeyan and Visvanathan, 2013). Lower temperatures during the process are reported to decrease microbial growth, substrate utilization rates, and biogas production (Kim et al., 2006; Trzcinski and Stuckey, 2010). Moreover, lower temperatures may also result in an exhaustion of cell energy, a leakage of intracellular substances or complete lysis (Kashyap et al., 2003). In contrast, high temperatures lower biogas yield due to the production of volatile gases such as ammonia which suppresses methanogenic activities (Fezzani and Cheikh, 2010).

Generally, anaerobic bacteria can grow at psychrophilic (10–30°C), mesophilic (30–40°C) and thermophilic (50–60°C) conditions (Karthikeyan and Visvanathan, 2013; Zhang et al., 2014; Mao et al., 2015; Gerardi, 2003). Thermophilic AD has a rate-advantage over mesophilic digestion as a result of its faster reaction rates and higher-load bearing capacity and, consequently, exhibits higher productivity compared with mesophilic AD. Thermophilic temperatures convert organic acids at a faster rate, with higher CH_4 production (25–35%) than the mesophilic system (Karthikeyan and Visvanathan, 2013). However, acidification may occur during thermophilic AD, inhibiting biogas production. Other disadvantages of thermophilic condition such as decreased stability, low-quality effluent, increased toxicity and susceptibility to environmental conditions, larger investments, poor methanogenis and higher net energy input have also been identified. In addition, this process is more sensitive to environmental changes than the mesophilic process (Mao et al., 2015). While mesophilic condition is reported better process stability and higher richness in bacteria. The operation in the mesophilic range is more stable and requires a smaller energy expense (Ward et al., 2008; Fernández et al., 2008). Castillo et al. (2006) found that the best operational temperature was 35°C with retention time in 18 days while a little fluctuation in temperature from 35°C to 30°C caused a reduction in the rate of biogas production (Chae et al., 2008).

Ambient/seasonal temperature AD has also been used to treat organic waste. This process does not require an extra heat supply but exhibits lower methane production and lower stability than the mesophilic process due to temperature changes in the surrounding environment. Hyperthermophilic AD exhibits greater resilience in treating co-substrates containing high concentrations of proteins, lipids, and nonbiodegradable solid matter (Mao et al., 2015).

2.3.2 pH

The ideal pH range for AD is quite narrow pH 6.8–7.2. The growth rate of methanogens is greatly reduced below pH 6.6 (Mosey and Fernandes, 1989) since an excessively alkaline pH can lead to the disintegration of microbial granules and subsequent failure of the process (Sandberg and Ahring, 1992). The optimal pH of methanogenesis is around pH 7.0 but the optimum pH of hydrolysis and acidogenesis has been reported as being between pH 5.5 and 6.5 (Kim et al., 2003a; Kim et al., 2003b)

Buffer capacity is often referred to as alkalinity in AD, which is the equilibrium of carbon dioxide and bicarbonate ions that provides resistance to significant and rapid changes in pH, and the buffering capacity is therefore proportional to the concentration of bicarbonate. Buffer capacity is a more reliable method of measuring digester imbalance than direct measurements of pH, as an accumulation of short chain fatty acids reduce the buffering capacity significantly before the pH decreases. Increasing a low buffer capacity is best accomplished by reducing the organic loading rate, although a more rapid approach is the addition of strong bases or carbonate salts to remove carbon dioxide from the gas space and convert it to bicarbonate, or alternatively bicarbonate can be added directly (Guwy et al., 1997). Direct bicarbonate addition is more accurate as converting carbon dioxide to bicarbonate requires a time lag for gas equilibrium to occur which could result in over-dosing. It has also been demonstrated that the inoculum-to-feed ratio can be modified to maintain a constant pH (Gunaseelan, 1995).

2.3.3 Feedstock/inoculum

The feedstock to inoculum ratio (F/I) is an important parameter affecting AD processes as well as in the assessment of anaerobic biodegradability of solid wastes (Neves et al., 2004). Theoretically, the F/I ratio has an effect only on the kinetics, and not on the ultimate methane yield, which only depends on the organic matter content (Gunaseelan, 1995; Raposo et al., 2006) but it is reported that too high F/I may be toxic while too low F/I may prevent induction of the enzyme necessary for biodegradation (Prashanth et al., 2006). Each substrate has its optimum F/I ratio, considering the potential amount of volatile fatty acids (VFAs) produced and its capacity to buffer the medium due to the ammonium produced by the hydrolysis of proteins (Lesteur et al., 2010).

According to Neves et al. (2004), a F/I ratio ranging between 0.5 and 2.3 g VS/g VS can prevent acidification phenomena. In the study, using granular inoculum prevented acidification during the anaerobic batch biodegradation of a kitchen waste for waste/inoculum ratios in the range of 0.5–2.3 g VS/g VS when the alkalinity/COD ratio was 37 mg $NaHCO_3$/g COD. While suspended sludge gave a significantly lower activity with similar condition, the methane production rates and the biodegradability were significantly lower and the pH decreased below 5.5 at the waste/inoculum ratio of 2.3 g VS/g VS. When the added alkalinity was decreased to 2 mg $NaHCO_3$/g COD, the ratio waste/inoculum was clearly more important than the inoculum activity, since, irrespective of the sludge used, acidification occurred at waste/inoculum ratios higher than 0.5 g VS/g VS. For a waste/inoculum ratio of 1.35, there were no significant differences between the results obtained for the biodegradability and maximum methane production rate, when the alkalinity decreased from 44 to 22 mg $NaHCO_3$/g COD.

While Owen et al. (1979) reported that the F/I proposed as a standard was approximately 1 g VS /g VS. Instability in the anaerobic process, such as high COD content in the effluent and VFAs accumulation, occurs with F/I ratio lower than 0.5 (Liu et al., 2009). Hashimoto (1989) and Labatut et al. (2011) reported that the minimum F/I ratio of 2 g VS substrate/g VS inoculum was required when digesting wheat straw at concentrations of 10–40 g VS/L and dairy manure at concentrations of $\geq$3 g VS/L, respectively. However, in the case of more recalcitrant wastes (woody feed stocks and municipal wastes), the rate of methane production in BMP assays was optimum at S/I of 0.5 g VS substrate/g VS inoculum (Chynoweth et al., 1993).

2.3.4 VFAs

Volatile fatty acids are a key intermediate in the AD process and decide the pH and stability of the AD reactor. According to Siegert and Banks (2005), pH of AD process decreases particularly the production of fatty acids, and when total VFAs concentration is above 4 g/L, fermentation of glucose is inhibited. Acetic acid is usually present in higher concentrations than other fatty acids during anaerobic digestion (Wang et al., 1999), but propionic and butyric acids are more inhibitory to the methanogens. Propionic acid concentrations over 3000 mg/L have previously been shown to cause digester failure (Boone and Xun, 1987), but in a recent study of Pullammanappallil et al. (2001) found that propionic acid was an effect rather than a cause of inhibition of anaerobic processes. Monitoring of fatty acids helps to increase the process stability (Ahring et al., 1995), as an increase in fatty acids can be indicative of an overload of the organic loading rate. The methanogens are not able to metabolise the acetate produced by the acetogenic organisms until the number of methanogenic organisms has increased sufficiently. This is true when feedstocks are rapidly hydrolysed such as vegetable and fruit wastes.

2.3.5 Alkalinity

Alkalinity or buffer capacity in anaerobic digestion is the equilibrium of carbon dioxide in the biogas and bicarbonate ions that provides resistance to significant and rapid changes in pH. The equilibrium between carbonic acid, bicarbonate alkalinity is a function of pH in AD process. Bicarbonate alkalinity is the primary source of carbon for methane-forming bacteria. AD process stability is enhanced by a high buffer capacity. A decrease in alkalinity below the normal operating level has been used as an indicator of pending failure. The reduction of alkalinity in AD can be caused by (1) an accumulation of organic acids due to the failure of methanogens to convert the organic acids to methane, (2) a feedstock that discharge of organic acids to the anaerobic digester, and (3) the presence of wastes that inhibit the activity of methane-forming bacteria (Ward et al., 2008). According to Guwy et al. (1997) buffer capacity can be increased by reducing the organic loading rate, however, a more rapid approach is the addition of strong bases or carbonate salts to remove carbon dioxide from the gas space and convert it to bicarbonate, or alternative bicarbonate can be added directly. It was discovered that many carbohydrate-rich feedstocks were found to require either co-digestion with other feedstocks or addition of alkaline buffer to ensure stable performance (Hills and Roberts, 1982; Knol et al., 1978). An alkalinity level ranging from 1000 to 5000 mg $CaCO_3$/L was recommended by Tchobanoglous et al. (1991).

As theory, the VFAs/TA should be maintained below 0.4 to keep stability for the AD process (USEPA, 1976). While Callaghana et al (2002) concluded that as the ratio of VFAs/TA is higher than 0.8 the AD digester is significant instability.

2.3.6 C/N ratio

The C/N ratio presents the relationship between an amount of carbon and nitrogen in organic materials. This ratio in feedstocks plays a pivotal role in anaerobic digestion. The unbalanced nutrients are considered as a factor limiting anaerobic digestion of organic wastes (Khalid et al., 2011). In case of a high C/N ratio, the methanogens consume nitrogen rapidly, which results in lower gas yield. Otherwise, a lower C/N ratio causes ammonia accumulation and pH values exceeding 8.5, which is toxic to methanogenic bacteria (Kothari et al., 2014). Optimum C/N ratio of the materials can be maintained by mixing materials of high and low C/N ratio; e.g. organic solid waste can be mixed with sewage or animal manure. Generally, a C/N ratio range of 20–30 was considered to be the optimum condition for AD (Zhang et al., 2014; Mao et al., 2015; Khalid et al., 2011; Ward et al., 2008; Kothari et al., 2014). However, optimal C/N ratio is a function of the type of feedstock and varies with types of feedstock.

For AD of fruit and vegetable waste, C/N ratio ranging from 22 to 25 is the most suitable (Bouallagui et al., 2009), whereas Romano et al. (2008) suggested that the optimal C/N ratio of onion juice and digested sludge should be maintained at 15. On the other hand, Zhu et al. (2014) studied AD of organic wastes and found C/N ratio between 15 and 18, when corn stover was inoculated with digested sewage sludge.

2.4 Biomethane potential assay of organic solid wastes

Before starting a full-scale AD reactor, biochemical methane potential (BMP) assays should be investigated. BMP assays analyze which types of substrates, have the highest biomethane potential among various substrates. The assays also determine the optimum ratios between substrates and inoculums; and evaluate the anaerobic biodegradation rate of substrates or retention time required for complete the digestion (Elbeshbishy et al., 2012). For the last 20 years, although various BMP methods have been formulated, their procedures are very similar, the only difference between BMP assays is the biomethane production from the inoculum. There are two methods to determine the biomethane production of the inoculum: the blank assay (Neves et al., 2004; Gunaseelan, 2004; Elbeshbishy et al., 2012) and pre-incubated inoculum (VDI, 2006; Prabhudessai et al., 2013; Elbeshbishy et al., 2012). In the blank method, the biomethane production from the inoculum (i.e. with water or medium only) is deducted from the biomethane production achieved in the assays with the substrate (Angelidaki and Sanders, 2004; Elbeshbishy et al., 2012; Gunaseelan, 2004; Labatut et al., 2011; Owen et al., 1979; Prabhudessai et al., 2013). For the pre-incubated inoculum method, the biomethane gas, initially presented in the inoculum, must be first depleted. Then it is used for the assays, which the process is maintained until no significant biomethane production is detected. (VDI, 2006; Elbeshbishy et al., 2012).

BMP assay values are sensitive to several parameters such as temperature, pH, solid waste particles size, and feedstock to inoculum ratios. Temperature has a direct effect on survival and growth of bacteria and their metabolic activities (Lesteur et al., 2010). Theoretically, on the AD process, the temperature only affects the digestion rates and not on the bio-digestion of a substrate. However, in very-low-temperature range (<20°C), the degradation rates can be decreased significantly and this lowers the biogas achievement than that of optimum temperature (Angelidaki and Sanders, 2004). The amount of inoculum added or feedstock to inoculum ratio (F/I) also plays a significant role in the BMP assays. The F/I ratio only influences theoretically on the kinetics, and not the biomethane yield, which only depends on the organic component (Elbeshbishy et al., 2012). However, the assays may be toxic when the F/I ratio is too high while too low F/I may inhibit the production of the necessary enzyme for the AD. This ratio also affects the lag phase, which is shorter for low ratios (Chen and Hashimoto, 1996). Each feedstock has the optimum F/I ratio, depending on the amount of VFAs produced, and the capability to buffer the medium of the ammonium produced from the proteins hydrolysis process. A small amount of inoculum is expected due to the internal biogas production can affect the results. But when the F/I ratios increase, it can lead to overloads due to the high VFA concentration. According to Hashimoto (1989) and Labatut et al. (2011), the F/I ratio should be higher than 2 g VS feedstock/g VS inoculum as digesting wheat straw and cow manure at concentrations of 10 – 40 g VS/L and higher than 3 g VS/L, respectively. For recalcitrant wastes (woody substrates and municipal wastes), the optimum biomethane production rate in the BMP assays was achieved at the F/I ratio of 0.5 g VS feedstock/g VS inoculum (Chynoweth et al., 1993). For kitchen waste, Neves et al. (2004) concluded that when the buffer capacity is lower than 2 mg $NaHCO_3$/g COD, the F/I ratio was more important than the inoculum activity; the acidification occurred and limited the methanization rate with F/I ratios higher than 0.5 g VS/g VS. With maize waste, four different F/I ratios of 0.3, 0.5, 0.6, 1 g VS/g VS showed the optimum methane production rate, the high initial feedstock load gave higher maximum specific biomethane production rate but a lower maximum methane production per load (Raposo et al., 2006). Table 2.2 shows previous BMP experiments on different operating conditions.

Table 2.2: Research BMP experiments on different operating conditions

Substrates	**Inoculum**	**F/I ratio**	**Temperature** (ºC)	**RT** (days)	**Biogas yield** (mL/g VS)	**% CH_4**	**VS reduction**	**Reference**
Coconut oil cake	Cow dung	VS loading 4 g VS/L	35	60	451 662	55	NR	(Prabhudessai et al., 2013)
Grass		4.5 g VS/L		25	501 557	51		
Food waste (gCOD/gVS)	AD sludge from WWTP	0.25	37	30	1000 (CH_4)	NR	NR	(Elbeshbishy et al., 2012)
		0.5			940 (CH_4)			
		1			1400 (CH_4)			
	AD sludge from SWT plant	0.25			440 (CH_4)			
		0.5			790 (CH_4)			
		1			440 (CH_4)			
Switchgrass	AD sludge from SWT plant	1	35	40		NR	NR	(Labatut et al., 2011)
Cabbage					257			
Potatoes					335			
Corn silage					296			
Food waste		3.1	35	25	430	57	51	

Substrates	Inoculum	F/I ratio	Temperature (ºC)	RT (days)	Biogas yield (mL/g VS)	% CH_4	VS reduction	Reference
(gVS/ gVS)	AD sludge from WWTP	1.6	50		778	66	94	(Liu et al., 2009)
		3.1	50		742	68	82	
		4.0	50		784	66	80	
Green waste		3.1	35		372	56	52	
		1.6	50		631	57	92	
		3.1	50		529	55	81	
		4.0	50		524	55	78	
Mixture of Food and Green waste (1:1 g VS)		3.1	35		358	52	48	
		1.6	50		716	60	91	
		3.1	50		613	59	83	
		4.0	50		671	58	81	

2.5 Categories of MSW AD technologies

Anaerobic digestion processes can be termed as “wet” and “dry” digestions depending on the total solids concentration of the feed substrate. Anaerobic digestion is defined as a wet process if the total solids concentration of the substrate is less than 15% and as a dry process if the concentration reaches 20 – 40%.

According to Vandevivere et al. (2002), typically anaerobic digesters or processes of solid waste can be categorized into several types, mostly according to the feeding mode (continuous mode: single stage, two stages and batch mode) and the moisture content of the substrate (wet or dry digestion). Furthermore, with those basic types, the anaerobic reactors can be arranged according to the digestion process temperature (mesophilic or thermophilic), mixing methods (gas injection or mechanical stirrers), and the shape of the reactors (vertical or horizontal). Stimulated by the increasing demand of anaerobic digester for organic solid wastes, several commercial anaerobic digester plant designs have been developed over the past two decades. Figure 2.3 presents the available anaerobic digestion technology for solid waste treatment.

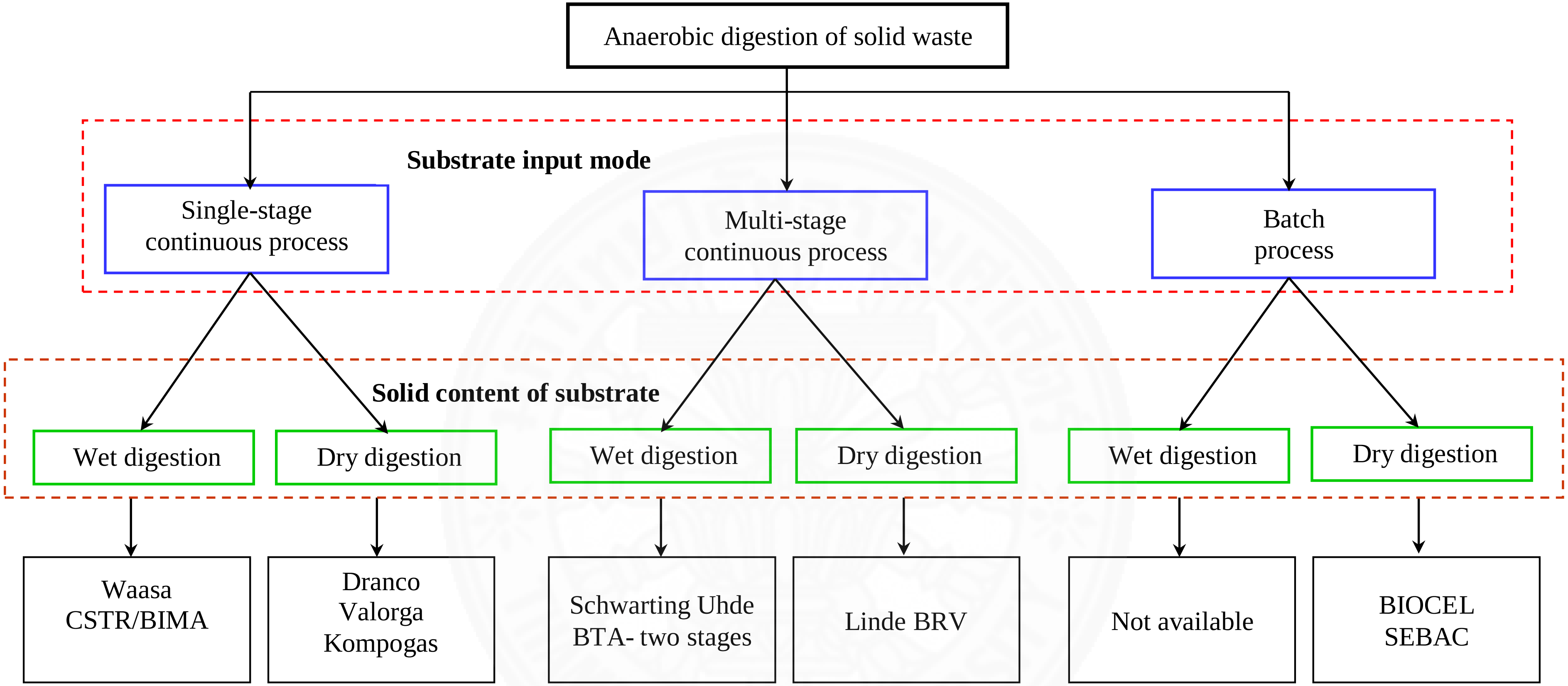


Figure 2.3: Various anaerobic digestion technologies available in current market

2.6 Dry Anaerobic Composting (DRANCO) system

2.6.1 DRANCO's structure design

The DRANCO system was developed in the late 1980s in Gent, Belgium. The system employs a one-stage anaerobic digestion process, which is followed by a short aerobic maturation phase. The total solids content of the digester depends on the waste material source but is in the range 15-40%. Although mostly operated at the thermophilic temperature (50-55°C), mesophilic operation (35-40°C) can also be applied to specific waste streams. The DRANCO system is typically a vertical plug-flow reactor. The digester is fed from the top of the reactor and the digested slurry is removed from the bottom at the same time. Usually, one part of the digested slurry is used as inoculum and mixed with six to eight parts of a fresh substrate. There are no mixing devices needed in the reactor other than the natural downward movement of the waste caused by fresh feeding and digestate withdrawal. The rest of the digested slurry contains active bacteria, some ammonia, and undigested solids and must be aerobically stabilized for use as agricultural compost.

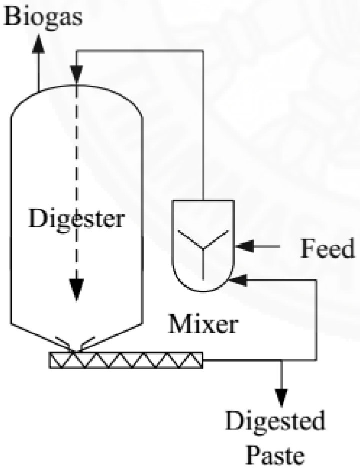


Figure 2.4: Schematic diagram of a DRANCO reactor

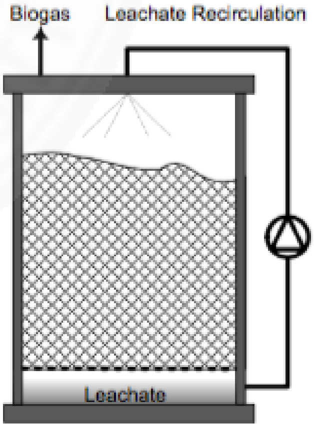


Figure 2.5: Schematic diagram of a BIOCEL reactor

2.6.2 DRANCO performance

For DRANCO system, food waste, fruits and vegetable waste, organic fraction MSW (OF-MSW) are effectively used as sole source of DRANCO system (Brummeler et al., 1991; Di Maria et al., 2012; Forster-Carneiro et al., 2008; Di Maria et al., 2013). Some of the studies conducted on DRANCO process are summarized in Table 3.2. With food waste feedstock, Banks et al. (2008) reported that with OLR varied from 3.5 – 4.5 kg VS m^{-3} d^{-1}, the mean methane yield achieved 390 L $kgVS^{-1}$ in mesophilic temperature. While with a little higher OLR range from 3.7 – 5.0 kg VS m^{-3} d^{-1}, the mean methane yield reached to 410 L $kgVS^{-1}$ in thermophilic temperature. Moreover, the study also pointed out that the mean VS reduction in thermophilic temperature is a little higher than mesophilic temperature. Considering OFMSW feedstock, DeBaere et al. (2008; DeBaere and Verstraete, 1984) also showed that the performance in term of methane yield of DRANCO reactors in thermophilic temperature is higher than mesophilic temperature. With the OLR range from 10.0 to 13.2 kg VS m^{-3} d^{-1}, the methane yield achieved from 260 to 235 L $kgVS^{-1}$ in mesophilic condition. While the thermophilic condition could be loaded with higher OLR range from 10.0 to 14.9 kg VS m^{-3} d^{-1}, and the methane yield reached to 310 L $kgVS^{-1}$.

2.7 Dry anaerobic batch digestion (BIOCEL) system

2.7.1 BIOCEL's structure design

The BIOCEL system was developed in the 1980s and 1990s in Holland at the Wageningen University as a part of the early research on high-solids digestion of MSW. The initial goal of the process was to reduce cost by simplifying material handling and eliminating the need for mixing while simultaneously achieving relatively high loading and conversion rates.

The BIOCEL system (Fig. 2.4) is based on a batch-wise dry anaerobic digestion. Digesters in the process are filled with wastes mixed with an inoculation material, usually taken from the previous run. Once the digesters are filled, they are closed with airtight doors and the reaction is allowed to go to completion. The reactors using this system have chambers under them that allow the collection of leachate. Temperature is controlled at 35-40^{o}C by spraying the leachate, which is preheated by a heat exchanger, from nozzles on top of the digesters. Typical retention time in this process is reported to be 15 – 21 days. This system is similar to a landfill cell, but it controls the leachate recirculation and is conducted at a higher temperature than in a landfill.

2.7.2 BIOCEL performance

Various research studies with different feedstock have been conducted using the BIOCEL process (Le Thi et al., 2013; Brummeler et al., 1991; Di Maria et al., 2013), and some of these studies are summarized in Table 2.3. Considering organic fraction of MSW (OFMSW) feedstock, Kim et al. (2013) reported that the mixture among OFMSW, digested OFMSW (DOFMSW) and pig manure gave the highest cumulative biogas yield and specific methane production compared with another mixture. The performance of the lab scale (45 L) showed a volatile solid (VS) reduction of 61% and specific methane production of 362 L kg VS-1, for a retention time of 30 days. While the pilot scale gave a VS reduction of 65% and specific methane production of 367 L kg VS-1, for 25 days. According to Forster et al. (2008), VS reduction for source selected organic fraction MSW (SSOFMSW) and mechanically selected organic fraction of MSW (MSOFMSW) was 45 and 56%, respectively. The cumulative methane yields per kg VS were not reported, however, cumulative biogas of 120 L (25 L CH_4) was achieved for SSOFMSW and 82 L (30 L CH_4) for MSOFMSW. While Brummeler et al. (1991) reported that mixture between OFMSW and DOFMSW gave the mean methane yield of 221 L/kg VS-1 for 36 days. Studies of Di Maria et al. (2013), with OFMSW and DOFMSW achieved the mean methane 212 L/kg VS for 74 days, however, the specific methane production achieved the high values from day 15th to 40th. In another study, the authors (Di Maria et al., 2012) reported that OFMSW mix with DOFMSW ratio 0.711 and 0.291 (OFMSW/Inoculum) gave the methane production of 180 L/kg VS, and 400 L/kg VS for 40 and 55 days, respectively.

Table 2.3: Experimental conditions and methane generation in BIOCEL and DRANCO system

Feedstock	**Feed TS (%)**	**Scale**	**Temp. (^{0}C)**	**RT (days)**	**OLR (kg VS m^{-3} d^{-1})**	**CH_4 yield (mL CH_4 g VS^{-1})**	**CH_4 (%)**	**(%) VS Reduction**	**Ref.**
				BIOCEL system					
OFMSW;	23	Lab 45 L	Ambient (20-38)	32	2.8	146	48	35	(Le Thi et al., 2013)
OFMSW+DOFMSW;	23	Lab 45 L		32	2.6	214	53	49	
	18	Pilot 5 m^3		25	2.7	244	60	51	
OFMSW+PM;	15	Lab 45 L		27	2.0	317	52	47	
	20	Pilot 5 m^3		25	3.3	351	51	54	
OFMSW+DOFMSW+PM	19	Lab 45 L		30	2.08	362	59	61	
	22	Pilot 5 m^3		25	3.59	367	62	65	
SSOFMSW+DS	21	Lab 5 L	55	60	NR	NR	21	45	(Brummeler et al., 1991)
MSOFMSW+DS	19						35	56	
OFMSW + DOFMSW	31	Lab 78 L	35	30	3.1	221	60	NR	(Di Maria et al., 2012)

Feedstock	Feed TS (%)	Scale	Temp. (^{0}C)	RT (days)	OLR (kg VS m^{-3} d^{-1})	CH$_4$ yield (mL CH$_4$ g VS^{-1})	CH$_4$ (%)	(%) VS Reduction	Ref.
OFMSW+ DOFMSW	33	Pilot 100 L	35	74	NR	212	NR	NR	(Forster-Carneiro et al., 2008)
OFMSW+ DOFMSW	40	Pilot 100 L	35	40	7.95	180	50	NR	(Di Maria et al., 2013)
	39			55	5.62	400	50		
DRANCO system									
Food waste + DFW	23	Pilot 1.5 m^3	56	32	3.7 – 5.0	410	58	70	(Banks et al., 2008)
			35.6	27	3.5 – 4.5	390	58	67	
MSOFMW	30	Lab 35 L	35 - 40	16 – 21	10.0	260	NR	NR	(De Baere, 1984)
	35				12.1	264			
					13.2	235			
			50 -55		10.0	286			
					12.1	282			
					13.2	283			
					14.9	310			
MSOFMW	25	Pilot	35	14	15	187	NR	NR	(DeBaere

Feedstock	Feed TS (%)	Scale	Temp. (^0C)	RT (days)	OLR (kg VS m^{-3} d^{-1})	CH_4 yield (mL CH_4 g VS^{-1})	CH_4 (%)	(%) VS Reduction	Ref.
	35	60 m^3	40	21					and Verstraete, 1984)
Vegetable + fruit + garden waste	43	Pilot 56 m^3	50	20	12	143	55	NR	(Six and De Baere, 1992)

OFMSW=organic fraction of MSW, DOFMSW=digested organic fraction of MSW, PM=pig manure, MSOFMSW=mechanically selected organic fraction of MSW, SSOFMSW=source sorted organic fraction of MSW, OLR=organic loading rate, RT=retention time, TS=total solid, NR=not reported, DS= digested sludge, FVW= fruit and vegetable waste, WMS= waste-mixed sludge

Chapter 3

Methodology

The framework of the research consists of two phases as presented in Figure 3.1. At first, composition and characteristics of solid waste generated from the vegetable market of Talaad Thai were investigated. Phase I of the research determined biochemical methane potential of the waste. In phase II, lab-scale reactors were conducted in AD batch process.

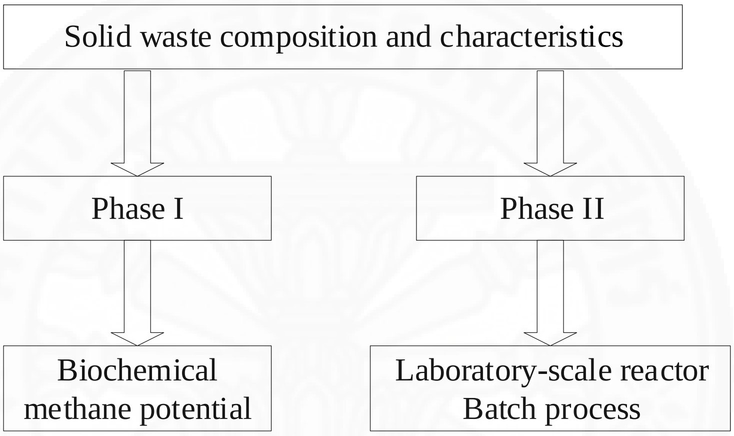


Figure 3.1: Overall the study

3.1 Organic solid waste and anaerobic sludge inoculum

3.1.1 Organic solid waste

The mixed vegetable wastes from the vegetable market in Talaad Thai, located in Thailand, was used as the feedstock. The feedstock was cut and ground in a blender to give a fraction with particle size of around 2 – 3 mm.

3.1.2 Anaerobic sludge (Inoculum)

The granular sludge from the up-flow sludge blanket reactor (UASB), Pathum Thani Brewery Co., Ltd. Thailand was used as the inoculum. After collecting organic waste and inoculum from the field, the feedstocks were determined for Total Solid (TS), Volatile Solid (VS), pH, alkalinity, total carbon and total nitrogen.

3.2 Solid waste composition and characteristics

3.2.1 Procedure for waste composition analysis

Waste composition analysis was performed as the method of ASTM D5231-92 (2008) and manual sorting of waste samples with standard protocols, followed by lab analyses of the representative samples for its physical and chemical parameters. These analyses are important to estimate the composition of the waste and are suitability for the anaerobic digestion process.

The determination of the mean composition of MSW based on the collection and manual sorting of a number of samples of waste was conducted in one week (7 days). The weight fraction of each component in the sorting sample was calculated from the weights of the components. The mean waste composition was calculated by using the results of the composition of each of the sorted samples. Garbage bins for sampling were selected randomly each day of the week during sampling period, so as to get representative of the waste stream. According to ASTM D5231-92 (2008), for a weekly sampling method of k days, the number of samples each day shall be approximately n/k, where n is the total number of sample to be selected for the determination of waste composition. A weekly period for this study is 7 days. The number of sorting samples (n) required to achieve the desired level of measurement precision is a function of the component (s) under consideration and the confidence level. The governing equation for n is as follows:

$$n = \left(\frac{t^{*}s}{e\bar{x}}\right)^{2} \qquad (1)$$

where:

t* = is the student t statistic corresponding to the desired level of confidence,
s = the estimated standard deviation,
e = desired level of precision,
$\bar{x}$ = estimated mean.

For the study, waste was the governing equation component at 90% confidence level, with 10% precision, it means

s = 0.030; e = 0.100; $\bar{x} = 0.100$; $t^{*}(\infty) = 1.645$

Using (Eq.1):

$$n = \left(\frac{1.645(0.030)}{0.100(0.100)}\right)^2 = 24 = n_o$$

For n = 24, t^*_{90} = 1.714, so n = 26 = n'.

Since 26 (that is, n') is within 10% of 24 (that is, n_0), 26 samples should be selected for analysis. Hence n = 26, where n is the total number of sample to be selected for the determination of waste composition. The per day samples is calculated by using a formula n/k, where n is the actual samples (26 in this case), and k is the number of days in a week (7 in this case). Thus, the average samples per day was 4, one sample was calculated as one available garbage bin at Talaad Thai market.

3.2.2 Tools and equipment for sorting

Table 3.1 shows the tools and equipment using for waste sorting. The Personal Protective Equipment (PPE) like gloves and face mask were used for safety during waste sampling and sorting at the market.

Table 3.1: Tools and equipment for Waste Sorting

Tool and equipment	Quantity
Canvas	1
Bucket 45 L	1
Scale 50 kg	1
Gloves	2
Face mask	1 (pack)
Plastic bag	2 (packs)
Zip lock bag	1 (packs)

3.2.3 Sorting procedure

As described in section 3.2.1, the average sample per day is 4 for 7 days. The sorting place was at the storage area of the vegetable market, Talaad Thai market. The garbage bin is selected randomly from the workers directing to the sorting place. The collected samples were poured on the prepared canvas for sorting. Manual sorting and visual characterization of the samples were used to classify MSW sample into 5 categories as follows: organic waste, wood, metal, plastic, paper, and glass. Each category of waste was weighed by a balance in kilogram unit. The percentage of each component was determined for each day. Eventually, an average percentage of the waste composition was calculated from the sum of the percentage of each component in every day divided by the total days of sampling.

3.2.4 Waste characteristics analysis

After determining the composition, each day of 7 sampling days, 1 kg of waste generated from the vegetable market of Talaad Thai was taken to analyze characteristics at SIIT laboratory. TS, VS, pH, total carbon, and total nitrogen of the waste were measured. Analysis methods and standards of the parameters is presented are table 3.4.

3.3 Biochemical methane potential (BMP) experiment (Phase I)

Bio-methane potential (BMP) test, which is considered the most suitable method for a relatively easy evaluation of the anaerobic digestibility (Labatut et al., 2011; Chynoweth et al., 1993). The tests are widely used in studies concerning the anaerobic digestion of organic solids. The assay is used to determine the methane yield from the organic material by using an anaerobic batch reactor. Although they are often criticized to be the time consuming, with an average length longer than 30 days, such tests are doubtless easy to be conducted, relatively inexpensive and repeatable. Moreover, BMP tests give significant information about the bio-methanation of specific substrates and provide experimental results essential to calibrate and validate mathematical models. The tests measure the maximum amount of biogas or bio-methane produced per gram of volatile solids (VS) contained in the organics used as substrates in the anaerobic digestion process. In this study, the BMP test was conducted with vegetable waste from Talaad Thai market co-digested with anaerobic sludge from Pathumthani Brewery Company Limited, Thailand.

3.3.1 Materials and equipment needed to conduct BMP test

Equipment and materials used for BMP test are presented in Table 3.2.

Table 3.2: Equipment for BMP test

Tool and equipment	Note
Blender	To reduce waste particle size into fine solids as possible
250 ml glass bottle	Glass bottles with thick rubber septum to be used as reactors. The exact volume of each bottle was determined by filling the bottles with measured volume of water;
Incubator	
1 ml glass syringe with pressure lock	To allow sampling of a fixed volume at actual pressure from the reactors
Gas chromatograph	To determine composition of biogas: CO_2, CH_4, H_2.
N_2 gas	-
Buffer solution	Sodium bicarbonate

3.3.2 BMP test procedure

BMP test was conducted to investigate the possible methane generation potential of solid waste generated from the vegetable market of Talaad Thai. This test was conducted to find optimum conditions to be used for lab-scale reactors, and also to evaluate the performance in term of biogas and methane generation.

The procedure of BMP followed was based on the principles described by Owen et al. (1979), and revised by others (Chynoweth et al., 1993; Hansen et al., 2004). The BMP assays were conducted in 120 ml glass serum bottles. To determine the effects of F/I ratio, the anaerobic bio-digestion and methane production of the feedstock was investigated over the following range of F/I 0.5, 0.75, 1.0, 1.5, 2.0, 3.0, 4.0, and 5.0 g VS feedstock /g VS of inoculum. The F/I ratios were calculated based on the initial VS of the feedstock and VSS of the inoculum. To evaluate the effect of temperature, the reactors with the above F/I ratio were conducted in ambient and mesophilic (37°C) temperature. The optimum condition of temperature for the AD of the mixed vegetable was determined in this step, and that temperature would be applied for other experiments.

In order to investigate the influence of alkalinity on the methanization rate, reactors were added $NaHCO_3$ at concentrations of 300, 500, 600, 900 to 1200 mg $NaHCO_3$ per g VS of feeding (i.e. feedstock and inoculum). The buffer solution was prepared by adding 100 g of $NaHCO_3$ to 1000 mL of water.

The headspace of reactors was purged with nitrogen gas at 5-10 psi for a period of 5 min and capped tightly with rubber stoppers and screw cap. All reactors were operated at the mesophilic temperature (37°C). All assays were conducted in duplicate and the average results are presented. In each experiment, blank reactors that contained the same amount of inoculum and buffer solution were also set up. The biogas/methane production from the blank (inoculum) reactors was deducted from treatment reactors (waste samples). Thus, the presented data is only the biogas and methane production from the feedstock and not from the inoculum.

Experiments in the BMP test was also conducted to investigate an ability to recycle the digestate from the previous AD process. The digestate is the suitable inoculum for the new digestion process due to the acclimation of bacteria with the feedstock and the high value of alkalinity. As the results from the rectors with adding buffer solution, the highest biogas and methane achieved at the F/I ratio of 1.0, and $NaHCO_3$ concentration of 500 mg. Therefore, a lab-scale AD digester at the optimum conditions from BMP test was conducted for the mixed vegetable waste and UASB sludge. The digestate from the digester was used as an inoculum for a new AD process. In the experiment of reuse digestate, three F/I ratios of 0.5, 1.0, and 2.0 at the alkalinity concentration of 150, 250, and 350 mg was conducted.

The BMP tests were conducted in 250 ml glass bottles with a thick rubber septum. The representative sample of organic waste was collected from the vegetable market of Talaad Thai according to waste sampling methodology as mention in section 3.2. This sample was subsequently ground and sieved as far as to have a homogeneous material composed of particles with size ranging between 1 and 2 mm. The bottles were shaken manually once a day. Biogas production was measured using water displacement technique as in figure 3.3. Gas samples were taken periodically for composition analysis by gas chromatography using helium as carrier gas. All rectors were operated until no significant gas produced.

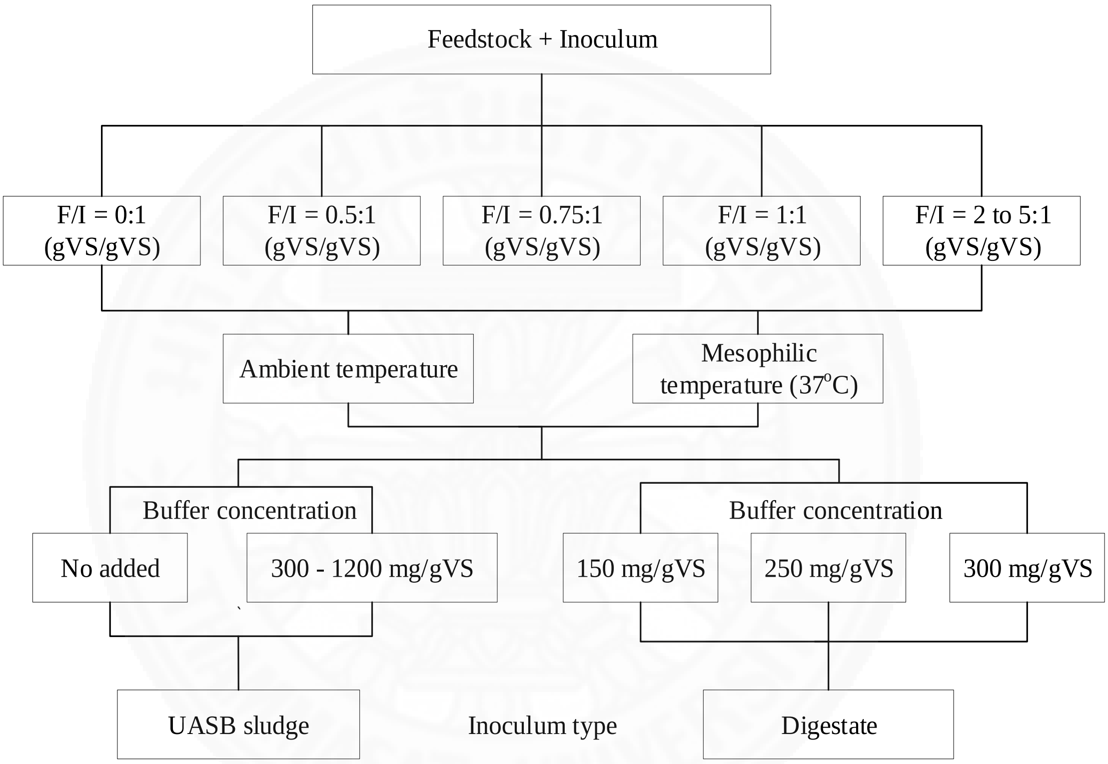


Figure 3.2 Schematic representation of BMP test

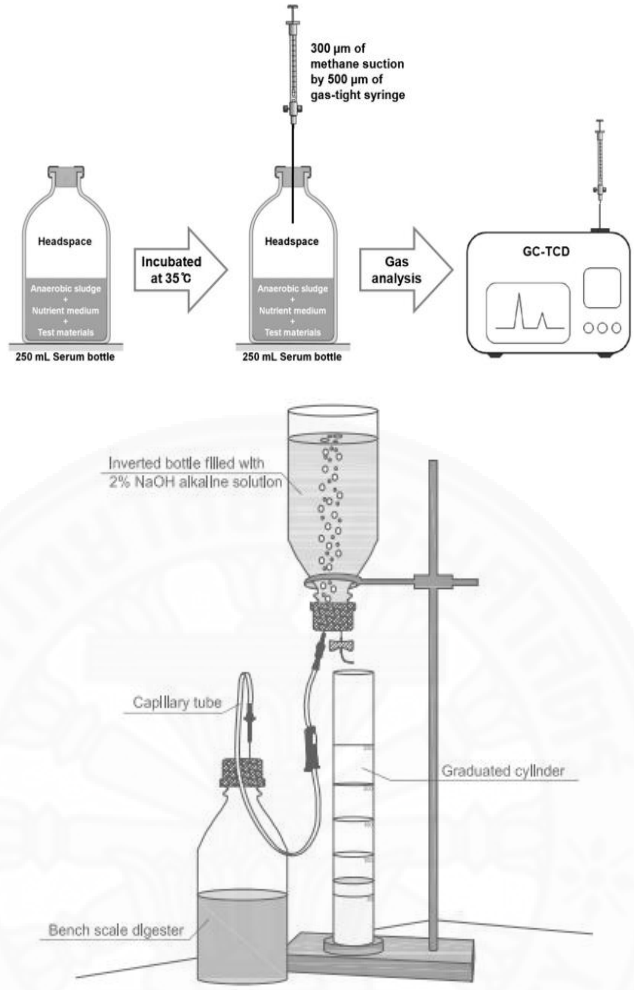


Figure 3.3 Experimental equipment used to measure the daily bio-methane production

3.4 Laboratory-scale AD reactors (Phase II)

After the optimum condition of temperature, F/I ratio, $NaHCO_3$ concentration, and retention time, the second phase of the study was conducted. In the phase, batch process was investigated.

3.4.1 Reactor design

In this experiment, the reactor was designed as the BIOCEL system. Digester design and dimensions are illustrated in figure 3.4. The reactor was made of acrylic. It had the cylindrical shape with screwed top and bottom covered with a layer of rubber gasket to avoid any air infiltration or leakages. The total volume of the reactor is approximately 6 L, while the designated volume available for the waste compaction is 4 L. The remaining volume of 0.9 L on the upper and lower part of the reactor serves as the available space for the generation of biogas and gravel support, respectively.

The reactor height is 350 mm. Two perforated plates with holes at 15 mm interval and thickness of 2 mm and consisting of holes with a diameter of 4 mm, were placed at both ends inside the reactor. The mixture feedstock and inoculum were loaded into the reactor and were stored between these two perforated plates to ensure distribution of leachate. The bottom part of the reactor was filled with gravel. The feedstock was separated from the gravel by the perforated plate. The upper 50 mm of the reactor is designed to provide allowable space for the installation of sprayer or sprinkler in order to distribute uniformly the feed water or recirculate leachate into the system and also provide a gap from the waste bed for biogas generation. The lower 24 cm of the reactor is designed for gravel support and drainage.

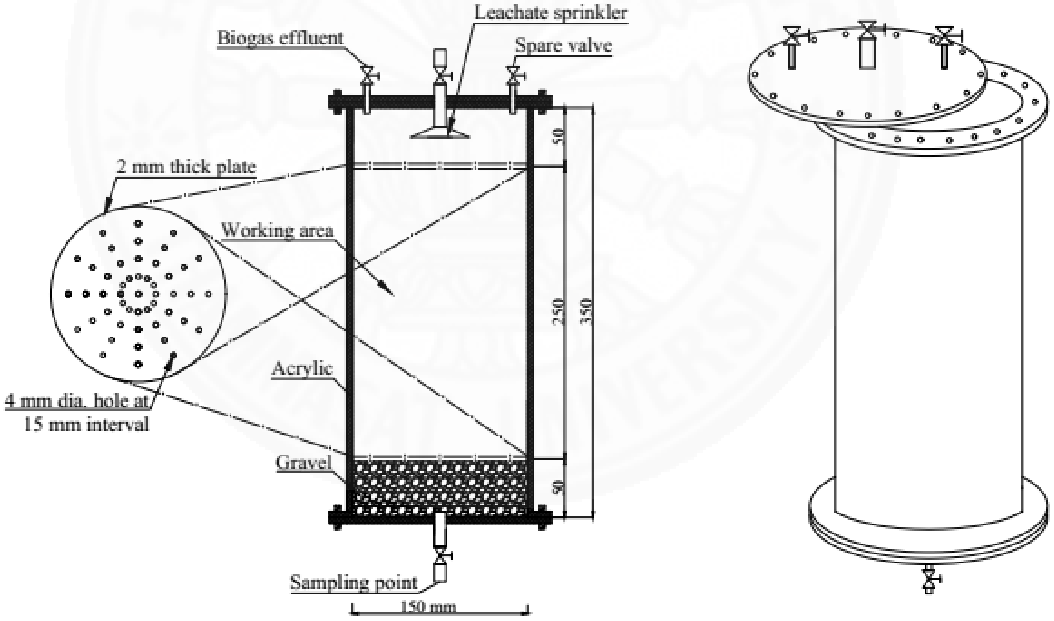


Figure 3.4 Lab scale reactor design for BIOCEL system

Leachate tank was made of plastic with the total volume of 2 L to store the wastewater from the BICOEL reactor. The tank was put inside a Styrofoam box to ensure the anaerobic condition (avoid the leakage of oxygen into the reactor). A submersible pump was also installed inside the leachate tank to recirculate the leachate into the BIOCEL reactor.

3.4.2 BIOCEL reactor

The reactor is an important vessel use to carry out anaerobic digestion of the waste bed. The other main equipment for the lab scale reactor consists of pressure sensor Keyence AP-30 series connect with a data logger to measure the daily volume of gas generated. 2 liters of Tedlar bags were used to store biogas generated from the reactor. The reactor was put in a box to control the desired temperature. The box was made from Styrofoam that has a rod for heating water. The temperature inside the box was controlled by a temperature controller that was connected to the rod. Details of the batch system are presented in figure 3.5.

The batch experiment was conducted to evaluate the performance of lab-scale BIOCEL reactor using optimum conditions obtained from BMP test. This helped to have modifications when a real scale AD reactor is built in the field. The first experiment was conducted at F/I ratio of 1.0 with the UASB sludge, the temperature of 37ºC, and $NaHCO_3$ concentration of 500 mg. The second experiment was conducted at the same conditions but the digestate from the last experiment was used as an inoculum.

(a)

(b)
Figure 3.5: BIOCEL reactor system

3.4.3 BIOCEL reactor operation

In the experiment, the optimum conditions on F/I ratios, temperature, and alkalinity concentration were applied for the BIOCEL reactor. The experimental procedure was operated as follows

- TS, VS, pH, and alkalinity of the vegetable waste and inoculum were determined before starting the experiment;
- The weight of the feedstock and inoculum were calculated following the optimum F/I ratio from BMP test;
- Mix the feedstock and inoculum completely before fed into the BIOCEL reactor;
- TS, VS, pH, and alkalinity of the mixtures were analyzed;
- Adding buffer solution as the optimum condition from BMP test;
- Checking the reactors to ensure having no leakage;
- Purging with nitrogen gas at 5 – 10 psi and capped tightly with the rubber seal and acrylic cap;
- Sampling and analyzing TS, VS, pH, and alkalinity of the mixture once every 5 days;
- Measuring biogas composition by GC machine once every 5 days;
- The leachate was recirculated four times per day by the pump installing inside the leachate tank;
- TS, VS, pH, and alkalinity of the digestate were analyzed at the end of the experiment.

3.5 Analytical method

3.5.1 Total solids and volatile solids

The total solids (TS) and volatile solids (VS) of the samples are determined by EPA Method 1684 (2001). For determining TS, samples with certain volume or weight are placed in ceramic vessels and dried in a drying oven at 105 ± 2 °C for 12 hours until constant weight. After cooling in the desiccators, the samples are weighed for TS measurement. The samples then oxidize at 550°C for 2 hours for VS determination. The VS is determined by subtraction of the minerals content of the sludge sample (residual ash after oxidation) from the total solids content.

$$\%\ \text{Total solids} = \frac{W_{\text{total}} - W_{\text{dish}}}{W_{\text{sample}} - W_{\text{dish}}} * 100$$

$$\%\ \text{Volatile solids} = \frac{W_{\text{volatile}} - W_{\text{dish}}}{W_{\text{total}} - W_{\text{dish}}} * 100$$

Where:

W_{total} = Weight of dried residue and dish (g)
W_{dish} = Weight of dish (g)
W_{sample} = Weight of wet sample and dish (g)
$W_{volatile}$ = Weight of residue and dish after ignition (g)

3.5.2 pH value

pH value of solid samples is determined by EPA Method 9045D. 20 g of waste sample in a 50-mL beaker, add 20 mL of reagent water, cover, and continuously stir the suspension for 5 min. Additional dilutions are allowed if working with hygroscopic wastes and salts or other problematic matrices. Stand waste suspension for about 15 min to allow most of the suspended waste to settle out from the suspension or filter or centrifuge off aqueous phase for pH measurement.

3.5.3 Biogas production and composition

The compositions of biogas (methane and carbon dioxide) were analyzed by a gas chromatograph (PerkinElmer, USA) equipped with a thermal conductivity detector

(TCD) and fitted with a Porapak Q, 50/80 mesh column. Helium gas was used as the carrier gas at a flow rate of 25 mL/min. The operating temperatures of column, detector, and injector were 45, 100, and 100ºC, respectively.

3.5.4 Total Kjeldahl nitrogen (TKN)

Total Kjeldahl Nitrogen (TKN) is used to determine the sum concentration of both organic nitrogen and ammonia nitrogen. The method involves a preliminary digestion to convert the organic nitrogen to ammonia, then distillation of the total ammonia into an acid absorbing solution and determination of the ammonia by titration method. The method employed sulphuric acid as the oxidizing agent. A catalyst was needed to hasten the oxidation of some of the more resistant organic substances. The oxidation proceeds rapidly at temperatures slightly above the boiling point of sulphuric acid (340 °C). The boiling point of the acid was increased by addition of sodium or potassium sulphate. When the organic nitrogen has been released as ammonia nitrogen, it was distilled into a solution of boric acid and determined titrimetrically with standard H_2SO_4 with a mixed indicator.

3.5.5 Elemental composition

Elemental composition of the feedstock and inoculum was analyzed by a Thermo Finnigan CHNS Flash EA 1112 elemental analyzer following the manufacturer's standard procedures.

3.5.6 VFAs

Total volatile fatty acids (VFAs) were determined by a gas chromatograph (PerkinElmer, USA) equipped with a flame ionization detector (FID) and fitted with a HP-INNOWax column. The injection port and detector temperature were at 250 and 275ºC, respectively. Oven temperature was set at 60ºC and was kept for 1 min, then raised to 170ºC by 10ºC/min. Helium gas was used as the carrier gas at 1.5 mL/min. The total VFAs was sum of all volatile acids produced.

3.6 Basic parameter calculations

3.6.1 Biogas production yield

Biogas and methane yields were determined by dividing the cumulative biogas and methane production by the amount of VS feedstock added into reactors.

3.6.2 TS reduction

The TS reduction was calculated based on measurements of the TS contents and total mass of feedstock and inoculum before and after the experiment.

$$\%\mathrm{TS} = \frac{\mathrm{M}_0 - \mathrm{M}_1}{\mathrm{M}_0}$$

M_0: dry weight of feedstock entering the reactor, g

$M_0 = TWW_0 \times TS_0$

TWW_0: total wet weight feedstock entering the reactor, g

TS_0: % of total solid of feedstock

M_1: dry weight of residual going out the reactor, g

$M_1 = TWW_1 \times TS_1$

TWW_1: total wet weight of residual going out the reactor, g

TS_1: % total solids of residual

3.6.3 VS reduction

The VS reduction was calculated based on measurements of the VS contents and total mass of feedstock and inoculum before and after the experiment.

$$\%\mathrm{VS} = \frac{\mathrm{N}_0 - \mathrm{N}_1}{\mathrm{N}}$$

N_0: weight of volatile solids of feedstock entering the reactor, g

$N_0 = M_0 \times VS_0$

VS_0: % of volatile solid of feedstock

N_1: weight of volatile solids of residual going out the reactor, g

$N_1 = M_1 \times VS_1$

VS_1: % volatile solid of residual

3.6.4 Mass removal

The mass removal is defined as the amount of substrate volatile solids and water (consumed during hydrolysis) converted to and removed as biogas. In this paper, the mass removal in term of biogas at the end of the digestion was calculated, based on the method by Richards et al. (1991) as follows:

$$\mathrm{MR_b} = \mathrm{V_0}(1.963 - 0.01249.\mathrm{CH_4})$$

Where MR_b = mass of biogas removed (g), and CH_4 = methane content of biogas (% of volume).

$$\mathrm{V_0} = \mathrm{DBF_t} \times \mathrm{V_t}$$

Where V_0 = dry (non-water) biogas volume at 0ºC (l); DBF_t = dry biogas factor (L dry at 0ºC) (measured at TºC)); V_t = biogas volume measured at temperature T (L)

Table 3.3: General information on the parameters analyzed for solid waste, leachate and gas characteristics

No.	Test parameter	Method	Standard	Experiments
		Organic waste, anaerobic sludge, and digestate		
1	Moisture content	Gravimetric analysis	EPA method 1684	BMP, Lab scale reactors
2	Volatile solid			
3	pH	pH meter	EPA method 9045D	
4	Total carbon		EPA method 9060A	
5	Total nitrogen	TKN method	EPA method 1687	
		Mixture of feedstocks		
6	Moisture content	Gravimetric analysis	EPA method 1684	Lab scale reactors
7	Volatile solid			
8	pH	pH meter	EPA method 9045D	
		Leachate		
9	Flow rate			Lab scale reactors batch system
10	pH	pH meter	EPA method 9045D	
11	COD			
12	VFA	Gas chromatograph (GC PerkinElmer, equipped with FID and fitted with a HP-INNOWAX column)	-	
13	Alkalinity	Volumetric titration method	EPA method	

No.	Test parameter	Method	Standard	Experiments
		Biogas		
14	Flow rate	Wet gas meter/gas counter	-	BMP, Lab scale reactors
15	Composition: CH_4, CO_2, H_2, H_2S	Gas chromatograph (GC PerkinElmer model AutoSystem XL, equipped with TCD and Porapak Q 50/80 mesh column)	-	

Chapter 4

Results and Discussion

4.1 Composition of solid waste from the vegetable market, Talaad Thai market

The composition of solid waste from the vegetable market is presented in Table 4.1. Twenty-seven samples were taken in one week (7 days), and the average values are shown. As shown in the data, organic wastes are the largest components of MSW, followed by plastics, and paper. The organic wastes were the mixture of tropical vegetable waste, mainly including leaves of cabbages, Chinese cabbage, cauliflower leaves, and few of broccoli stem, water spinach, lettuce, etc. The high biodegradable composition of the MSW from the Talaad Thai are favorable for biogas production. The waste composition at the vegetable market from Thailand's largest agricultural market can be used as reference data for another vegetable markets in the South East Asia region due to the similarities in culture and habits of people in the countries.

Table 4.1: Composition of solid waste from the vegetable market, Talaad Thai market, Thailand

No	Component	Composition (%)
1	Organic wastes	88.22
2	Plastic	7.52
3	Paper	2.32
4	Glass	1.79
5	Metal	0.05
6	Wood	0.09
	Total	100

Values represents the average ± SD of 27 samples

4.2 Characteristics of Feedstock and Inoculum

The characteristics of mixed vegetable wastes and granular sludge are shown in Table 4.2. As shown in the table, the vegetable wastes had very high moisture content. The VS of the mixed vegetable waste was 79.31% of TS, therefore this feedstock could be easily degraded in the AD process. As calculated from table 4.2, 1 g VS of the feedstock was about 19.94 g wet weight of the waste, while 1 g VS of inoculum was about 14.76 g wet weight of the sludge. pH of the mixed vegetable waste and inoculum in the study were 6.5 and 7.83, respectively. The required pH for fermentative bacteria is in a range of 5.0–8.5 (Zhang et al., 2014). From the elemental composition, the C/N ratio of the mixed vegetable waste and granular sludge were about 10 and 5.2, respectively.

Table 4.2: Feedstock and inoculum characteristics

Parameter	Units	Vegetable waste[a]	Granular sludge[a]
Total solid (TS)	(%)	9.60 ± 0.27	8.05 ± 0.03
Volatile solid (VS)	(%)	79.31 ± 0.99	89.36 ± 0.12
pH	-	6.8 ± 0.20	7.83 ± 0.10
Alkalinity	mg $CaCO_3$/L	5690 ± 590	1512 ± 314
	Elemental composition[b]		
Nitrogen (N)	(%)	3.82	8.83
Carbon (C)	(%)	37.91	45.99
Hydrogen (H)	(%)	7.45	7.25
Sulphur (S)	(%)	^{oo}ND	0.70
Oxygen (O)	(%)	50.82	37.93

[a] Values represents the average ± SD of 5 samples; [b] CHNS analysis of dry samples.

4.3 Effect of F/I ratio and temperature on biogas production

In this experiment, reactors in the BMP tests were conducted without adding external alkalinity. As presented in Chapter 3, F/I ratios of 0.5, 0.75, 1.0, 2.0, 3.0, 4.0 and 5.0 were operated in AD reactors to investigate the effect of F/I ratio on biogas production. The experiment also identified the effect of temperature on the AD performance through controlling two temperature conditions (i.e. 37°C and ambient temperature: 30±4°C).

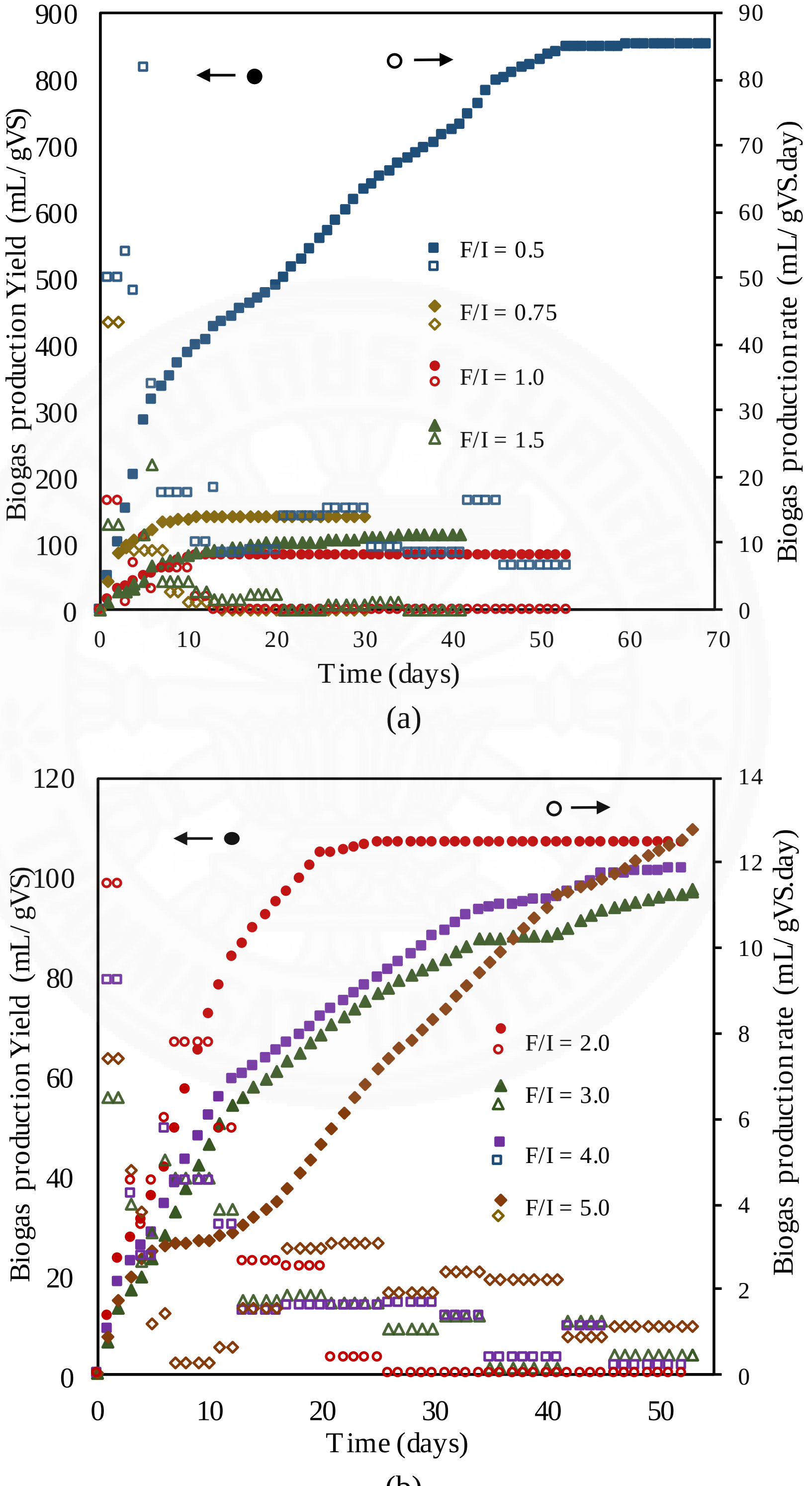
4.3.1 Biogas production at different F/I ratios at constant temperature 37°C

The biogas production of biodegradable solid wastes depends on the content of digestible carbohydrates, lipids, and proteins, as well as on the content of more resistant cellulose, hemicellulose and lignin (Hartmann and Ahring, 2006). Figure 4.1 shows the daily biogas production rate and yield at different F/I ratios at constant temperature (37°C). As the data shown in graphs, biogas started immediately after incubation at all F/I ratios and buffer concentrations. This may indicate fast acclimation of the AD microorganisms to the feedstock.

At the F/I of 0.5, daily biogas production rate was obtained very high comparing to the other F/Is. The daily biogas production rates at the F/Is of 0.75, 1.0, 2.0, 3.0. 4.0, and 5.0 achieved their peak values of 43, 17, 12, 7, 9, and 7 mL/g VS per day on the second day of experiment time, respectively. And the biogas production rate at F/I 0.5 and 1.5 reached the peak values of 82 and 22 mL/g VS per day on the fifth day. It can be seen from the figure, at F/I ratios of 0.75, 1.0, and 1.5, 90% of the biogas was released after the day 15 of experiment time. After 15 days, the biogas production ceased and even upon prolonged incubation, no biogas was evolved anymore. However, at F/I ratio of 0.5, 2.0, 3.0, 4.0, and 5.0, a large amount of biogas is still generated after day 15 of the experiment time.

As shown in the figure, the biogas production yields were not proportional to the VS added into reactors. The highest yield was obtained at F/I of 0.5 at the value of 855 mL/g VS, while the production yield of higher F/I ratios was lower than 200 mL/g VS. The lowest production yield was at F/I of 1.0 with 84 mL biogas per g VS added into the reactors. This is in line with the conclusion of Lesteur et al. (Lesteur et al., 2010), each feedstock has its optimum F/I ratio, depending on the amount of VFAs produced, and the capability to buffer the ammonium produced from the proteins hydrolysis process.

In addition, Fig 4.1 also depicts the optimum retention time (i.e. biogas generation time) of each F/I ratio. At F/I ratios of 0.75, 1.0, and 1.5 g VS feedstock/g VS inoculum, the maximum volume of biogas production was achieved after 15 days of experiment time. While F/I ratios of 2.0 and 3.0 g VS feedstock/g VS inoculum reached the maximum of biogas production after 25 and 30 days of experiment time, respectively. However, at F/I ratios of 0.5, 4.0, and 5.0, biogas is still being generated after 53 days of the experiment. The results are very important because they determine the optimum retention time of the feedstock when designing a pilot or full-scale anaerobic digestion reactors.



(a)

(b)

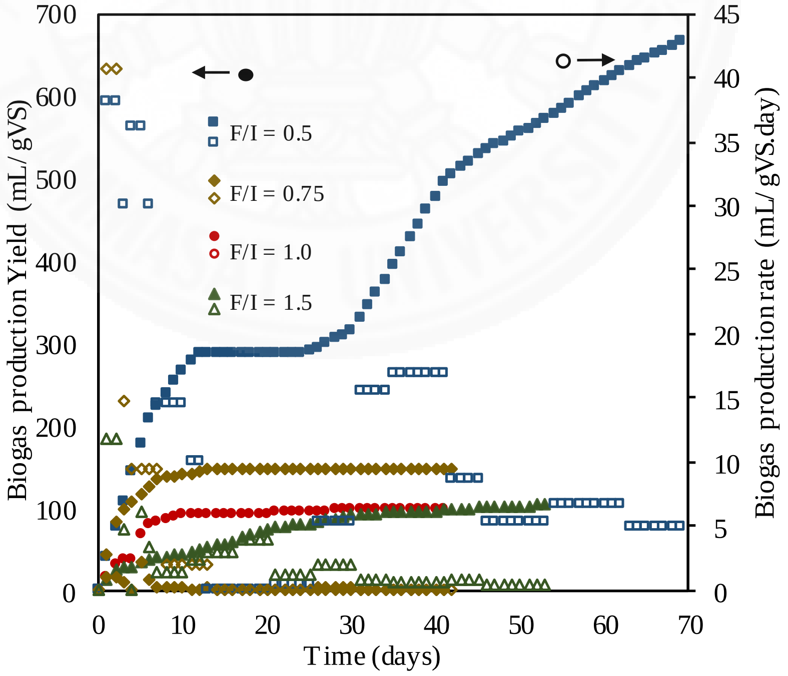
Figure 4.1: Biogas production rate at the different F/I ratios at temperature of 37ºC

(a) F/I ratios of 0.5 to 1.5; (b) F/I ratios of 2.0 to 5.0

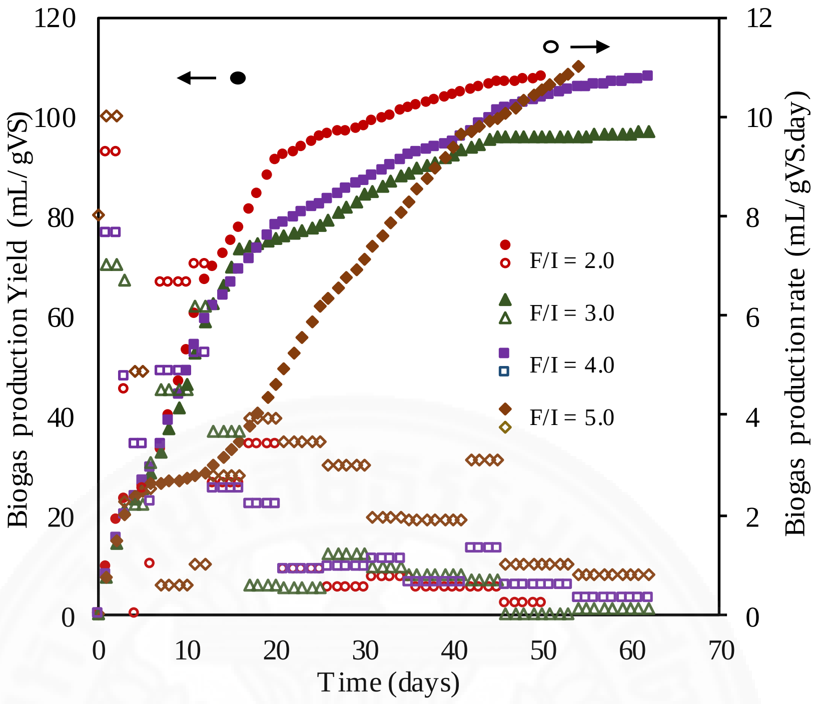
4.3.2 Biogas production at different F/I ratios at ambient temperature

The biogas production rate and yield of different F/I ratios at ambient temperature (30 ± 4°C) are shown in Fig 4.2. As presented in the figure, all results achieved at the ambient temperature is in line with results at the temperature of 37ºC. Similar to reactors at 37ºC, F/I of 0.5 gave a very high production rate compared to other F/I ratios. The daily biogas production rates of all F/I ratios achieved their peak values on the second day while, at F/I of 3.0, the production rate reached the peak at day 5 of experiment time. The peak daily production rates were 39, 41, 32, 12, 9, 7, 8, and 10 mL/g VS.day at F/I ratios of 0.5, 0.75, 1.0, 1.5, 2.0, 3.0, 4.0, and 5.0, respectively.

At all F/I ratios except F/I of 4.0 and 5.0 g, the biogas production at the constant temperature (37°C) was higher than that of the ambient temperature. The differences in biogas production between the constant and ambient temperature were 31%, 37%, 13%, 11%, and 9% at F/I of 0.5, 0.75, 1.5, 2.0, and 3.0, respectively. However, at the F/I ratios of 4.0 and 5.0 g VS, the biogas production under the ambient condition is higher than that of constant temperature, and the difference is about 5%.



(a)



(b)

Figure 4.2: Biogas production rate at the different F/I ratios at ambient temperature

(a) F/I ratios of 0.5 to 1.5; (b) F/I ratios of 2.0 to 5.0

From the data shown in graphs, biogas generation started immediately after incubation at all F/I ratios and buffer concentrations. The results from reactors having no external alkalinity demonstrate that biogas and methane yield is influenced by the F/I ratio: the higher the F/I ratio, the lower the biogas and methane yields. As shown in Table 4.2, the mixed vegetable wastes have low total solids and high volatile solids. Thus, the hydrolysis process of the feedstock is rapid. This leads to acidification of the digester and consequent inhibition of methanogenesis (Ward et al., 2008).

In comparison between two temperature conditions, the highest biogas production yields were achieved at F/I ratios of 0.5 for both temperatures. After 70 days of the experiment time, at the F/I ratios of 0.5, the cumulative biogas yields were 855 and 664 mL/g VS at 37ºC and 30±4°C. The results depict the important role of temperature in AD process of the mixed vegetable waste

4.4 Effect of F/I ratio and temperature on methane content

Methane, carbon dioxide, and hydrogen contents of biogas produced at difference F/I ratios under constant temperature (37°C) and ambient temperature (30±4ºC) are shown in Fig. 4.3 and 4.4. The average methane contents of the biogas produced from the mixed waste at the F/I of 0.5 was 52.3, and 42.1% (from 5 to 70 days) at 37ºC and ambient temperature. While the methane contents were very low at higher F/I ratios for both temperature conditions. The result gave percentages of methane in the biogas lower than that found in a batch experiment with maize waste carried out by Raposo et al. (2006) who obtained a methane percentage of 53% using similar F/I ratio at 37°C.

It can be seen from the Fig. 4.3, at F/I of 0.5, carbon dioxide was dominant in the mixture of biogas from day 3 to 25 of the experiment time at reactors of 37°C. At the same F/I ratio, reactors of ambient temperature had dominance of carbon dioxide for longer time, from day 3 to 35 of the experiment. The average value of carbon dioxide content at ambient temperature was also higher than that of 37°C reactors at 52.3 and 72.3%, respectively, during the experiment. This proved the methanogens in the study preferred temperature of 37°C compared with ambient temperature.

After 70 days of experiment time, cumulative methane yields at the F/I of 0.5 at 37ºC and ambient temperature were calculated to be 306 mL/g VS, and 230 mL/g VS, respectively. These values were comparable to the previous study of Nguyen (2004) who conducted BMP test on fresh market waste and obtained a value of around 300 mL/g VS. The results indicated the significant role of temperature on methane production yield. At the constant temperature (37ºC), the net methane yield of feedstock was higher than 46% compared to that yield at ambient temperature.

The methane conversion efficiency was very low for the other F/I ratios, after 70 days of experiment time. Even though high volumes of biogas were obtained, methane production of all F/I ratios is very low. From Fig. 4.3 and 4.4, at the F/I ratios higher than 2.0, a significant amount of hydrogen was produced in both temperature conditions. The details of hydrogen production are discussed in Section 4.4.

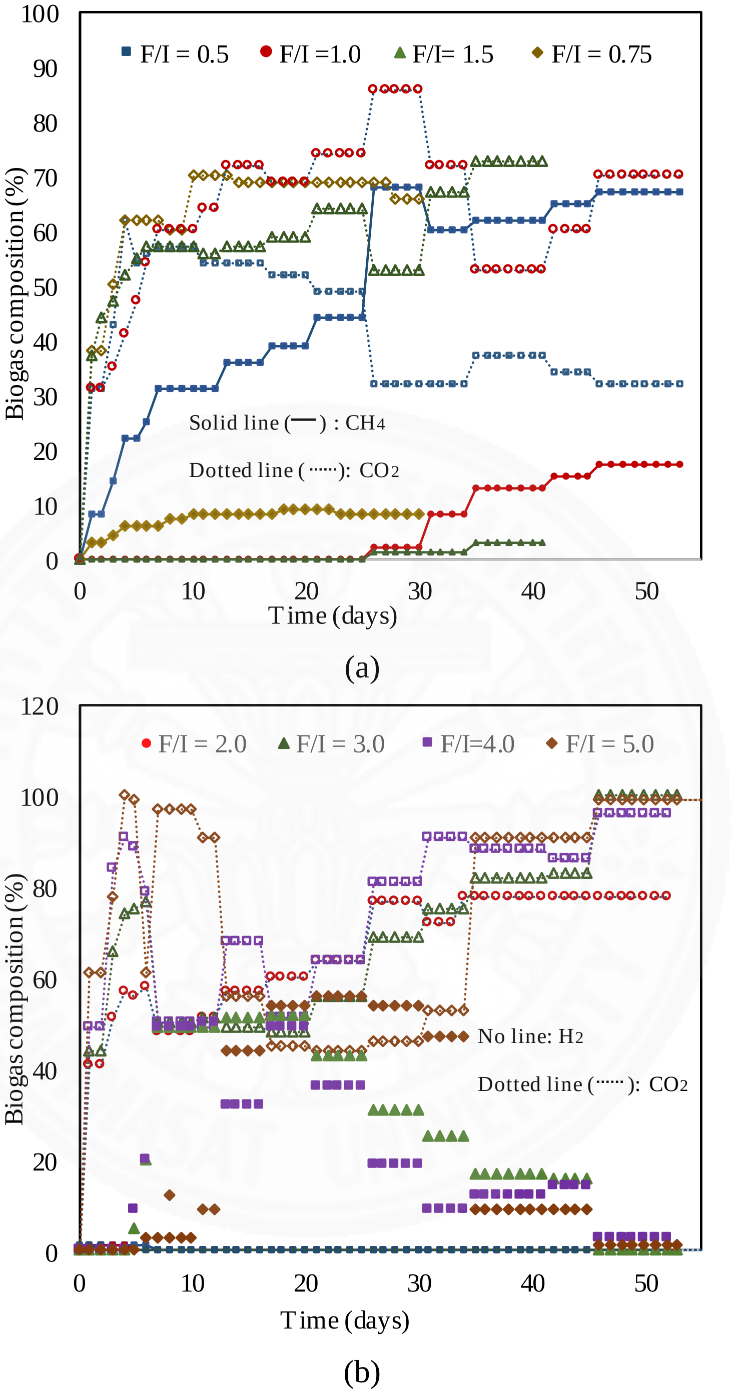


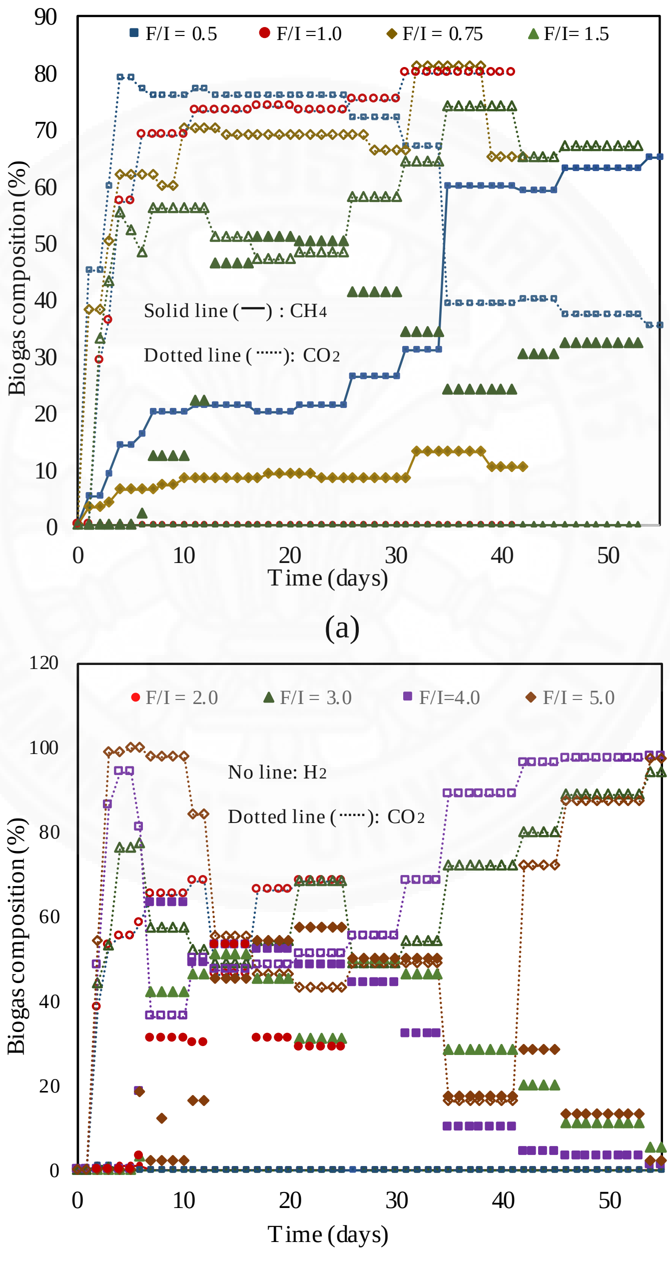
Figure 4.3: Biogas compositions at different F/I ratios at 37ºC

(a) F/I ratios of 0.5 to 1.5; (b) F/I ratios of 2.0 to 5.0

As shown in Section 2.1, the AD of an organic material such as the mixed vegetable wastes consists of four metabolic stages (Knol et al., 1978) (Fig. 2.1). First, particulate organic materials of the vegetable wastes like cellulose, hemicellulose, pectin, and lignin are liquefied by extracellular enzymes before being taken up by acidogenic bacteria (Koster, 1984). The rate of hydrolysis is a function of factors, such as pH, temperature, composition, and particle size of the substrate and high concentrations of intermediate products (Kübler and Schertler, 1994; Veeken et al., 2000). After that, soluble organic components including the products of hydrolysis are converted into organic acids, alcohols, hydrogen, and carbon dioxide by acidogenic. The products of the acidogenesis are then converted into acetic acid, hydrogen, and carbon dioxide. Finally, methane is produced by methanogenic bacteria from acetic acid, hydrogen, and carbon dioxide as well as directly from other substrates of which formic acid and methanol are the most important (Adney et al., 1991).

In this study, the low efficiency of methane conversion can be explained by VFAs accumulation when F/I ratio is higher than 0.5. According to Bouallagui et al. (2005), the AD is normally inhibited by the hydrolysis process. However, the AD of cellulose-poor wastes like the vegetable wastes is limited by methanogenesis rather than by the hydrolysis (Cecchi et al., 1986; Mata-Alvarez et al., 1990). These wastes, are very rapidly acidified to VFAs and tend to inhibit methanogenesis when the feedstock is not adequately buffered. VFAs accumulation at high organic loading decreases pH and inhibit methanogenesis (Neves et al., 2004; Zhang et al., 2014). After 70 days of experiment time, the pH of reactors operated at 37°C at an F/I of 1.0, 1.5, 2.0, 3.0, 4.0 and 5.0 were 5.5, 5.8, 5.7, 5.9, 6.0 and 5.8, respectively. This is a consequence of the VFAs accumulation in high concentrations at F/I ratios of 1.0, 1.5, 2.0, 3.0, 4.0, and 5.0 at 7994, 3662, 4937, 3078, 2376, and 3439 mg/L, respectively. From the values, the ratio of VFAs and total alkalinity (VFAs/TA) can be calculated as 2.47, 1.65, 1.87, 1.53, 1.64, and 1.60 at F/Is of 1.0, 1.5, 2.0, 3.0, 4.0, and 5.0, respectively. However, from theory, the VFAs/TA should be maintained below 0.4 to keep the AD process stable (USEPA, 1976). Callaghana et al (2002) concluded that if the ratio of VFAs/TA is higher than 0.8, the AD digester has significant instability. While the VFAs concentration at F/I of 0.5 was 2820 mg/L, and pH at the ratio was 8.5. This can explain the results of high biogas and methane yields at an F/I of 0.5 at 37ºC as the VFAs/TA was only 0.52. The results are in line with the study of Raposo et al. (2006). High concentrations of VFAs were found at F/I ratios higher than 0.3.

The VFAs concentrations were not proportional to the increase of F/I ratio. As shown in Fig. 1, the biogas yield at F/I of 2.0 is higher than that of another F/Is. This means that high amounts of fatty acids were converted to biogas at the acetotrophic phase in the reactors of 2.0 F/I. Therefore, this may decrease the amount of VFAs at F/Is higher than 2.0. The biogas mass removal, VS reduction, and final pH of all reactors in the experiment after 70 days of experiment time are shown in Table 4.4.



(a)

(b)

Figure 4.4: Biogas compositions at different F/I ratios at ambient temperature.

(a) F/I ratios of 0.5 to 1.5; (b) F/I ratios of 2.0 to 5.0

From the results, it can be concluded that without adding any buffer solutions, the optimum F/I ratio of the methane conversion rate of the mixed vegetable waste was 0.5. At a higher F/I ratio, external alkalinity sources are necessary to maintain a suitable pH range for methanogenic bacteria during the fermentative process. It is necessary to improve the AD performance at F/I ratio higher than 0.5. This helped to reduce the operation fee due to more mixed vegetable waste can be loaded for the same volume of a reactor in the AD treatment. The experiment also found the suitable temperature for operating the AD reactors was 37°C. Therefore, the following experiments were conducted at the temperature of 37°C.

4.5 Effect of F/I ratio and temperature on hydrogen production

Theoretically, the BMP assays are only conducted to determine the biomethane potential from biodegradable organic matters. However, when the experiment was operated, significant amounts of hydrogen gas were also detected. According to the principle of anaerobic digestion, in the acetogenesis process, VFAs are converted into acetate, hydrogen gas, and carbon dioxide. This conversion is governed by the action of obligate hydrogen producing acetogenic bacteria, which are considered as acetogenins and acidogenic. The latter occurs under syntrophy phenomena. After this step, hydrogen is converted to methane gas by methanogenic bacteria through hydrogenotrophic methanogenesis process. However, at the F/I ratios higher than 2.0 g VS feedstock/ g VS inoculum, the VFAs was accumulated at high organic loading and inhibited activities of methanogenic bacteria. Thus, in the study, the anaerobic digestion was stopped at acetogenesis process, and large amounts of hydrogen gas were obtained.

High amounts of hydrogen were found at F/I ratios in a range of 2.0, 3.0, 4.0, and 5.0 g VS feedstock/ g VS inoculum (Fig. 4.5). The hydrogen generation started after 6 days of experiment time and lasted for 40 days. The hydrogen production also increased with increasing F/I ratio (i.e. the hydrogen biogas production was proportional to the VS added). At the F/I ratios of 2.0 and 3.0, the hydrogen production at constant temperature (37°C) was higher than that of ambient temperature. However, for the F/I ratios of 4.0 and 5.0 g VS feedstock/ g VS inoculum, the achievement of hydrogen production under ambient temperature condition was higher than that of the constant temperature condition.

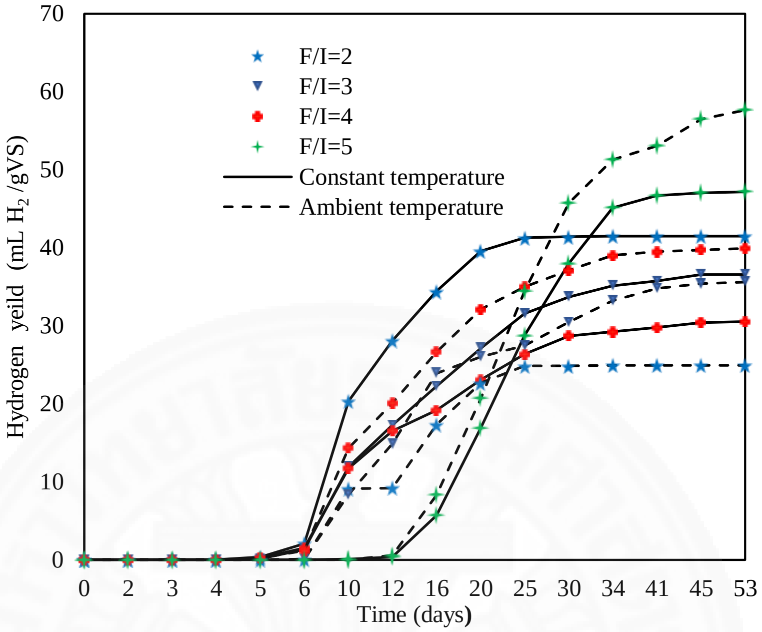


Figure 4.5: Hydrogen production yields at the different F/I ratio

(constant temperature 37°C, Ambient temperature 30±4°C)

4.6 Digestion of the mixed vegetable waste at different F/I ratios and buffer solution concentrations

Based on the results without alkalinity as above, external alkalinity as $NaHCO_3$ was added into reactors to improve the biogas yields, and methane contents. The concentrations of sodium bicarbonate were calculated on the total VS mass of feedstock and inoculum added into reactors. Four concentrations at 300, 500, 600, 900, 1200 mg $NaHCO_3$/g VS with F/Is of 0.5, 1.0, and 2.0, were investigated in the study. All reactors were operated at 37°C as the optimum condition found in Section 4.4.

4.6.1. Digestion of the mixed vegetable waste at an F/I of 0.5

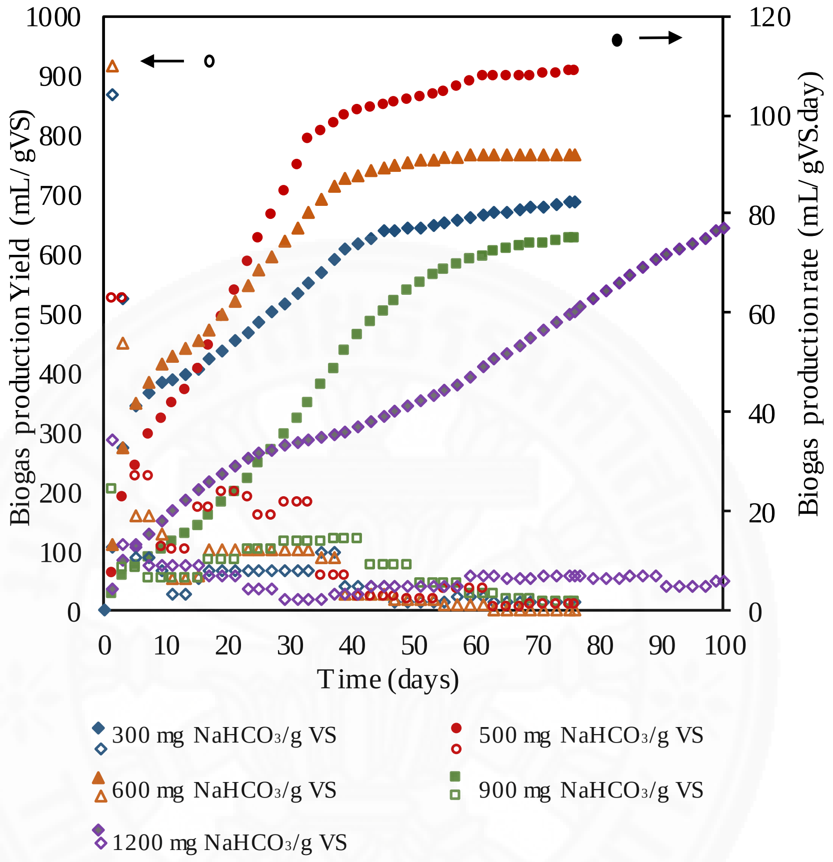
The biogas yield and production rates of the mixed vegetable wastes at an F/I of 0.5 during the experiment time are presented in Fig. 4.6 (a). The biogas production also started from the first day, same as the reactors without adding buffer solution. The daily biogas production rates at sodium bicarbonate concentrations of 300, 500, 600, 900, and 1200 mg, reached their peak values of 104, 110, 24, 63, 34 mL/g VS per day, respectively, on the second day.

Unlike reactors having no external buffer, the biogas production continued after 70 days and lasted until 75 days of experiment. After 75 days of experiment time, the cumulative biogas yields were 688, 907, 767, and 626 mL/g VS at $NaHCO_3$ concentrations of 300, 500, 600, and 900 mg/g VS, respectively. While at a buffer concentration of 1200 mg, the biogas production lasted until 100 days with the cumulative biogas yields of 637 mg/L. After 70 days of experiment time, compared to the same F/I of 0.5 in reactors having no external buffer solution, the biogas yields at buffer concentrations of 300, 600, and 900 mg/g VS were lower. However, at a $NaHCO_3$ concentration of 500 mg/g VS, the biogas yield at the F/I of 0.5 was higher than that of reactors having no external buffer solution.

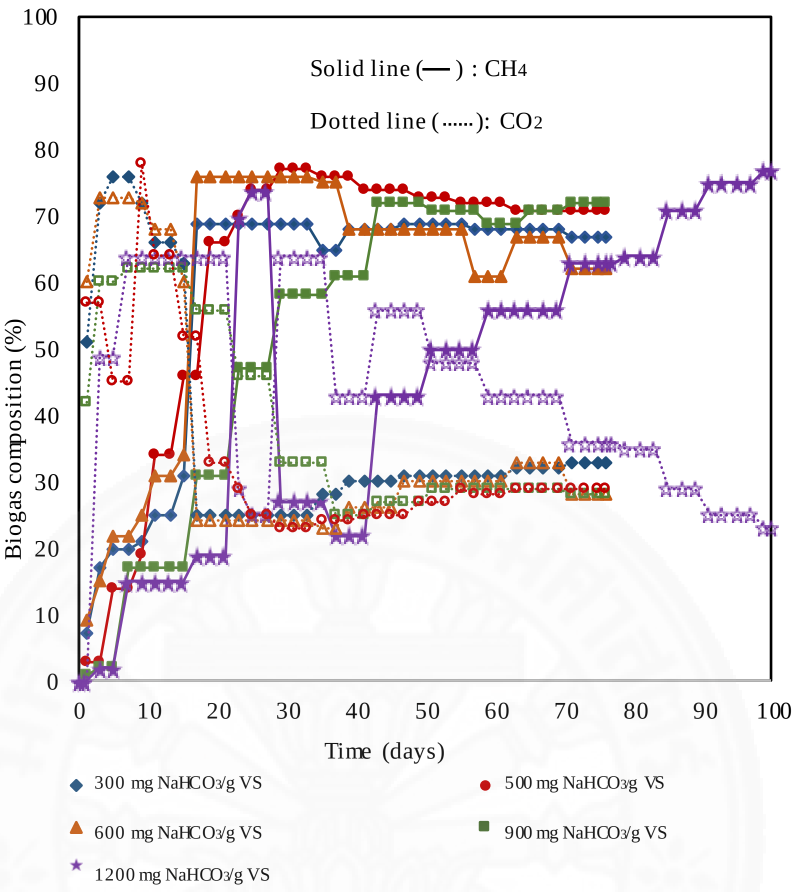
As shown in Fig. 4.6 (b), the average methane contents of the biogas produced from the mixed vegetable wastes were 61.5, 65.4, 62.0, 55.0, and 47.4%, (from 5 to 75 days) at buffer concentrations of 300, 500, 600, 900, and 1200 mg/g VS. Thus, the cumulative methane yields were 306, 371, 327, 314, and 265 mL/g VS, respectively. The results depicted that higher maximum methane yields were obtained when adding external buffer solution at the same F/I ratio. At an F/I of 0.5, adding 500 mg of $NaHCO_3$ gave the highest biogas and methane yields.

At $NaHCO_3$ concentrations of 300, 500, and 600 mg, carbon dioxide was dominant in the mixture of biogas from day 1 to 15 of the experiment. At the buffer concentration of 900 and 1200 mg, the longer time for dominance of carbon dioxide found from day 1 to 21 of the dig experiment time. The average values of carbon dioxide content at $NaHCO_3$ concentration of 300, 500, 600, 900, and 1200 mg were 67.2, 58.6, 68.1, 54.9, and 60.8%, respectively, during the experiment. In comparison to digesters having no external buffer, the lag phase of methane production was improved as $NaHCO_3$ added.

It can be concluded that at the F/I of 0.5, the biogas production yields and methane contents reduced as the $NaHCO_3$ concentration increased higher than 600 mg. This may due to the excess alkalinity at increased pH of the environment over the optimum range of AD bacteria. The biogas mass removal and VS reduction of reactors after 75 days of experiment time are shown in Table 4.4. The alkalinity values at the end of experiment were also analyzed. At buffer concentrations of 300, 500, 600, 900, and 1200 mg/g VS_{added}, the final alkalinity values were 7680, 9440, 8160, 14560, and 16800 mg $CaCO_3/L$, as shown in Table 4.3.



(a)



(b)

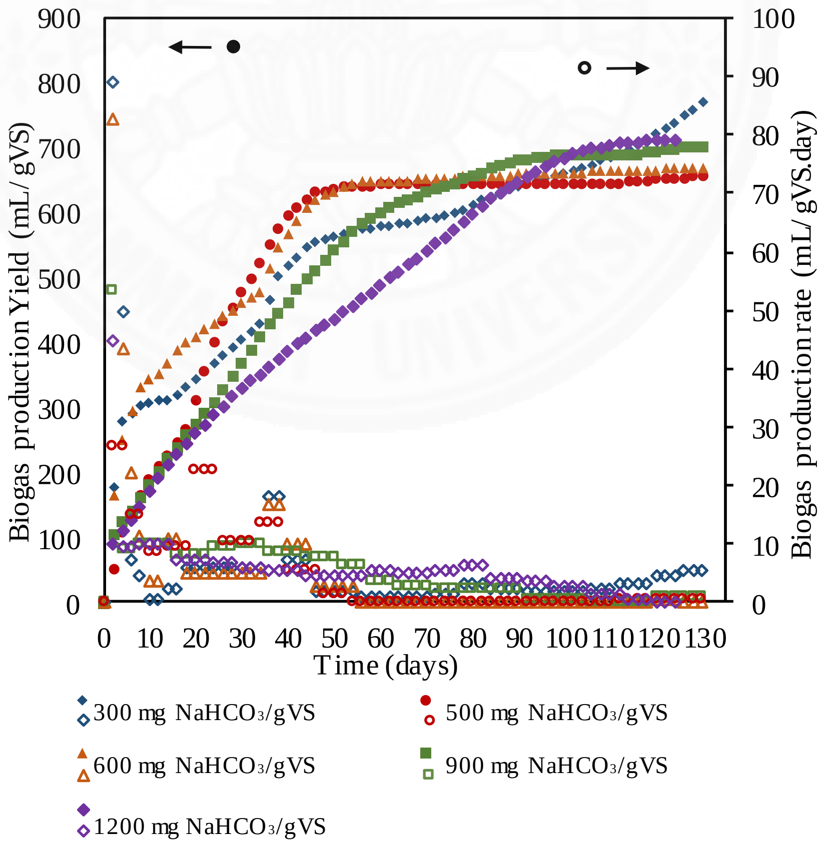
Figure 4.6: Anaerobic digestion of mixed vegetable waste at F/I of 0.5 and temperature of 37°C. (a) Biogas production (b) Biogas composition

4.6.2 Digestion of the mixed vegetable waste at an F/I of 1.0

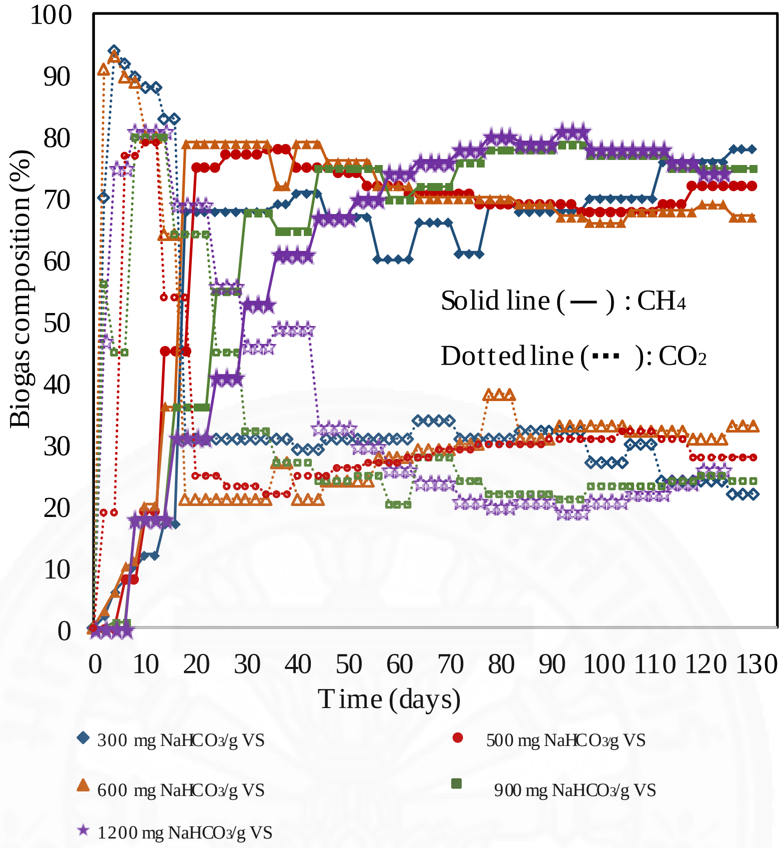
Figure 4.7 (a) shows the daily production rates and biogas yields of the mixed vegetable wastes at an F/I of 1.0. The biogas production rates at buffer concentrations of 300, 500, 600, 900 and 1200 mg reached their peak values of 89, 27, 83, 54, and 45 mL/g VS per day, respectively, on the second day of experiment time. Compared to an F/I of 0.5, these peak values are lower. However, the biogas production time at an F/I of 1.0 is longer and lasted until 130 days. Thus, the cumulative biogas yields of 1.0 F/I at the end of experiment were higher than that of 0.5 F/I. From Fig. 4.7 (a), it can be depicted that 90% of biogas was released after 50 days of experiment time at the $NaHCO_3$ concentrations of 500 and 600 mg. While biogas still produced in a significant amount at buffer concentrations of 300, 900 and 1200 mg.

The average methane contents in the mixture of biogas from the digestion of the vegetable waste were 64.1, 67.3, 67.3, 64.7, and 64.2% (from 5 to 130 days), at buffer concentrations of 300, 500, 600, 900, and 1200 mg, respectively, as shown in Fig. 4.7 (b).

At $NaHCO_3$ concentrations of 300, 500, and 600 mg, carbon dioxide was dominant in the mixture of biogas from day 1 to 16 of the experiment. At the buffer concentration of 900 and 1200 mg, the longer time for dominant of carbon dioxide found from day 1 to 25 of the experiment time. The average values of carbon dioxide content at $NaHCO_3$ concentrations of 300, 500, 600, 900, and 1200 mg were 87.7, 53.8, 78.1, 63.5, and 70.2%, respectively, during the experiment time. The results again show the low AD performance of reactors at $NaHCO_3$ concentrations of 900 and 1200 mg. The biogas mass removal, cumulative methane yields, and VS reduction of the vegetable wastes after 130 days of the experiment time are presented in Table 4.4.



(a)



(b)

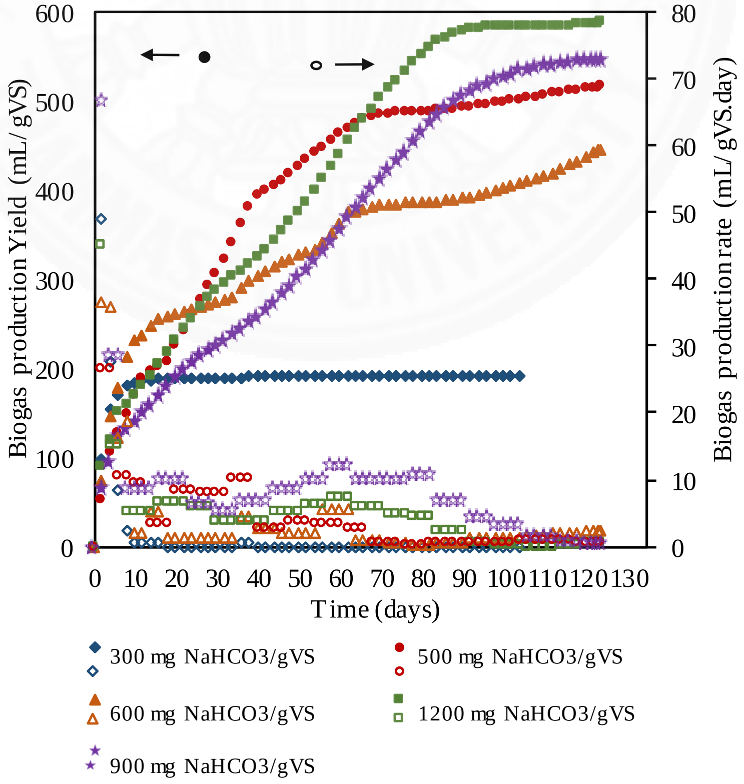
Figure 4.7: Anaerobic digestion of mixed vegetable waste at F/I of 1.0 and temperature of 37°C. (a) Biogas production (b) Biogas composition

4.6.3 Digestion of the mixed vegetable waste at an F/I of 2.0

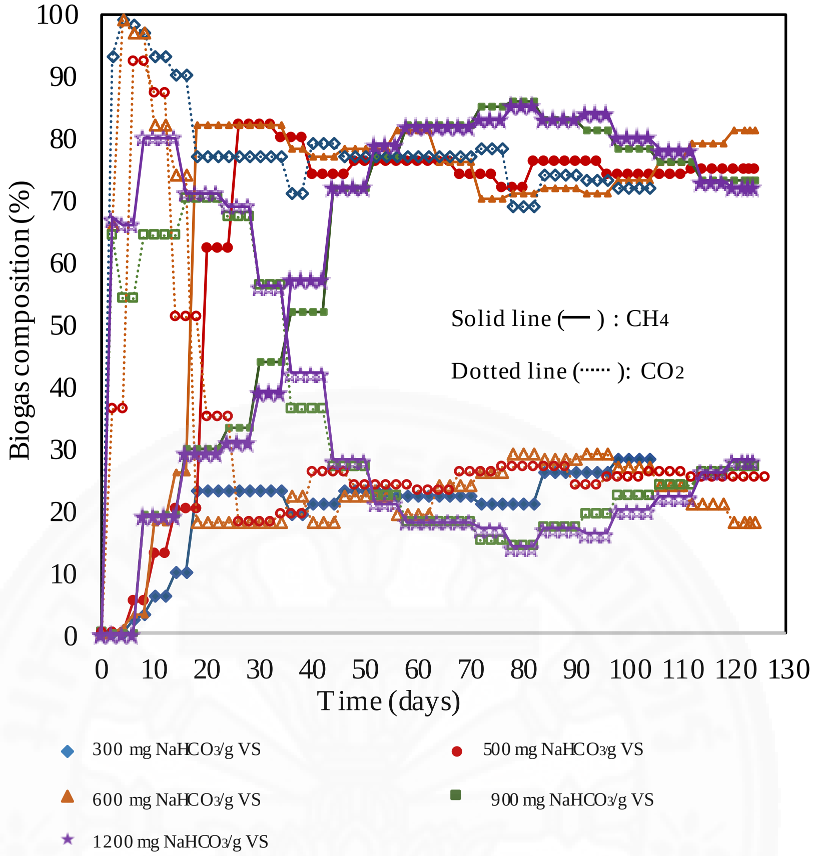
The biogas production yields and daily biogas production rates during 125 days of experiment at an F/I of 2.0 are presented in Fig. 4.8 (a). Similar to an F/I of 1.0, on the second day, reactors achieved their peak values of biogas production rates at 49, 27, 37, 45, and 34 mL/g VS per day with a buffer concentration of 300, 500, 600, 900, and 1200 mg, respectively. The biogas production at a concentration of 300 mg $NaHCO_3$/g VS stopped at 15 days of experiment. At the concentrations of 500, 600, 900, and 1200 mg, the biogas was still produced until 125 days of experiment. From Fig. 4.7, it can be depicted that 90% of biogas was released after 50 days of experiment time at the $NaHCO_3$ concentrations of 500, and 600 mg. While biogas still produced in a significant amount at buffer concentrations of 900 and 1200 mg. It can be seen from Fig. 4.8 (a), at the $NaHCO_3$ concentration of 300 mg, 95% of biogas was released after 10 days of experiment.

The average methane contents of the biogas produced from the mixed vegetable wastes at four buffer concentrations of 300, 500, 600, 900, and 1200 mg were 22.2, 68.2, 71.0, 64.9, 65.1%, respectively. Biogas composition as contents of methane and carbon dioxide under different conditions during the experiment are shown in Fig. 4.8 (b). As shown in the figure, average values of carbon dioxide content at $NaHCO_3$ concentrations of 500, 600, 900, and 1200 mg were 66.8, 86.6, 63.1, and 69.6%, respectively, during the experiment time. The results again show the low AD performance of reactors at $NaHCO_3$ concentrations of 900 and 1200 mg.

At the $NaHCO_3$ concentration of 300 mg, carbon dioxide content was dominant in the mixture of biogas during 105 days of experiment time. As shown in Table 4.3, the final alkalinity at an F/I of 2.0 and $NaHCO_3$ of 300 mg was 6720 mg $CaCO_3$/L. The value is higher than that of the reactors having no external alkalinity. However, it maybe not enough to maintain the effectivity of methanogens. The final pH value, and biogas mass removal in all reactors at the end of experiment are shown in Table 4.4. Alkalinity values, before and after operation of the experiments, are presented in Table 4.3.



(a)



(b)

Figure 4.8 Anaerobic digestion of mixed vegetable waste at F/I of 2.0 and temperature of 37°C. (a) Biogas production (b) Biogas composition

As expected, adding external alkalinity as $NaHCO_3$ improves the biogas and methane yields significantly at high F/I ratios of 1.0 and 2.0. As shown in Table 4.4, cumulative biogas yields at an F/I of 1.0 achieved the highest values at a buffer concentration of 300 mg. At concentrations of 500 and 900 mg, an F/I ratio of 1.0 gave a lower biogas yield. However, the methane yield was not much different, compared to the bicarbonate concentrations of 300 mg. At an F/I of 2.0, the biogas and methane yields achieved peak values at $NaHCO_3$ concentrations of 500 and 900 mg, but the ratio gave lower values of gas production, compared to an F/I of 0.5 and 1.0 at all buffer concentrations. With a concentration of 300 mg, the biogas yield was improved significantly compared to experiments having no external alkalinity. However, the methane content in the biogas was still very low. This again shows a shortage of alkalinity at high F/I ratios. The improvement of methane contents at 1.0 and 2.0 F/I benefits the economic costs. The higher the F/I ratio, the lower the inoculum added into the reactors. This means that the reactors can be loaded with more mixed vegetable wastes for the same volume of a reactor in the treatment process.

Table 4.3 presents the alkalinity values of all experiments at the beginning and ending of experiment. It can be seen that at the beginning of the AD process, the alkalinity of all reactors is lower than that of the ending process. In the hydrolysis process of the AD, VFAs are generated in high concentrations, decreasing the pH rapidly. However, if the buffer capacity is suitably maintained in the process, methanogens consume VFAs and produce alkalinity (Guwy et al., 1997; Ward et al., 2008). This makes the pH of the digester increase with increased stability. Thus, as the AD reactors are operated properly, the alkalinity at the end of the process is higher than the value at the beginning. For reactors having no external buffer source, an F/I of 0.5 gave a higher alkalinity concentration than the values at an F/I of 1.0 and 2.0. This is in line with the conclusion of Gunaseelan (1995) that the feed to inoculum ratio can affect the buffer capacity and pH of AD.

The data from the study also shows that the biogas and methane yield are not proportional to the increased bicarbonate concentration. At an F/I of 0.5, as the $NaHCO_3$ concentration increases from 300 to 500 mg, biogas and methane yields were improved to 27 and 21%. However, at concentrations of 600, 900, and 1200 mg $NaHCO_3$, biogas and methane decreased. This may be due to excess alkalinity at increased pH of the process and the rapid conversion rate of ionized ammonia nitrogen into free ammonia nitrogen (FAN). The increased amount of FAN inhibits the methanogenic bacteria, resulting in the accumulation of VFAs (Rajagopal et al., 2013). The biogas and methane yields of reactors at an F/I of 1.0 decreased as the bicarbonate concentration increased from 300 and 600 mg. At the buffer concentration of 900 mg, the biogas and methane yields increased, but was still lower than from 300 mg $NaHCO_3$. Similar to an F/I ratio of 1.0, cumulative biogas and methane yields of reactors at an F/I of 2.0 increased with increased bicarbonate concentration. However, at a concentration of 600 mg, biogas and methane production decreased.

When considering the cumulative biogas and methane yields, and retention time of the AD, the optimum conditions were at $NaHCO_3$ concentration of 500 mg for all F/I ratios of 0.5, 1.0 and 2.0. The methane yeilds of the conditions are presented in Fig 1 (Appendix B).

Table 4.3: Alkalinity values before and after operation in all reactors

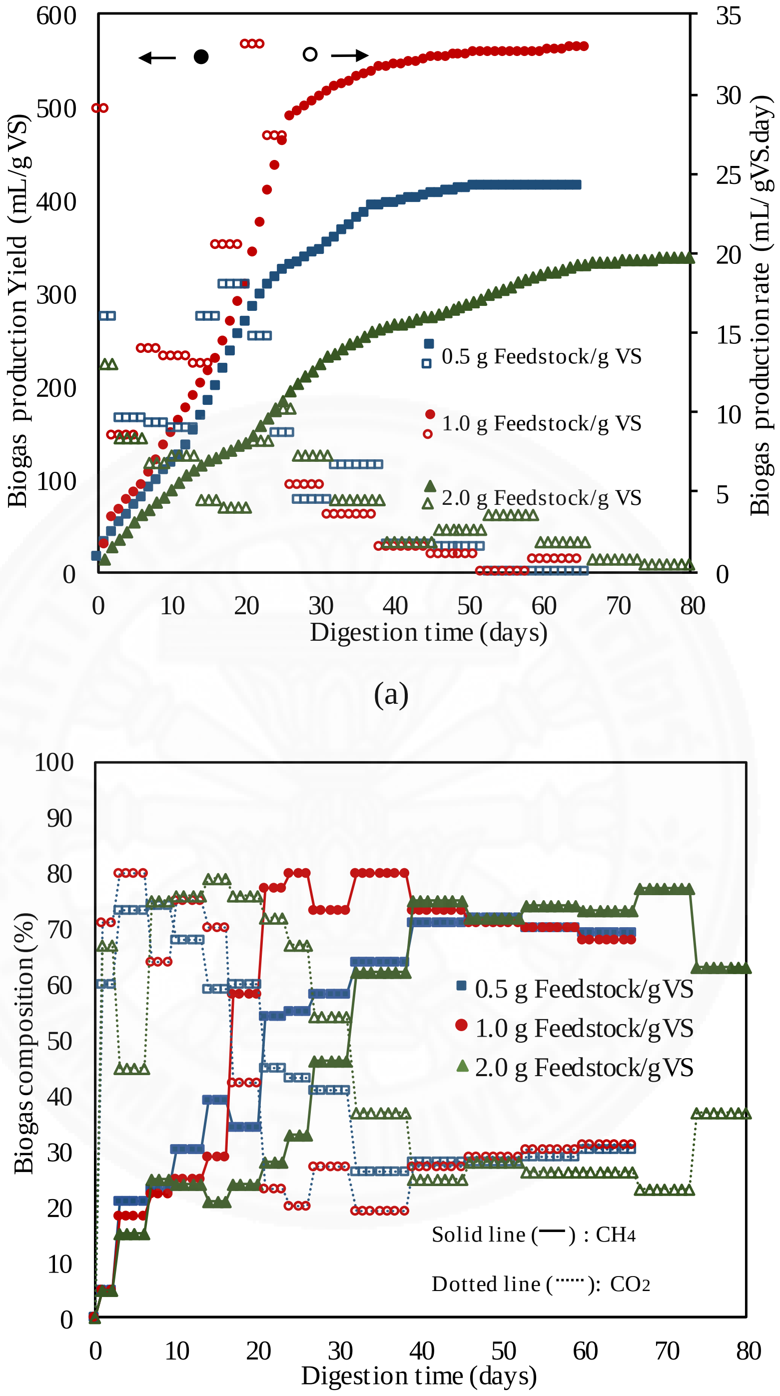
Buffer concentration added (mg $NaHCO_3$/g VS)	F/I ratio	Alkalinity (mg $CaCO_3$/L) Before operation	Alkalinity (mg $CaCO_3$/L) After operation
Without addition (37°C)	0.5	1120	5400
	1.0	1280	3240
	2.0	1440	2640
300	0.5	4960	7680
	1.0	5920	8320
	2.0	6080	6720
500	0.5	6760	9440
	1.0	8320	10240
	2.0	8640	9600
600	0.5	6840	8160
	1.0	8733	9440
	2.0	7600	8160
900	0.5	11040	14560
	1.0	11360	12320
	2.0	10560	10560
1200	0.5	13400	16800
	1.0	12560	16320
	2.0	12050	15680

4.6.4 Effect of thermophilic temperature on AD performance at different F/I ratios

As shown in previous studies (Hansen et al., 2004; Gannoun et al., 2007; Kothari et al., 2014), a higher VS reduction and biogas yield were achieved as AD reactors are operated at thermophilic temperature compared to mesophilic reactors. Thus, the experiment was conducted to investigate the effect of thermophilic temperature on the AD performance of the mixed vegetable wastes. From results in Section 4.6.1, 4.6.2, and 4.6.3, the AD reactors showed a high performance at $NaHCO_3$ concentration of 500 mg for all F/I ratios at temperature of 37°C. Therefore, the experiment was operated at F/I ratios of 0.5, 1.0, and 2.0 at the buffer concentration of 500 mg and temperature of 50°C.

The biogas production yields and daily biogas production rates at different F/I ratios are presented in Fig. 4.9 (a). Similar to other experiments, the biogas at the condition was released in the first days of experiment. The peak values of biogas production rates were at 16, 29, and 13 mL/g VS per day with F/I ratios of 0.5, 1.0, and 2.0, respectively. As shown in Fig 4.8 (a), almost 90% of biogas was produced on the day 35, 30, and 50 of the experiment at F/I ratios of 0.5, 1.0, and 2.0, respectively. The cumulative biogas yield was highest at the F/I of 1.0 with the shortest experiment time. The cumulative biogas yields were 177, 276, and 130 mL/g VS at F/I of 0.5, 1.0, and 2.0, respectively.

The average methane contents in the mixture of biogas from the digestion of the vegetable waste were 54.9, 60.5, and 54.2%, at F/I ratios of 0.5, 1.0, and 2.0 mg, respectively, as shown in Fig. 4.9 (b). It can be seen from the figure that carbon dioxide was dominant in the mixture of biogas from day 1 to 20 of the experiment at F/I ratios of 0.5 and 1.0. However, at F/I of 2.0, the longer time for dominance of carbon dioxide was found from day 1 to 31 of the experiment time.



(a)

(b)

Figure 4.9: Anaerobic digestion of mixed vegetable waste at different F/I ratios and temperature of 50°C. (a) Biogas production (b) Biogas compositions

In comparison to AD reactors operated at the buffer concentration of 500 mg and 37°C, the lower AD performance was found for AD reactors operated at the temperature of

50°C. The thermophilic reactors shorten the experiment time compared to the mesophilic reactors. However, the final biogas yields at thermophilic reactors were lower than that of mesophilic reactors at 54, 16, and 36%, at F/I of 0.5, 1.0, and 2.0, respectively. It can be explained because the inoculum used in the experiment was from a mesophilic UASB reactor. Thus, as the inoculum was applied in the thermophilic temperature, the activity of anaerobic microorganisms was limited. In the experiment, to determine the effect of thermophilic temperature on the AD, the inoculum should be taken from thermophilic reactors. Unless the mesophilic inoculum has to be operated several cycles in thermophilic to make acclimation of microorganisms with the high-temperature condition.

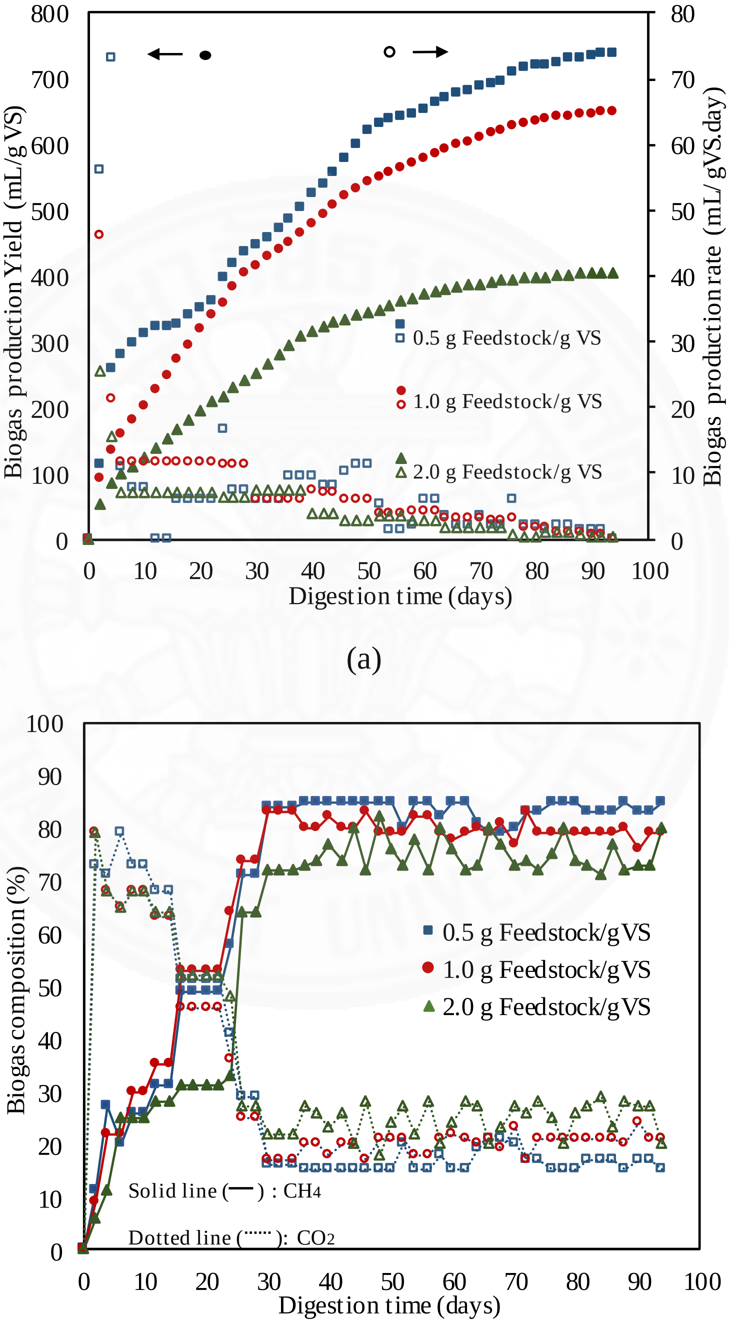
4.6.5 Recycling of digestate as a source of inoculum and buffer solution

As shown in Section 4.5.3, the biogas and methane yield were improved by adding external buffer solution. At the end of experiments in the study, the alkalinity remained at a high concentration in all reactors. This can be a suitable inoculum source with high values of alkalinity in the digestate that can help to buffer the environment in new processes. Therefore, the digestate from an anaerobic digester with an F/I ratio of 1.0 at the $NaHCO_3$ concentration of 500 mg was recycled in the experiment. Three F/Is of 0.5, 1.0, and 2.0 at a $NaHCO_3$ concentration of 150 mg were investigated in the study.

As presented in Fig. 4.10 (a), at F/Is of 1.0 and 2.0, daily biogas production rates obtained peak values at 35 and 87 mL/g VS per day, respectively, on the second day. While the F/I of 0.5 reached its peak on the third day of experiment time at 73 mL/g VS per day. At all reactors, biogas produced until day 40, then gradually leveled off and stopped at 90 days. The cumulative biogas yields were 737, 648, and 404 mL/g VS at F/Is of 0.5, 1.0 and 2.0, respectively.

Biogas composition at different F/I ratios during the experiment are shown in Fig. 4.10 (b). As shown in the figure, carbon dioxide was dominant in the mixture of biogas from day 1 to 20 of the experiment at all F/I ratios. The average methane contents at the end of the experiment were 70.3, 68.9, and 61.7% at F/I of 0.5, 1.0, and 2.0, respectively.
In comparison with reactors using the UASB sludge as inoculum, the biogas and methane yields of recycled digestate gave comparable results with experiments. Thus,

it is possible to reuse the digestate of the last AD as an inoculum and alkalinity source. The calculated mass of biogas produced, cumulative methane yields, and VS reduction of the vegetable wastes after 130 days of the experiment time are presented in Table 4.4.



(a)

(b)

Figure 4.10: Anaerobic digestion of mixed vegetable waste with digestate as inoculum at three different F/I ratios and temperature of 37°C.
(a) Biogas production (b) Biogas compositions

The results achieved in this study are compared with previous studies and summarized in Table 4.4. Gunaseelan (2004) investigated the digestion of separated vegetables as leaves of cabbage and cauliflower. In the study, F/I ratios were not mentioned. Nutrient and buffer solutions were also added into the reactors, but the concentrations are not shown. The methane yields were reported at 309 and 190 (average 250) L/g VS for cabbage and cauliflower, respectively. In the current study, the mixed feedstock also mainly included leaves of cabbage. At an F/I of 0.5 and 1.0 at all $NaHCO_3$ concentrations, the average methane yields were higher than that of Gunaseelan's study. Another study by Labatut et al. (2011) showed a methane yield of 257 mL/g VS from whole cabbage waste digested at an F/I ratio of 1.0. In the current study, at the same F/I ratios, the methane yields are higher. However, the digestion time in the experiment of Labatut et al. (2011) was 40 days shorter than the current study. This can be explained due to the different feedstocks. Cabbage was used in their study while mixed vegetable waste was used in our work. Rajeshwari et al. (1998) and Bouallagui et al. (2001) used mixed fruit and vegetable waste as the feedstock. Cumulative methane yields and VS reduction of our study were comparable with their results (Cho et al., 1995; Buffiere et al., 2006).

The achieved results also show good agreement of the biogas yields and VS reductions. As shown in Table 4.4, the biogas yields are proportional to VS reductions in all reactors, and at all buffer solution concentrations. According to Richards et al. (1991), the biogas mass removal should be higher than the VS reduction. The mass of biogas removed includes both converted substrate mass and the mass of water. In the current study, the theory is correct at an F/I of 0.5 with all buffer concentrations (including without buffer). However, at a higher F/I of 1.0 and 2.0, the VS reduction is higher than the biogas mass removal. This error may be due to the excessive loss of VFAs and other volatile compounds during the drying process of VS measurement (Liu et al., 2009).

The results of the BMP test are valuable to understand the AD process of vegetable waste at the Talaad Thai market, Thailand. It can be concluded that without adding an external alkalinity source, the optimum F/I ratios of the AD was at 0.5 g VS/g VS. When external alkalinity was added into reactors, the optimum conditions were at F/I of 1.0, $NaHCO_3$ concentration of 500 mg and 37°C. The conditions were applied in the lab-scale reactor in the next experiment.

Table 4.4: Results of BMP tests of mixed vegetable wastes at different F/Is and buffer concentration in the current and previous studies

Current Study(a)									Previous studies			
Buffer concentration (mg $NaHCO_3$/g VS)	F/I ratio	Time (days)	CH_4 content (%)	Cumulative biogas yield (mL/g VS)	Cumulative CH_4 yield (mL/g VS)	VS reduction (%)	Biogas mass removal (g/g VS added)	Final pH	Digestion time	Cumulative CH_4 yield (mL/g VS)	VS reduction (%)	Reference
No adding (37ºC)	0.5	70	52.3	855	306	42	0.96	8.5	40	257(b)	-	(Labatut et al., 2011)
	0.75	30	7.7	107	11	21	0.23	5.7				
	1.0	53	0	84	0	22	0.14	5.5				
	1.5	41	1.0	112	0	25	0.19	5.4				
	2.0	53	0.7	107	0	23	0.18	5.7	100	309(c) 291(d) 190(e) 331(f)	-	(Gunaseelan, 2004)
	3.0	53	0	97	0	20	0.17	5.5				
	4.0	53	0	102	0	18	0.17	5.3				
	5.0	53	0	110	0	19	0.19	5.4				
No adding (ambient 30±4ºC)	0.5	70	42.1	664	230	34	0.82	8.3				
	0.75	42	8.9	145	13	22	0.23	5.8				
	1.0	41	0	99	0	21	0.17	5.5	30	277(b)	-	(Cho et al., 1995)
	1.5	53	0	153	0	25	0.17	5.4				
	2.0	50	0	109	0	23	0.18	5.6				
	3.0	62	0	97	0	20	0.16	5.5				
	4.0	62	0	109	0	21	0.18	5.3				
	5.0	62	0	124	0	22	0.21	5.3				
300	0.5	75	61.5	688	293	43	0.71	8.3	-	160(g)	65	(Rajeshwari et al., 1998)
	1.0	130	64.1	839	359	64	0.43	8.2				

Current Study(a)									Previous studies			
Buffer concentration (mg NaHCO$_3$/g VS)	F/I ratio	Time (days)	CH$_4$ content (%)	Cumulative biogas yield (mL/g VS)	Cumulative CH$_4$ yield (mL/g VS)	VS reduction (%)	Biogas mass removal (g/g VS added)	Final pH	Digestion time	Cumulative CH$_4$ yield (mL/g VS)	VS reduction (%)	Reference
	2.0	105	22.2	465	10	45	0.28	6.9				
500	0.5	75	65.4	907	371	63	0.49	8.0				
	1.0	130	67.3	672	343	53	0.64	8.0				
	2.0	125	68.2	527	245	51	0.52	8.1	-	260(g)	68	(Bouallagui et al., 2001)
600	0.5	75	62.0	767	327	75	0.77	8.3				
	1.0	130	67.3	671	280	68	0.64	8.2				
	2.0	125	71.0	447	187	70	0.41	8.4				
900	0.5	75	55.0	626	314	63	0.68	8.4				
	1.0	130	64.7	706	361	71	0.69	8.4				
	2.0	125	64.9	589	278	73	0.57	8.2				
1200	0.5	100	47.4	643	265	65	0.76	8.5	-	294(h)	-	(Buffiere et al., 2006)
	1.0	125	64.2	713	343	62	0.71	8.6				
	2.0	125	65.1	549	273	66	0.54	8.6				
Recycled digestate and 150 mg	0.5	95	70.3	737	385	70	0.69	7.3				
	1.0	95	68.9	648	338	61	0.61	7.5				
	2.0	95	61.7	404	181	58	0.21	6.9				

(a) Mixed vegetable waste was used as the feedstock in the current study. Different types of feedstocks were used in previous studies, such as whole cabbage (b); leaves and stems of cabbage (c), and (d); leaves and stems of cauliflower (e) and (f); fruit and vegetable (g), lettuce (h).

4.7 Digestion of the mixed vegetable waste in lab-scale BIOCEL reactor

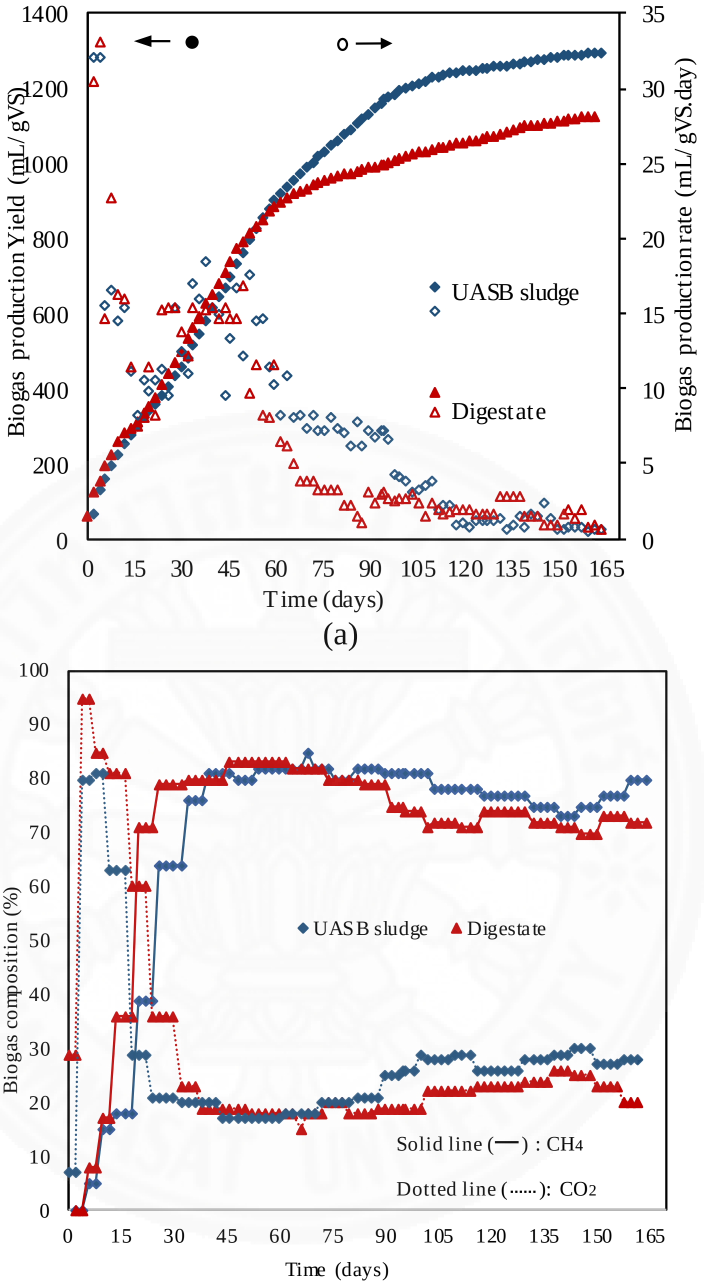
4.7.1 Biogas production and methane contents

In this experiment, the mixed vegetable wastes with two inoculum sources as UASB sludge and reused digestate was investigated. The optimum conditions achieved in the BMP tests were applied for the lab-scale BIOCEL reactors. All reactors were fed with F/I ratio of 1.0, $NaHCO_3$ concentration was at 500 and 150 mg for UASB sludge and digestate reactors, respectively. Performance in anaerobic digestion of lab-scale reactor with the UASB sludge and digestate is shown in Fig. 4.11 (a), (b). Different from the BMP tests, biogas production yield in the experiment includes the biogas generated from inoculums (i.e. UASB sludge and digestate). Therefore, the biogas production yield of the reactors was higher than that of BMP tests at the same F/I ratio and $NaHCO_3$ concentrations.

At the same conditions, for the UASB sludge, the biogas production yield was 907 and 1028 mL/g VS for the BMP test and lab-scale reactor, respectively, after 75 days of experiment. For the digestate, the biogas yield was 737 mL and 998 mL/g VS for the BMP test and lab-scale reactor, respectively, after 95 days of experiment time.

As presented in the Fig. 4.11, the biogas yield from the reactor with UASB sludge was higher than that value of the reactor used digestate as inoculum. The reason is that biogas presented in digestate was depleted in the last digestion while the gas still remains in UASB sludge. Therefore, biogas contributes from the UASB sludge is higher than digestate.

Average methane contents of reactors using UASB sludge as inoculum for BMP test and lab-scale were 65 and 71%, respectively. While average methane contents of reused digestate were 70 and 72% for BMP test and lab-scale reactor, respectively. After 165 days of experiment, the methane content of digestate reactor was higher than UASB sludge reactor. This was due to the acclimation of the AD bacteria to the vegetable wastes as reuse of digestate.



(a)

(b)

Figure 4.11: Anaerobic digestion of mixed vegetable waste in BIOCEL reactor at temperature of 37°C. (a) Biogas production (b) Biogas compositions

4.7.2 pH and Alkalinity

The variation of pH in UASB sludge and digestate reactors over 165 days of experiment is shown in Fig. 4.12. The pH of UASB sludge reactor was in a range of 6.2 to 8.0, while that of digestate reactor was 5.9 to 7.2. At the beginning of experiment, pH of UASB sludge reactor was higher than digestate reactor. The reason is that the UASB sludge reactor was operated at $NaHCO_3$ concentration of 500 mg/g VS, while that value of digestate reactor was only 150 mg/g VS. In the first two weeks of the experiment, pH of two reactors decreased rapidly. The pH of two reactors reached the lowest values of 6.2 and 5.9 on the 15th day of experiment. This was due to the high concentration of VFAs produced (Fig. 4.14). The concentration of VFA and alkalinity together decreased with the time in the two reactors and the pH curves were quite stable. The alkalinity was also found lower and reached to 5013 and 4332 mg/L $CaCO_3$ with UASB sludge and digestate reactor, respectively, during this period.

It depicted the important role of alkalinity to stable the pH of the AD process. If the external buffer solution was not added into the reactor, pH and alkalinity of the AD would be lower, and inhibit activity of AD microorganisms. This was in line with the conclusion of Mata-Alvarez (2002), pH value is strongly affected by the buffer capacity of the system and bicarbonate, VFA, and ammonia are the main process controllers in terms of buffering capacity. Due to lower alkalinity and pH, the methanogenic activity was not initialized and the methane contents of the two reactors were below 50% (Fig .4.11) during 0-15 days of the experiment. From day 25, the pH and alkalinity were almost found steady.

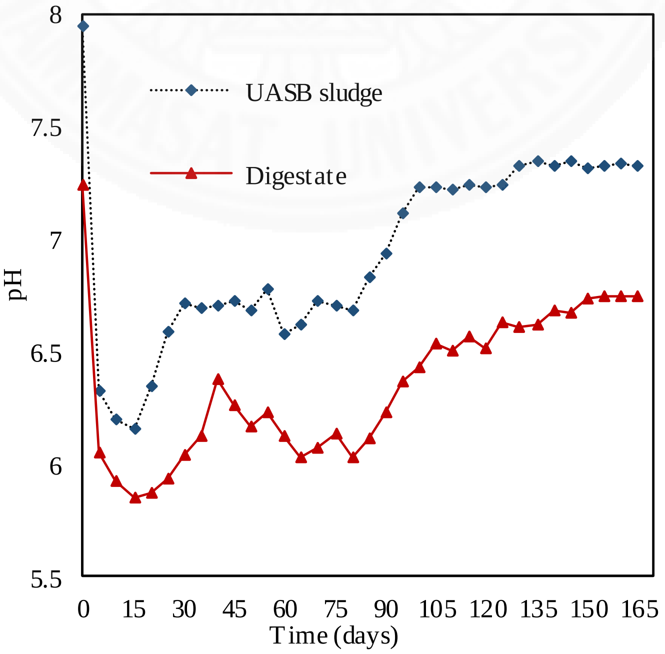


Figure 4.12: Variation of pH with experiment time in BIOCEL reactor with two types of inoculum.

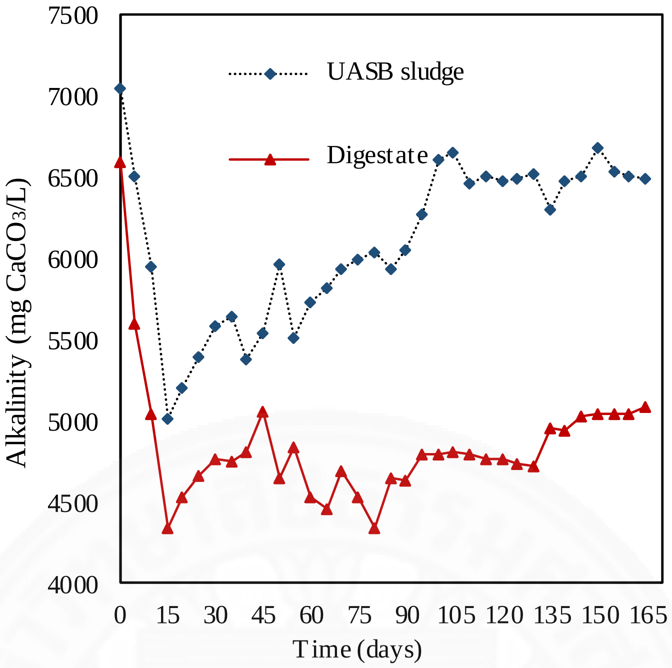
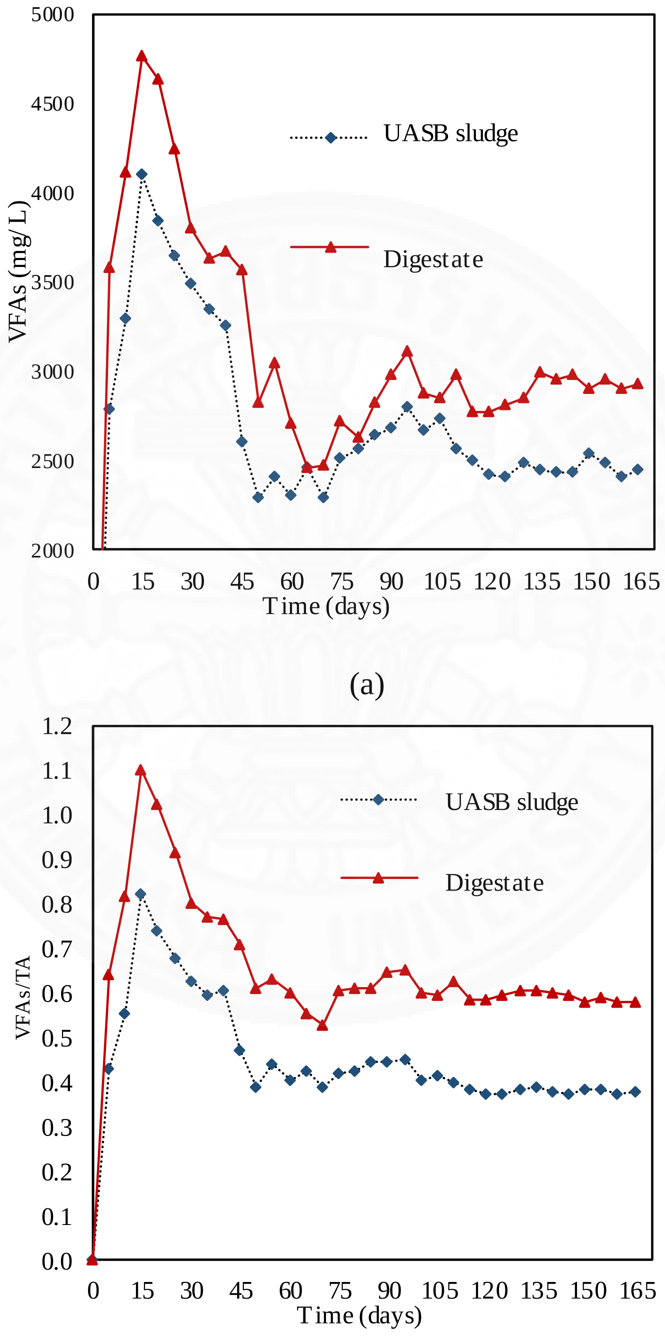


Figure 4.13: Variation of total alkalinity with experiment time in BIOCEL reactor with two types of inoculum.

Comparing two reactors, during 165 days of the experiment, pH and alkalinity of UASB sludge reactor were higher than that of digestate reactor. As shown in Fig. 4.13, VFAs accumulated in high concentration at the digestate reactor due to the contribution of VFAs in the digestate sludge from the previous digestion. Thus, the alkalinity of the digestate reactor was consumed rapidly.

4.7.3 VFA

Figure 4.14 shows the trend of VFAs concentrations of two reactors used UASB sludge and digestate as inoculum. Both reactors showed an initial stage with hydrolytic and acidogenic activity among 0 - 45 days. As the result, fatty acids were generated in high concentration. The VFA generation in the beginning was high due to higher acidogenesis and lower methanogenic activity. As the vegetable wastes consist of highly putrescible fraction, they were degraded quickly and the concentration of VFA was found elevated. The VFA production reached the peak values of 4185 and 4639 mg/L at the UASB sludge and digestate sludge, respectively, on the 15th day. This made the pH and alkalinity of the two reactors decreased rapidly. After day 30, the VFA concentration decreased due to methanogenic activity in which the intermediate organic acids started to convert into biogas such as methane and carbon dioxide. The VFA concentration reached around 2500 mg/L after day 30 of the experiment. The pH dropped in the beginning, corresponding to the transient accumulation of volatile acids, but then increased as the VFAs were converted to methane.



(a)

(b)

Figure 4.14: Variation of VFAs concentration (a) and VFAs/TA (b) with experiment time in BIOCEL reactor with two types of inoculum.

The ratio of VFAs and total alkalinity (VFAs/TA) is an important parameter to show the stability of AD process. According to Callaghana et al (2002), as the VFAs/TA is less than 0.8, the process is consider stable. At the digestate reactor of the study, the ratio reached to 1.1 on the 15th day of the experiment. However, the AD process was still continuous due to the high buffer ability and the acclimation of bacteria as reuse digestate. Thus, it is very important to measure the VFAs/TA ratio as operating an AD reactor. When the ratio is increase rapidly, it is necessary to adding more buffer solution or adjust the pH of the reactor.

As shown in figure 4.11, UASB sludge reactor achieved higher biogas production yields after 165 days of experiment time compared to the digestate reactor. The average methane content was around 70% for both reactors. Therefore, the cumulative methane yield of UASB sludge was higher than that of digestate reactor. It can be concluded that AD performance of digestate reactor was lower than that of UASB reactor. This is because the alkalinity of digestate reactor was not enough to maintain the optimum environment for AD microorganisms. It can be seen from the figure 4.14 (b), the VFAs/TA ratios of digestate reactor was higher than that values of UASB sludge. The VFAs/TA should be maintained below 0.4 to keep stability for the AD process (USEPA, 1976). Callaghana et al (2002) concluded that if the ratio is higher than 0.8, the AD digester depicted significant instability. As shown in Fig. 4.14 (b), during 165 days of experiment, the VFAs/TA ratio of UASB sludge was always lower than 0.8. However, the ratios at digestate reactor was higher than 0.8 during the beginning of experiment from day 10 to 30. Then the ratio was more stable after day 35, but it was still around 0.6. Therefore, as operated a full-scale reactor, it is very important to analysis the VFAs/TA ratios during the experiment time in AD reactors. When the ratio is higher than 0.5, it is necessary to add more buffer solution as $NaHCO_3$.

At the sample ratios and conditions, anaerobic performance of the lab-scale reactors was higher than that of BMP reactors. For the reactor with UASB sludge, the methane yield was 404 and 756 mL/g VS at BMP and lab-scale reactor, respectively. At the reactor with digestate, the methane yield was 395 and 672 mL/g VS at BMP and lab-scale reactor, respectively.

The results achieved in this study are higher than previous studies and summarized in Table 4.5. Bouallagui et al. (2001) investigated the digestion of fruit and vegetable waste. In the study, the feedstock was digested in the BIOCEL reactor (5 L) similar to the current study. After 32 days of experiment, the methane yield was 260 mL/g VS with VS reduction of 65%. Another study of Rajeshwari et al. (1998) also determined the methane production yield of fruit and vegetable waste. The reactor was designed at 10 liters. After 47 days of experiment, methane yields were reported at 160 ml/g VS with VS reduction of 58%. The both studies gave lower results in methane yields and VS reductions comparing to the current study. Le et al. (2013) used organic fraction of municipal solid waste (OFMSW) as feedstock and digested OFMSW as inoculum. In the study, two F/I ratios at 5 and 10, were investigated. The cumulative methane yields were 87 and 113 mL/g VS at F/I ratios of 5 and 10, respectively, after 32 days of digestion. Cumulative methane yields of our study were higher with their results. The methane yeilds of the conditions are presented in Fig 2 (Appendix B).

Table 4.5: Results of lab-scale reactors of mixed vegetable wastes

Current Study						Previous Studies	
Inoculum type	CH_4 content (%)	Cumulative biogas yield (mL/g VS)	Cumulative CH_4 yield (mL/g VS)	VS reduction (%)	Final pH	Cumulative CH_4 yield (mL/g VS)	Reference
UASB sludge	71	1297	756	69	7.3	87	(Le Thi et al., 2013)
						160	(Rajeshwari et al., 1998)
Digestate	72	1128	672	72	6.8	260	(Bouallagui et al., 2001)
						387	(Velmurugan, 2011)

Chapter 5
Conclusions and Recommendations

5.1 Conclusions

This study successfully demonstrated the potential of utilizing the mixed vegetable waste from Talaad Thai market as a renewable energy source through anaerobic digestion. The optimum conditions such as F/I ratios, temperature, external alkalinity concentration was determined by the BMP tests in the study. Moreover, the optimum conditions from the BMP tests were applied in the lab-scale rector. The lab scale reactor helped to confirm the results of BMP tests and have modifications when a real scale AD is built in the field. A summary of the results achieved in this study is shown as below.

- Without adding $NaHCO_3$
  - The achieved results showed a negative relation between biogas yields and F/I ratios at ambient (30±C) and constant temperature (37°C). The higher the F/I ratio, the lower the biogas and methane yield. This was because the high concentration of VFAs accumulated at the F/I ratio higher than 0.5 and inhibited the activity of anaerobic microorganisms.
  - Methanogens in the study preferred temperature of 37°C compared with ambient temperature. Thus, the biogas and methane production at 37°C was generally higher than that of the ambient temperature (30±4°C) at all F/I ratios except F/I of 4.0 and 5.0 g VS/g VS.
  - The optimum ratio was at F/I of 0.5 and temperature of 37°C. The biogas and methane yield were at 851 and 306 mL/g VS when no external buffer was added.
  - At F/I ratios of 2.0, 3.0, 4.0, and 5.0 g VS/ g VS, VFAs was generated in high concentration and inhibited the activity of methanogens. This made the anaerobic digestion stopped at the acidogenesis step and hydrogen could not transfer to methane. Therefore, high amounts of

hydrogen were found in the mixture of biogas at the F/I ratios for both temperature.

- Adding $NaHCO_3$
  - When the external buffer was added into reactors at F/I of 1.0 and 2.0, the biogas yields and methane contents were improved significantly. The external alkalinity helped to maintain pH of environment in the neutral range. Therefore, anaerobic microorganism could be active and digest the waste during the experiment time.
  - At F/I of 1.0, the biogas yield was reached the highest value at the buffer concentration of 300 mg. While the F/I of 2.0 achieved its highest values of biogas and methane yields at the buffer concentration of 900 mg. The maximum biogas yields were 839 and 589 mL/g VS at F/I of 1.0 and 2.0 with the external alkalinity concentration, respectively
  - When considering the cumulative biogas, methane yields, and retention time of the AD, the optimum conditions were at $NaHCO_3$ concentration of 500 mg for all F/I ratios of 0.5, 1.0 and 2.0.
  - Due to high value of alkalinity of the digestate from the previous digestion, it can be reused as a source of inoculum and buffer solution. When the digestate is recycled, the sodium bicarbonate can be reduced at the concentration of 150 mg per gram VS added.

- BIOCEL reactor
  - The reactor with UASB sludge achieved higher biogas production yields after 165 days of experiment time compared to the reactor with digestate. This was due to the lack of alkalinity in the reactor with digestate at the hydrolytic step of the AD.

- The experiment also depicted the important role of VFAs/TA ratio to maintain the stability of the AD process. It is necessary to measure the parameters during the experiment and operate the ratio below 0.5.

Overall, the BIOCEL reactors showed the higher AD performance compared to BMP reactors. For the reused digestate, the optimum conditions were similar but the $NaHCO_3$ can be decreased to 150 mg. However, other types of AD reactor should be investigated to find out the optimum one. Here, some recommendations are suggested, which require deeper investigations.

5.2 Recommendations

- More combinations of organic wastes consisting of other sources of substrates should be investigated. For example, solid wastes from another market such as fruits, flowers, meat, fish, and etc. should be mixed together to improve the C/N ratio.
- Another AD technologies such as DRANCO, KOMPOGAS, SEBAC should be investigated to determine the optimum one for the digestion of the mixed vegetable waste.
- For the effluents from the lab-scale reactors, it should have further experiments to utilize the digestate. Use of the digestate should be investigated as a feedstock for the composting process to make a green fertilizer.

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Appendices

Appendix A

Photographs

Preparation for feedstock and inoculum

Raw vegetable waste

Cut into size 20 – 30 mm

Grind into size 2 – 3 mm

Mix with AD sludge

Feed into reactors and shake to mix
feedstock and inoculum well

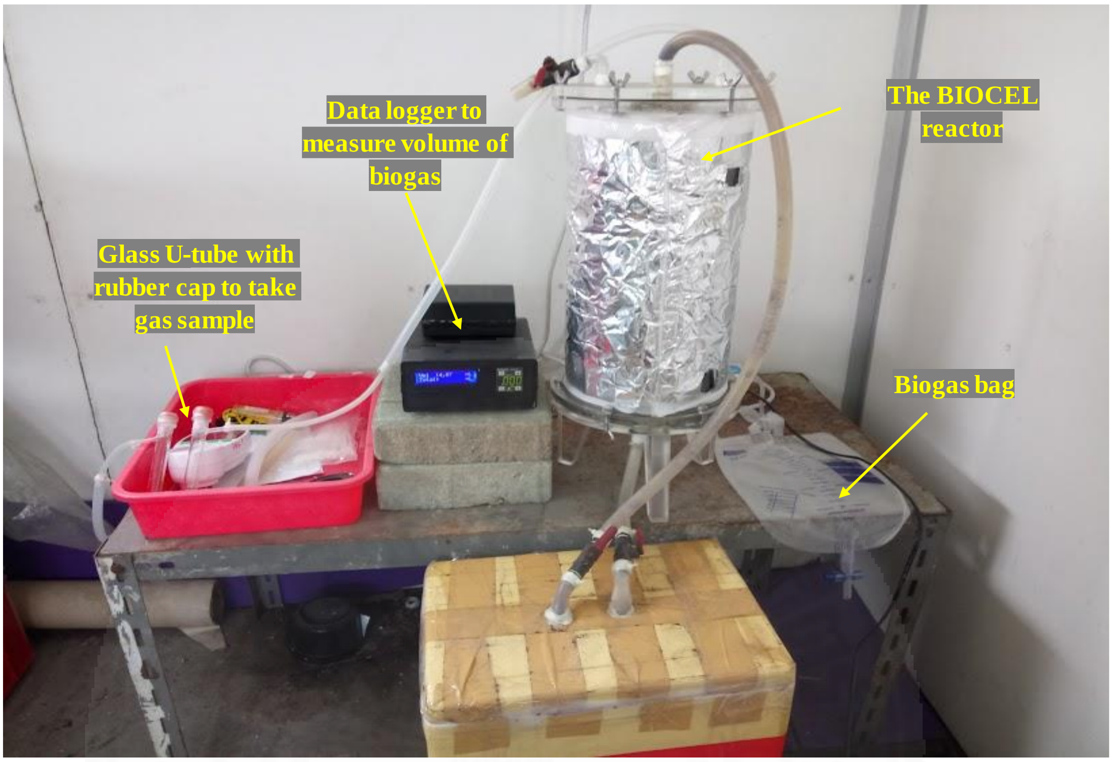
Determine the influences of temperature

Wrap with aluminum foil to
avoid photosynthetic bacteria

Incubator 37ºC

Ambient condition 30 ± 4˚C

BIOCEL system



Appendix B

Methane production yields

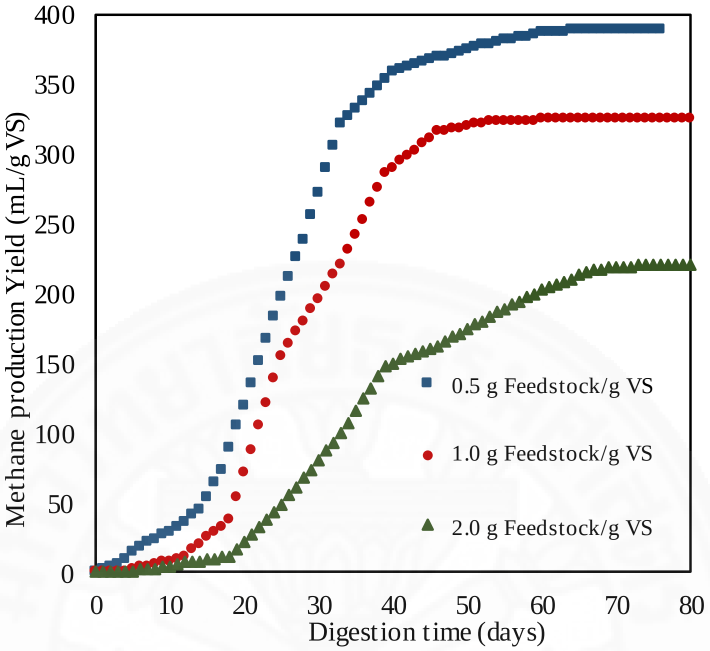


Fig. 1: Methane yield at optimum conditions of the BMP test with UASB sludge (i.e. $NaHCO_3$ concentration of 500 mg, temperature of 37°C)

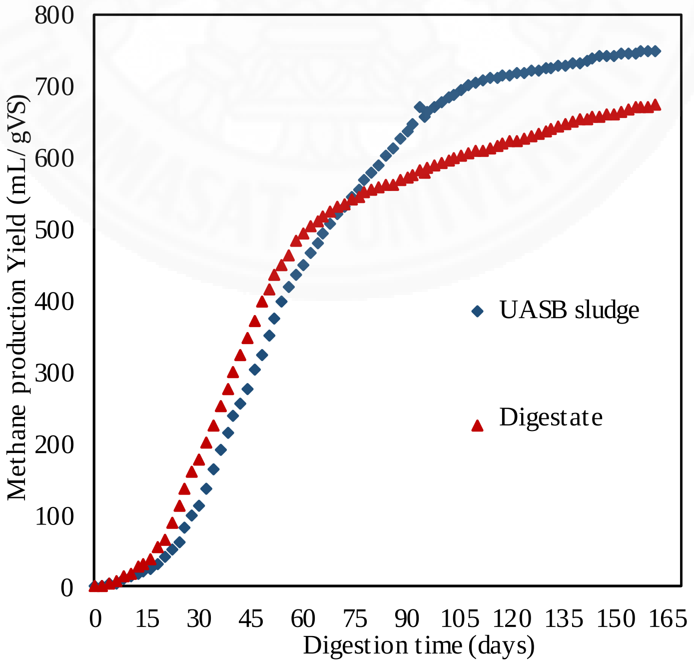


Fig. 2: Methane yield at lab-scale reactors (i.e. reactor with UASB sludge and digestate)