การผลิตก๊าซชีวภาพแบบการหมักร่วมของขยะเศษอาหารและเศษใบจามจุรี โดยใช้ถังหมักไร้อากาศแบบขั้นตอนเดียวและแบบสองขั้นตอน

นางสาวพรรณเลขา หมั่นเพ็ชร

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรมหาบัณฑิต
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บัณฑิตวิทยาลัย จุฬาลงกรณ์มหาวิทยาลัย
ปีการศึกษา 2557

BIOGAS PRODUCTION FROM CO-DIGESTION OF FOOD WASTE
AND RAIN TREE LEAF USING SINGLE-STAGE AND
TWO-STAGE ANAEROBIC DIGESTERS

Miss Panlekha Manpetch

A Thesis Submitted in Partial Fulfillment of the Requirements
for the Degree of Master of Science Program in Environmental Science
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Thesis Title	BIOGAS PRODUCTION FROM CO-DIGESTION OF FOOD WASTE AND RAIN TREE LEAF USING SINGLE-STAGE AND TWO-STAGE ANAEROBIC DIGESTERS
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Field of Study	Environmental Science
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พรรณเลขา หมั่นเพ็ชร : การผลิตก๊าซชีวภาพแบบการหมักร่วมของขยะเศษอาหารและเศษใบจามจุรีโดยใช้ถังหมักไร้อากาศแบบขั้นตอนเดียวและแบบสองขั้นตอน (BIOGAS PRODUCTION FROM CO-DIGESTION OF FOOD WASTE AND RAIN TREE LEAF USING SINGLE-STAGE AND TWO-STAGE ANAEROBIC DIGESTERS) อ.ที่ปรึกษาวิทยานิพนธ์หลัก: รศ. ดร. ชวลิต รัตนธรรมสกุล, 89 หน้า.

การวิจัยนี้มีวัตถุประสงค์เพื่อศึกษาผลของระบบถังหมัก แบบขั้นตอนเดียวและแบบสองขั้นตอนต่อการผลิตก๊าซชีวภาพและการบำบัดของเสีย โดยทำการหมักร่วมระหว่างเศษอาหารและเศษใบจามจุรี ที่อัตราส่วนต่างๆ ดังนี้ 85:15 90:10 95:5 100:0 ถังหมักก๊าซชีวภาพต้นแบบระบบขั้นตอนเดียว มีรูปทรงกระบอกแนวนอน ภายในมีใบพัดกวน ตั้งอยู่บริเวณโรงอาหารอาคารจุลจักรพงษ์ และระบบถังหมักก๊าซชีวภาพต้นแบบระบบสองขั้นตอน คือจะแยกหมักระหว่างถังหมักกรด และถังหมักก๊าซ เพื่อเพิ่มประสิทธิภาพการทำงานของเชื้อจุลินทรีย์ ตั้งอยู่บริเวณโรงอาหารอาคารมหิตลาธิเบศร ซึ่งในงานวิจัยแบ่งผลการทดลองออกเป็น 2 การเปรียบเทียบ คือเปรียบเทียบระหว่างระบบถังหมักขั้นตอนเดียวและแบบสองขั้นตอน และการเปรียบเทียบอัตราส่วนที่เหมาะสมของการหมักร่วมระหว่างเศษอาหารและเศษใบจามจุรีต่อการผลิตก๊าซชีวภาพ

ผลการทดลองในส่วนของการเตรียมอัตราส่วนของการหมักร่วมเศษอาหารและเศษใบจามจุรีพบว่า ที่ 95:5 ให้ผลการผลิตก๊าซชีวภาพที่สูงในทั้งสองระบบ วัดค่าคาร์บอนต่อไนโตรเจนอยู่ในช่วง 20.77 มีความเหมาะสมต่อการเจริญเติบโตของจุลินทรีย์ในระบบถังหมัก ในส่วนของประสิทธิภาพการกำจัดซีโอดีพบว่า ระบบถังหมักก๊าซชีวภาพแบบขั้นตอนเดียวสามารถกำจัดซีโอดีได้ร้อยละ 61.95–89.44 แต่ระบบถังหมักก๊าซชีวภาพแบบสองขั้นตอนนั้นสามารถกำจัดซีโอดีได้ร้อยละ 71.90 – 88.84 และการกำจัดของแข็งทั้งหมดพบว่าทั้งในระบบขั้นตอนเดียวและสองขั้นตอนสามารถกำจัดของแข็งได้ร้อยละ 73.54 - 81.70 และ 59.03- 83.10 ตามลำดับ ผลการทดลองพบกว่าการแยกถังหมักออกเป็นสองขั้นตอนสามารถเพิ่มประสิทธิภาพการกำจัดซีโอดีและของแข็งทั้งหมดได้ดียิ่งขึ้น ในส่วนของการผลิตก๊าซชีวภาพ พบว่าระบบถังหมักแบบสองขั้นตอนสามารถผลิตก๊าซชีวภาพได้สูงถึง 4.213 ลบ.ม.มาตรฐาน และ 1.965 ลบ.ม.มาตรฐาน โดยมีค่าเฉลี่ย 3.194 ลบ.ม.มาตรฐาน ในส่วนของระบบถังหมักก๊าซชีวภาพแบบขั้นตอนเดียวพบว่าได้ค่าเฉลี่ยอยู่ที่ 1.612 ลบ.ม.มาตรฐาน

สาขาวิชา วิทยาศาสตร์สิ่งแวดล้อม	ลายมือชื่อนิสิต ....
ปีการศึกษา 2557	ลายมือชื่อ อ.ที่ปรึกษาหลัก ....

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PANLEKHA MANPETCH: BIOGAS PRODUCTION FROM CO-DIGESTION OF FOOD WASTE AND RAIN TREE LEAF USING SINGLE-STAGE AND TWO-STAGE ANAEROBIC DIGESTERS. ADVISOR: ASSOC. PROF. CHAVALIT RATANATAMSKUL, Ph.D., 89 pp.

This research is aim to study the anaerobic digestion system of single-stage and two-stage anaerobic digester for biogas production and waste treatment. This research also study the possibility of co-digestion of food waste and rain tree leaf as a feed stock. The ratio of food waste and rain tree leaf was done in 85:15 90:10 95:5 and 100:0 respectively. Single-stage anaerobic digester has a horizontal cylinder shape located at the canteen of Chulachakrabongse building while two-stage anaerobic digester separated into two reactors. acid fermentation reactor and methane reactor. Two-stage anaerobic digester located at the canteen of Mahitaladhibesra building.

The result obtain in feedstock was found that the ratio of co-digestion of food waste and rain tree leaf at 95:5 gave the significantly high in biogas production. Carbon to nitrogen ratio was measured, the result was 20.77 optimal C/N ratio for biogas production. The efficiency of COD removal found in single-stage was 61.95% – 89.44% while two-stage was 71.9% – 88.84% and the efficiency of total solid removal was found that ratio of co-digestion has significantly high in both single-stage and two-stage 73.54-81.7 and 59.03-83.10 respectively. The result found that separate reactor into two-stage gave more efficiency in waste reduction. Biogas production was found that two-stage anaerobic digestion had significantly high in biogas production rate at maximun 4.213 Nm^3 and 1.965 Nm^3 with average 3.194±0.189 Nm^3 in two-stage while single-stage anaerobic digester can produced biogas average 1.612 ± 0.094 Nm^3.

Field of Study: Environmental Science Student's Signature ....

Academic Year: 2014 Advisor's Signature ....

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CHAPTER I

INTRODUCTION

1.1 Introduction

The total amount of solid waste generated in Thailand has been rising up annually. In 2012, the amount of solid waste generation was reported approximately 67,577 tons/day of MSW (municipal solid waste) which consists of food waste as the largest waste stream 44.34% of total MSW (Pollution Control Department, 2012). Trend in energy demand in each year has also been rising up, lead to import of crude oil. Crude oil imported in 2012 was 1,079 KBD of crude oil equivalent, increasing by 8.3% of previous year. The imports value increased to 1,119.3 billion Baht, or an increase of 14.5% since the crude oil price was not much change from the previous year (The Energy Policy and Planning Office, 2013). Nowadays, the renewable energy has been focus to replace conventional fuels.

Anaerobic digestion is regarded as a clean energy technology (Abbasi & Abbasi, 2010; Ratanatamskul et al, 2014) that is able to convert energy directly from organic waste by microorganism. Nowadays, this technology has been promoted greatly in Thailand to produce biogas as the renewable energy. Rapid biodegradation of the organic fraction of the MSW is an importance key to identify environmental more responsible way to process it rather than landfilling or composting. Furthermore, anaerobic digestion is closed and controlled process and based on fugitive emissions is more preferable than landfilling and aerobic composting (Levis et al, 2010).

To enhance biogas production, anaerobic co-digested of different wastes has been focus to achieve synergetic effects. The benefits of anaerobic co-digestion can improve: the dilution of toxic compound, improved balance of nutrients, and increased load of biodegradable organic matter.(Sosnowski et al, 2003)

Anaerobic digestion process can be divided into two phases. The first one is acid fermentation phase where hydrolysis and acidogenesis is occurred lead to the production of intermediate products predominated by the volatile organic acids; Second phase is known as methane fermentation phase resulting in the conversion of intermediates substances to methane (Park et al, 2005; von Sachs et al, 2003). Almost all of anaerobic digesters are single-stage system where acidogenesis and methanogenesis both occur in the same digestive tank. Due to bacterial varieties different is required in anaerobic digestion. To separate reactor in two-stage digester makes each phase in the suitable environmental conditions (Cooney et al, 2007; Held et al, 2002).

In this study, co-digestion of food waste from canteens (Chulachakrabongse and Mahitaladhibesra buildings) and rain tree or Chamchuri leaf has been considered in various ratio to gain the suitable carbon to nitrogen ratio. Rain tree is one of the important Chulalongkorn University symbols. Rain tree leaf is found to be the major gardening wastes. Furthermore, single-stage and two-stage anaerobic digestion has also been studied to compare and evaluate the organic degradation and biogas production from co-digestion of food waste and rain tree leaf in both process to gain the optimal condition to produce the highest biogas production.

1.2 Objectives

1.2.1 To compare and evaluate organic degradation and biogas production of single-stage and two-stage anaerobic co-digestion of food waste and rain tree leaf

1.2.2 To study the effect of substrate ratio in co-digestion of food waste and rain tree leaf on system performance.

1.2.3 To study the biogas production quantity and quality from anaerobic co-digestion process.

1.2.4 To study characteristic of the liquid digestate

1.3 Scope of work

The study of single-stage and two-stage anaerobic co-digestion of food waste and rain tree leaf in various ratio has been done within a scope below.

1.3.1 This experiment was done in pilot scale a single-stage and a two-stage anaerobic digestion tanks. Single-stage anaerobic digestion tank locates next to Chulachakrabongse canteen and two-stage anaerobic digestion tank locates next to Mahitaladhibesra canteen. Feedstock of food waste was from both canteens and the rain tree leaf was from around Chulalongkorn University. Total feeding volume was around 20 kilogram per day.

1.3.2 This experiment was done to compare the single-stage and two-stage anaerobic digester system efficiency, and the ratio of feedstock food waste mixed with rain tree leaf to see the optimal condition. The results will be analyzed in 2 different comparatives.

1) Compare the efficiency of anaerobic systems of both single-stage and two-stage anaerobic digesters with all the feedstock ratio
2) Compare the optimal ratio of feed stock (food waste mixed with rain tree leaf) as below
    - 85% food waste with 15% rain tree leaf
    - 90% food waste with 10% rain tree leaf
    - 95% food waste with 5% rain tree leaf
    - 100% food waste without adding rain tree leaf

1.3.3 Analytical parameters

- Total chemical oxygen demand (COD)	- Total volatile fatty acid (TVF)
- Total solid (TS)	- Total volatile solid (TS)
- Temperature	- Pressure
- C/N ratio	- Alkalinity
- Biogas composition	- pH

1.4 Expected Benefits & Application

1. To reduce organic waste in the university both food waste and gardening waste.

2. To know that which anaerobic digestion system (single-stage or two-stage) give more efficiency in biogas production from co-digestion of food waste and rain tree leaf.

3. To know the optimal ratio of co-digestion of food waste and rain tree leaf in biogas production.

4. To support biogas to university canteen as application of recover energy from food wastes and rain tree leaf

CHAPTER II

LITERATURE REVIEW

2.1 Fuel Energy

Trend in energy demand in each year has been rising up annually. Crude oil imported in 2012 was 1,079 KBD of crude oil equivalent, increasing by 8.3% of previous year. The imports value increased to 1,119.3 billion Baht, or an increase of 14.5% since the crude oil price was not much change from the previous year (The Energy Policy and Planning Office, 2013). Nowadays, the renewable energy has been focus to replace conventional fuels.

2.1.1 Energy from biomass

The term "biomass" refers to organic matter that has stored energy through the process of photosynthesis. It exists in one form as plants and may be transferred through the food chain to animals' bodies and their wastes, all of which can be converted for everyday human use through processes such as combustion. Many of the biomass fuels used today come in the form of wood products, dried vegetation, crop residues, and aquatic plants. Biomass has become one of the most commonly used renewable sources of energy. There are 2 types of conversion.

2.1.1.1 Biochemical conversion

Biochemical conversion of biomass involves bacteria, microorganisms and enzymes activities to breakdown biomass into gaseous or liquid fuels, such as biogas or bioethanol. The most popular biochemical technologies are anaerobic digestion and fermentation. Anaerobic digestion is a chemical reactions during organic material is decomposed through the metabolic pathways of naturally occurring microorganisms in an anaerobic environment. Biomass wastes can also yield liquid fuels, such as cellulosic ethanol, which can be used to replace petroleum-based fuels.

2.1.1.2 Thermal Conversion

These technologies can be classified according to the principal energy carrier produced in the conversion process. Carriers are in the form of heat, gas, liquid and solid products, depending on the extent to which oxygen in air is admitted to the conversion process. The major methods of thermal conversion are combustion in excess air, gasification in reduced air, and pyrolysis in the absence of air.

Conventional combustion technologies raise steam through the combustion of biomass. This steam may then be expanded through a conventional turbo-alternator to produce electricity.

Gasification of biomass takes place in a restricted supply of oxygen and occurs through initial devolatilization of the biomass, combustion of the volatile material and char, and further reduction to produce a fuel gas rich in carbon monoxide and hydrogen.

Pyrolysis is the term given to the thermal degradation of wood in the absence of oxygen. It enables biomass to be converted to a combination of solid char, gas and a liquid bio-oil. Pyrolysis technologies are generally categorized as fast or slow according to the time taken for processing the feed into pyrolysis products.

2.1.1.3 Advantages and disadvantages of biomass energy

Adventages : - Abundant and renewable: biomass products are abundant and renewable. Since they come from living sources, and life is cyclical, these products potentially never run out, it turns living things components and waste products into energy.

- Reduce dependency on fossil fuels: as developed an alternate source of fuel. This can help to reduce dependency on fossil fuels

- Reduce landfills: an anaerobic digestion technology is handle biological waste, which can reduce the landfill volume required and control the organic pollutants as a source reduction method (Kim et al, 2006)

Disadvantages:- Inefficient as compared to fossil fuels: as the biomass from crop cultivation depends on season. The productivity is uncertain and sometime not enough. And also the changing of cultivation land into city.

- Expensive: some technologies to recover energy from wastes are complicated and expensive in investment.

2.2 Municipal solid waste

Municipal solid waste (MSW) is a term usually applied to a heterogeneous collection of wastes produced. Two broad categories are fermentable, and non-fermentable. Fermentative wastes tend to decompose rapidly and unless carefully controlled, decompose with the production of objectionable odors and visual unpleasantness. Non-fermentable wastes tend to resist decomposition and, therefore, break down very slowly. A major source of fermentative waste is food preparation and consumption a typified by crop and market debris.

The Pollution Control Department (2007) made an attempt to present a general qualitative and quantitative description of organic waste generation in Thailand. It is estimated that its total amount reaches about 50,000 tons a year and it is 60% of municipal solid waste generated within the country. The whole mass of organic biodegradable waste is dominated by the waste originating from food residues, which amounts to about 70 %. Since food waste contains high moisture content and also high organic composition, it can contribute to odor problem, leachate, and greenhouse gas production in landfill (Zhang et al, 2007).

2.2.1 Situation of municipal solid waste in Thailand

In 2012, Thailand approximately generated 24.73 million tons of municipal solid waste or 67,577 tons/day. Of the total waste, 15.90 million tons were disposed of in waste containers by local residents. Local Administrative Organizations were able to collect 11.90 million tons of waste. About 5.83 million tons could be properly managed and a total of 5.28 million tons were utilized. The remaining 13.62 million tons were improperly disposal. (Pollution Control Department, 2012)

Table 2.1 Amount of Solid Waste Generated Per Day in 2012

Area	Amount of Waste (Tons/Day)	
	2011	2012
Bangkok Metropolitan Administration	11,470	11,000
Pattaya City	425	426
Municipalities (2,266 locations)	19,011	25,046
Subdistrict Administrative Organizations (5,509 locations)	38,544	31,105
Total	69,450	67,577

Source : (Pollution Control Department, 2012)

2.2.2 Impacts of solid waste

2.2.3.1. Impacts of solid waste on health

The population in areas where there is no proper waste disposal method, especially children and adult, waste workers, and workers in facilities producing toxic and infectious material. Other high-risk group include population living close to a waste dump and those, whose water supply has become contaminated either due to waste dumping or leakage from landfill sites. Uncollected solid waste also increases risk of injury, and infection. Exposure to hazardous waste can affect human health, children being more vulnerable to these pollutants. In fact, direct exposure can lead to diseases through chemical exposure as the release of chemical waste into the environment leads to chemical poisoning.

2.2.3.2 Impacts of solid waste on environment

Fire and explosion hazard are not limited to incidents away from the landfill. On site fires are common and the formation of a mixture of methane and oxygen that can sustain a fire. Odors are mainly the result of the presence of small concentration of odorous constituents such as esters, hydrogen sulfide, volatile fatty acid, alkybenzenes etc. in landfill gas emitted into the atmosphere. (El-Fadel et al, 1997)

Ground water pollution, leachate occurrence is the most significant treat to ground water. Once it reaches the bottom of the landfill or an impermeable layer within the landfill, leachate either travels laterally to a point where it discharges to the ground's surface.

Air pollution, emission of methane and carbon dioxide from landfill surfaces contribute significantly to global warming or the greenhouse effect. It is more effective at trapping infrared radiation and tends to persist longer in the atmosphere.

2.2.3 Food waste

Food waste refers to food appropriate for human consumption being discarded, whether or not after it is kept beyond its expiry date or left to spoil. Often this is because food has spoiled but it can be for other reasons such as oversupply due to markets, or individual consumer shopping or eating habits. The whole mass of organic biodegradable waste is dominated by the waste originating from food residues, which amounts to about 70 %. Since food waste contains high moisture content and also high organic composition, it can contribute to odor problem, leachate, and greenhouse gas production in landfill (Zhang et al, 2007)

2.2.3.1 Causes of food waste

Causes of food waste are common to households and businesses. Food is lost or wasted along the whole food supply chain: on the farm, in processing and manufacture, in shops, in restaurants and canteens and in households.(European-Commission, 2010) Factors contributing to food waste include:

- Insufficient shopping and meal planning and promotions like "buy one get one free" leading to too much food being purchased or prepared

- Misunderstandings about the meaning of "best before" and "use by" date labels leading to edible foods being thrown away

- Standardized portion sizes in restaurants and canteens

- Stock management issues for manufacturers and retailers

- Overproduction or lack of demand for certain products at certain times of the year

- Product and packaging damage (farmers and food manufacturing)

- Inadequate storage and transport at all stages of the food chain

2.3 Anaerobic Digestion Process

Anaerobic digestion is regarded as a clean energy technology (Abbasi & Abbasi, 2010; Ratanatamskul et al, 2014) that is able to convert energy directly from organic waste by microorganism. Nowadays, this technology has been promoted greatly in Thailand to produce biogas as the renewable energy. Rapid biodegradation of the organic fraction of the MSW is of key importance to identify environmental more responsible way to process it rather than landfilling or composting it. Furthermore, anaerobic digestion is closed and controlled process and based on fugitive emissions is more preferable than landfilling and aerobic composting (Levis et al, 2010)

$$\text{Organic Matter} \xrightarrow{\text{Anaerobic digestion}} CH_4 + CO_2 + NH_3 + H_2 + H_2S$$

There are four fundamental steps of anaerobic digestion that include hydrolysis, acidogenesis, acetogenesis, and methanogenesis as shown in Figure 2.1. Throughout this entire process, large organic polymers that make up biomass are broken down into smaller molecules by chemicals and microorganisms. Upon completion of the anaerobic digestion process, the biomass is converted into biogas, mainly methane and carbon dioxide, as well as digestate and wastewater.

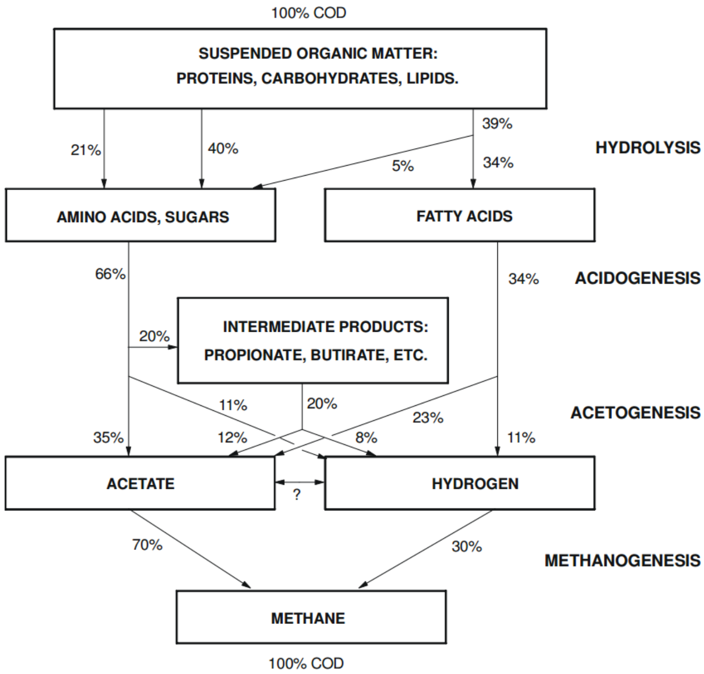


Figure 1.1 Anaerobic digestion process

Source : (W. Gujer, 1983)

2.3.1 Biochemical reactions in anaerobic digestion

2.3.1.1 Hydrolysis

Extracellular microbial enzymes catalyzing this reaction are known as hydrolyses. Depending on the type of the reaction they catalyze, these hydrolyses can be esterase (enzymes that hydrolyze ester bonds), glycosidase (enzymes that hydrolyze glycosides bonds), or peptidase (enzymes that hydrolyze peptide bonds). Hydrolytic bacteria. Proteolytic enzyme, cellulolytic enzyme, lipolytic enzyme.

$$\text{Carbohydrate} \xrightarrow{\text{hydrolysis}} \text{Sugar} \quad \text{Alcohol}$$

$$\text{Protein} \xrightarrow{\text{hydrolysis}} \text{Peptide} + \text{Amino acid}$$

$$\text{Lipid} \xrightarrow{\text{hydrolysis}} \text{Glycerol} \quad \text{Fatty acid}$$

2.3.1.2 Acidogenesis

Hydrolysed product was then digest to volatile fatty acid such as Acetic acid, Propionic acid, Butyric acid, Alcohol and Aldehyde. After anaerobic bacteria acidogenesis will drop pH down to 4. Below reactions show digestive of glucose to volatile fatty acid.

$$C_6H_{12}O_6 + 2H_2O \longrightarrow \underset{\text{(acetic acid)}}{2CH_3COOH} + 2CO_2 + 4H_2$$

$$C_6H_{12}O_6 + 2H_2 \longrightarrow \underset{\text{(propionic acid)}}{2CH_3CH_2COOH} + 2H_2O$$

$$2C_6H_{12}O_6 \longrightarrow \underset{\text{(butyric acid)}}{2CH_3CH_2CH_2COOH} + 4CO_2 + 4H_2$$

2.3.1.3 Acetogenesis

The products of the acidification are converted into acetic acids, hydrogen, and carbon dioxide by acetogenic bacteria. The first three steps of anaerobic digestion are often grouped together as acid fermentation. It is important to note that in the acid fermentation, no organic material is removed from the liquid phase: it is transformed into a form suitable as substrate for the subsequent process of methanogenesis.

$$\underset{\text{(ethanol)}}{CH_3CH_2OH} + H_2O \longrightarrow \underset{\text{(acetic acid)}}{CH_3COOH} + 2H_2$$

$$\underset{\text{(propionic acid)}}{CH_3CH_2COOH} + H_2O \longrightarrow \underset{\text{(acetic acid)}}{CH_3COOH} + 3H_2 + CO_2$$

$$CH_3CH_2CH_2COOH + 2H_2O \longrightarrow 2CH_3COOH + 2H_2$$

(butyric acid) (acetic acid)

2.3.1.4 Methanogenesis

In the final step of the anaerobic digestion process, the products of the acid fermentation (mainly acetic acid) are converted into CO_2 and CH_4. Only then will organic material be removed, as the produced methane gas will largely desorb from the liquid phase. The free energy released in the reactions is partially used for synthesis of the anaerobic bacterial populations.

Methane-forming bacteria can be divided into two types. 70% of methane is use acetic acid as substrate to produce methane and carbon dioxide by methane forming bacteria. The other methane is from the reaction between carbon dioxide and hydrogen to produce methane and water by hydrogen utilizing methane bacteria as below equation.

$CH_3COOH \longrightarrow CH_4 + CO_2$ (Methane Forming Bacteria)

$CO_2 + 4H_2 \longrightarrow CH_4 + 2H_2O$ (Hydrogen Utilizing Methane Bacteria)

2.3.2. The Microbiology of anaerobic digesters

Bacteria that are commonly found in wastewater treatment processes are divided into groups according to 1) their response to free molecular oxygen and 2) their enzymatic ability to degrade substrate. In the anaerobic digester can be divided into 2 groups by their enzymatic ability which are Acid-forming bacteria and Methane forming bacteria (Gerardi, 2003)

2.3.2.1 Acid-forming bacteria

During the degradation of wastes of acid forming bacteria within an anaerobic digester, facultative anaerobic and some strictly anaerobic bacteria are involved. These bacteria can be divided into 2 groups.

1) Hydrolytic and Fermentative bacteria

Hydrolytic bacteria or facultative anaerobes and anaerobes that are capable of performing hydrolysis achieve breakage of the unique bonds. Hydrolysis is the lysis of a compound with water. pH around 4.0 – 6.5

This stage complex substrate such as cellulose, hemicellulose, pectin, starch, protein and lipid were hydrolyzed to form simple molecule which bacteria cell can use.

This bacteria is in family *Streptococcaceae*

Enterobacteriaceae Bacillacea Lactobacillceae Bacteroides, Clostridium, Butyrivibrio, Eubacterium, Bifidobacterium, Lactobacillus, Propionibacterium, Ruminococcus, Acetivibrio, Peptostreptococcus, Peptococcus, Selenomonas, Desulfovibrio, Corynebacterium, Actinomyces, Staphylococcus, and Escherichia coli

2) Acetate-forming bacteria grow in a symbiotic relationship with methane-forming bacteria. Acetate serves as a substrate for methane-forming bacteria. The intermediate substrate are volatile fatty acid which drop pH then methane-forming bacteria digest it to balance pH. There are 2 groups of acetate-forming bacteria. A hydrogen producing acetate forming bacteria and homoacetate-froming bacteria

(1) Hydrogen producing acetate-forming bacteria

When acetate-forming bacteria produce acetate, hydrogen also produced. If the hydrogen accumulates and significant hydrogen pressure occurs, the pressure results in termination of activity of acetate-forming bacteria and lost of acetate production. However, methane-forming bacteria utilize hydrogen in the production of methane and significant hydrogen pressure does not occur.

$$CH_3CH_2OH + H_2O \longrightarrow CH_3COO^- + H^+ + 2H_2$$

(ethanol)

$$CH_3CHOHCOOH + H_2O \longrightarrow CH_3COOH + CO_2 + 2H_2$$

(lactate)

$$CH_3CH_2COOH + 2H_2O \longrightarrow CH_3COOH + CO_2 + 3H_2$$

(propionate)

(2) Homoacetate forming bacteria

This group of bacteria only produce acetate. Hydrogen and carbondioxide are absent. For example of homoacetate-forming bacteria is *Clostridium thermoaceticum* and *Butyribacterium methylotrophicum* which can produce acetate and butyrate

$$2C_6H_{12}O_6 + 2H_2O \xrightarrow{\textit{Butyribacterium methylotrophicum}} 2CH_3COOH+C_3H_7COOH+4CO_2+6H_2$$

$$2H_2 + CO_2 \xrightarrow{\textit{Clostridium thermoaceticum phicum}} CH_3COOH$$

2.3.2.2 Methane-forming bacteria

Methane-forming bacteria are some of the oldest bacteria and are grouped in the domain Archaebacteria. Methane-forming bacteria are oxygen-sensitive, fastidious anaerobes and are free-living terrestrial and aquatic organisms. Although methane-forming bacteria are oxygen sensitive, this is not a significant disadvantage. Methane-forming bacteria are found in habitats that are rich in degradable organic compounds. In these habitats, oxygen is rapidly removed through microbial activity. The domain thrives in heat. Archaebacteria comprise all known methane-forming bacteria, the extremely halophilic bacteria, thermoacidophilic bacteria, and the extremely thermophilic bacteria. The optimal pH range required by methanogenic bacteria is 6.8 – 7.6 (Appels, 2011)

Methanobacterium formicium and *Methanobrevibacter arboriphilus* are two of the dominant methane-forming bacteria in anaerobic digesters. Substrate are Hydrogen and carbon dioxide, methanol, formate, acetate, and trimethylamine as below equation.

$$4H_2 + CO_2 \longrightarrow CH_4 + 2H_2O$$

$$4HCOOH \longrightarrow CH_4 + 3CO_2 + 2H_2O$$

$$CH_3COOH \longrightarrow CH_4 + CO_2$$

$$4CH_3OH \longrightarrow 3CH_4 + CO_2 + 2H_2O$$

$$4(CH_3)_3N + H_2O \longrightarrow 9CH_4 + 3CO_2 + 6H_2O + 4NH_3$$

2.3.3 Factors affecting anaerobic digestion process

2.3.1.1 Temperature

Optimum temperature for bacterial growth has 3 ranges as shown in Table 2. They are Psychrophilic, Mesophilic, and Thermophilic. Anaerobic digestion can be developed for different temperature ranges including, mesophilic temperatures of approximately 35°C and thermophilic temperatures ranging from 55°C to 60°C (Kim et al, 2006).

Table 2.2 Temperature ranges for bacterial growth

Type	Temperature (°C)	Optimum temperature (°C)
Psychrophilic	10-30	12-18
Mesophilic	20-50	25-40
Thermophilic	35-75	55-65

Source: (Eddy, 2004)

Methanogen is sensitive to environmental changes. The changing of temperature may lead to lead to give slower reaction rates, lower gas production, and lower rates of the destruction of pathogens.

Operating temperature has limitation from different weather. In area with cold weather may need to control temperature with heater, but it is very suitable for Thailand a hot and humid weather with temperature around 20-35 °C. Mesophilic operation without adding any heat.

2.3.1.2 pH

The importance of the pH is due to the fact that methanogenic bacteria are very sensitive to acidic conditions and their growth and methane production are inhibited in acidic environment. It has been proven that the optimal range of pH for obtaining maximal biogas yield in anaerobic digestion is 6.5–7.5, the range is relatively wide in the plants and the optimal value of pH varies with substrate and digestion technique (Liu et al, 2008)

2.3.1.3 Alkalinity

The buffering capacity of an anaerobic digester is determined by the amount of alkalinity present in the system. The bicarbonate ion (HCO_3^-) is the main source of buffering capacity to maintain the system's pH in the range of 6.5 – 7.6. The concentration of HCO_3^- in solution is related to the percent of carbon dioxide in the gas phase. In a typical manure digester with a pH 7.4 and a percent CO_2 of 35%, the bicarbonate alkalinity is about 5,500 mg/L as CaCO3. Such alkalinity usually provides enough buffering capacity to withstand moderate shock loads of volatile fatty acids. In fact, cow manure can play an important role in co-digestion operations by increasing the pH and buffering capacity of the influent mixture when high-strength, easily degradable industrial wastes are used as co-substrates.

The acid/alkalinity ratio will change before the pH begins to drop. If the acids increase and drive the ratio out of the normal operating range, the digester may become upset. The volatile acid to alkalinity ratio should be kept below 0.4. The value of nearly 0.8 could cause system failure immediately from weak buffering capacity.

2.3.1.4 Toxic

A wide variety of substances have been reported to be inhibitory to the anaerobic digestion processes in substantial concentrations. Problems such as low methane yield and process instability are often encountered in anaerobic digestion. The inhibitors commonly present in anaerobic digesters include ammonia, sulfide, light metal ions, heavy metals, and other anthropogenic organic compounds such as solvents and pesticides in the waste.

1) pH impact

Both methane-forming and acid-forming microorganisms have their optimal pH. Failing to maintain pH within an appropriate range could cause reactor failure even though ammonia is at a safe level. Earlier researches showed the control of pH within the growth optimum of microorganisms is able to reduce ammonia toxicity.

2) Ammonia inhibition

Ammonia is one of the intermediate substances derived from hydrolysis and formed during the degradation of nitrogenous organic materials such as proteins and urea. Ammonium ion (NH_4^+) or free ammonia (NH_3) are produced in aqueous solution from degradation of amino acids and proteins, which could partly be converted into ammonium bicarbonate (NH_4HCO_3).

$$NH_4^+ \longleftrightarrow NH_3 + H^+ \text{ (p}K_a = 9.27 \text{ ที่ } 35\ ^{o}\text{C)}$$

If pH is higher 7.2, NH_3 could be found more which can cause the inhibition of microorganism activity. The inhibition could be found when the concentration reach 7,000-9,000 mg/l.

2.3.1.5 Volatile fatty acid

The concentration of all VFA increased during the digestive process, the rise in acetate concentration and the decreased of pH. The acetate concentration and the propionate to acetate ratio (P/A ratio) can be seen from ratio as valuable indicators to predict process failure. For manure, an acetic acid concentration of 0.8 g/l and a P/A ratio of 1.4 have been proposed as limit values (Krause, 1993)

2.4 Biogas

Biogas typically refers to a mixture of different gases produced by the breakdown of organic matter in the absence of oxygen. 2 types of bacteria was involved in this reaction, acid forming bacteria and Methane-forming bacteria. The main product of the anaerobic digestion process is a gas mixture of methane and carbon dioxide.

2.4.1 Typical biogas composition

Most of biogas composition is methane 50-80%. Other composition consists of Carbondioxide (CO_2) Hydrogen sulfide (H_2S) Nitrogen (N_2) Oxygen (O_2) Vapor (H_2O) as shown in Table 2.3.

Table 2.3 Typical biogas composition

Type	Ratio
Methane (CH_4)	50 - 80 % vol.
Carbondioxide (CO_2)	34 – 50 % vol.
Hydrogen sulfide (H_2S)	50-5,000 ppm
Ammonia (NH_3)	0-300 ppm
Oxygen (O_2)	< 1 % vol.
Nytrogen (N_2)	1 - 4 % vol
Vapor	2-5 % wt

Source : (Naskeo, 2009)

2.4.2 Benefits uses of biogas

Reduces pollution, Reduces time wastage while collecting firewood, Reduces reliance on fossil fuels, Lowers fuel import, Reduces deforestation, Improves living standards in rural areas, Reduces global warming, Produces good quality enriched manure to improve soil fertility, Effective and convenient way for sanitary disposal of organic wastes, Improving the hygienic conditions.

2.4.3.1 Application of biogas to energy

1) Heat generation : Biogas can be used directly as it has flammable activity. This application use in household as cooking gas.

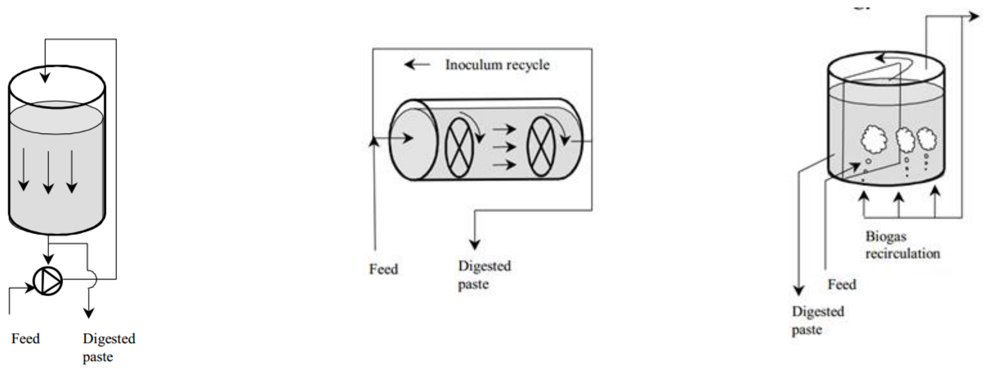
2) Power generation: Biogas can be used to operate a dual fuel engine to replace up to 80 % of diesel-oil. Diesel engines have been modified to run 100 per cent on biogas. Petrol and CNG engines can also be modified easily to use biogas.

2.5 Types of anaerobic digesters

The most common type of anaerobic digesters for solid wastes were compared based on biological, technical, performance and reliability. Thus there are 3 types of anaerobic digester system. They are one-stage, two-stage, and batch systems (Vandevivere, 2002).

2.5.1 Single-stage anaerobic digester system

The biomethanization of organic wastes is accomplished by a series of biochemical transformations, which can be roughly separated into a first step where hydrolysis, acidification and liquefaction take place and a second step where acetate, hydrogen and carbon dioxide are transformed into methane. In one-stage systems, all these reactions take place simultaneously in a single reactor. The fermenting wastes move via plug flow inside the reactors. There are 3 types of single-stage anaerobic digestion reactor shown in Figure 2.2



A.Dranco process B.Kompogas process C.Valorga process

Figure 2.1 3 types of single-stage anaerobic digestion reactor

Source : (Vandevivere, 2002)

A. Dranco process

In the Dranco process, the mixing occurs via recirculation of the wastes extracted at the bottom end, mixing with fresh wastes (1 fresh waste: 6 digested wastes), and pumping to the top of the reactor. This simple design has been shown effective for the treatment of wastes ranging from 20 to 50 % TS.

B. Kompogas Process

The Kompogas process works similarly as Dranco process, except that the plug flow takes place horizontally in cylindrical reactors. The horizontal plug flow is aided by slowly-rotating impellers inside the reactors, which also serve for homogenization, degassing, and resuspending heavier particles. This system requires careful adjustment of the solid content around 23% TS inside the reactor. At lower values, heavy particles such as sand and glass tend to sink and accumulate inside the reactor while higher TS values cause excessive resistance to the flow.

C. Valorga Process

The Valorga process is different from other type. It has a circular movement of waste. A mixing occurred by adding high pressure gas at the bottom of the tank every 15 minutes.

Fruteau de Laclos had studied Valorga Process with operation temperature 40 ^{o}C. The process can achieve even the organic loading rate was high. Ammonia concentration was higher than 3 g/l. There was not found the inhibition of ammonia in that research. It might be because there was not complete mixing inside the digester so only the first zone of the digester had high ammonia concentration then balance after transport to the next zone (Fruteau de Laclos, 1997).

2.5.1.1 Advantages and disadvantages of single-stage systems

Advantages

- Smaller reactor, less investment

- Complete hygienization

- Plug flow movement help to prevent shock load failure due to slow movement can divided zone of bacteria reaction

Disadvantages

- Loss of bacteria when release sludge may lead to loss of methane production efficiency

- Pre-treatment before put waste in the reactor

2.5.2 Two-stage anaerobic digestion system

Anaerobic degradation process can be separated into two phases. The first is acid fermentation phase where liquefaction-acidification reactions is occurred lead to the production of intermediate products predominated by the volatile organic acids; Second phase is known as methane fermentation phase resulting in the conversion of intermediates substances to methane. Due to bacterial varieties different is required in anaerobic digestion. To separate reactor in two-stage digester makes each phase in the suitable environmental conditions may lead to a larger overall reaction rate and biogas yield.

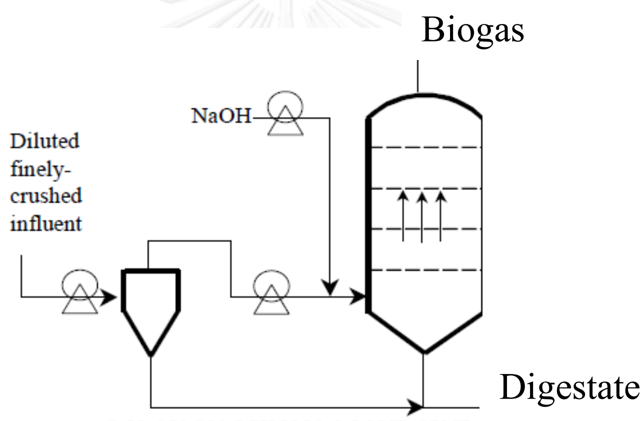


Figure 2.2 Two-stage anaerobic digester diagram

Source : Modified from (Vandevivere, 2002)

2.5.2.1 Advantages and disadvantages of two-stage systems

Advantages

- A greater biological reliability for wastes which cause unstable performance in one-stage systems

- Bacterial varieties different is required in anaerobic digestion. To separate reactor in two-stage digester makes each phase in the suitable environmental. (Optimum condition)

- Decrease methanogen lost as the acidogen has faster growth rate

- Only those two-stage systems with biomass retention schemes display stable performance with wastes excessively charged with nitrogen or other inhibitors

Disadvantages

- Complex design

- Larger investment

- More space

2.6 Litterature reviews

Anaerobic digestion of fruit and vegetable wastes to produce biogas was studied. It was carried out over 6 months to evaluate the most suitable operating parameters of the process depending on the availability of different kinds of fruit and vegetable wastes over the different periods of the year. Overall, the optimum daily loading rate of wastes was 35 kg/d, with a corresponding hydraulic residence time of 27 days. The optimum organic loading rate ranged from 2.5 to 3.0 kg_{VS}/m^3 d and the average specific biogas production was about 0.78 Nm^3/ kg_{VS}, with a specific methane yield of about 0.43 Nm^3/ kg_{VS}. A pay-Back Time of about 7.25 years can be achieved in the case of dispatching the electrical energy to the national grid (Scano et al, 2014).

Characterization of food waste as feedstock for anaerobic digestion was carried in pilot scale at the city of San Francisco, California. The anaerobic digestibility, biogas and methane yields of food waste were evaluated using batch anaerobic digestion perform at 50 ^{o}C. The ratio of volatile solid to total solid (VS/TS) were around 70 – 89%. The average methane content in biogas was 73% (Zhang et al, 2007).

Anaerobic digestion of autoclaved (160 °C, 6.2 bar) and untreated source food waste (FW) was compared. The untreated source of food waste was highger in methane yield 5-10% (maximum 0.483 ± 0.013 m^3 CH_4/kg VS at 3 kg VS/m^3 d) than autoclaved FW (maximum 0.439 ± 0.020 m^3 CH_4/kg VS at 4 kg VS/m^3 d). The residual methane potential of both digestates at all OLRs was less than 0.110 m^3 CH_4/kg VS, indicating efficient methanation. Autoclaved FW showed lower ammonium and hydrogen sulphide concentrations, probably due to reduced protein hydrolysis as a result of formation of Maillard compounds (Tampio et al, 2014).

Dry full scale anaerobic digestion of municipal solid waste (MSW) was studied compare between pure organic MSW, organic MSW with grey water and sludge. The result shown that the different organic composition effects the biogas production and digestate. Organic MSW was found to produce biogas 200 m^3/Ton of organic waste with methane 0.4 m^3 methane/kg_{VS} while the biogas production of organic MSW together with grey water and sludge can produce only 60 m^3/Ton of organic waste with methane 0.13 m^3 methane/kg_{VS} (Bolzonella, 2006).

The paper presents the results of a pilot- and full-scale experimental campaign on the anaerobic co-digestion of waste activated sludge and biowaste both in mesophilic and thermophilic conditions. The study demonstrated the possibility to increase the specific biogas production from 0.34 to 0.49 m^3/kgTVS and the gas production rate from 0.53 to 0.78 m^3per m^3 of reactor per day changing the reactor temperature from the mesophilic (37°C) to the thermophilic (55°C) range. The experimental work was carried out at pilot-scale, and the results match the full-scale behaviour(Cavinato et al, 2013).

The effect of ammonia-N accumulation in a dry anaerobic digestion was studied effectively using pilot-scale thermophilic reactor. Two simulations were prepared to attain C/N ratio 27 and C/N ratio 32 using bio-degradable feedstocks such as food waste, fruit and vegetable waste, green waste and paper waste. Organic loading rates and digestate recirculation rates were varied during different time intervals and the performance was evaluated using parameters like pH, VFA, Alkalinity, ammonia-N and biogas yield. Results showed that the simulation with C/N ratio 32 had about 30% less ammonia in digestate as compared to that with C/N ratio 27. The system performed well up to organic loading rate (OLR) 7-10 $kg_{VS}/m^{(3)}d$ and retention time up to 19 days, with surplus energy production of 50-73% (Zeshan et al, 2012).

A comparative evaluation of single-stage and two-stage anaerobic digestion processes for biomethane and biohydrogen production using thin stillage was performed to assess the impact of separating the acidogenic and methanogenic stages on anaerobic digestion. The separation of acidification stage increased the TVFAs to TCOD ratio from 10% in the raw thin stillage to 54% due to the conversion of carbohydrates into hydrogen and VFAs. Comparison of the two processes based on energy outcome revealed that an increase of 18.5% in the total energy yield was achieved using two-stage anaerobic digestion (Nasr et al, 2012).

CHAPTER III

METHODOLOGY

3.1 Process description

This research study prototype pilot scale anaerobic digestion system to compare both single-stage and two-stage systems and also compare the different feedstock ratio of rain tree leaf and food waste to get the optimal condition. This study can be divided into 2 experiment.

1) Experiment 1: Compare biogas production efficiency between Single-stage and Two-stage anaerobic digesters

2) Experiment 2: Compare the feedstock ratio of food waste and rain tree leaf by different in percent %food waste and % rain tree leaf. 85% food waste: 15% rain tree leaf, 90% food waste: 10% rain tree leaf, 95% food waste: 5% rain tree leaf, and 100% food waste.

Food waste in this experiment was collected from Chulachakrabongse and Mahitaladhibesra canteens, Chulalongkorn University, which was composed of food residues, vegetables, rice, meat and grease, etc. The food waste was shredded by food grinder into 5-10 mm size. Rain tree leaf was collected from area in Chulalongkorn University. The rain tree leaf was shredded by a leaf chipper shredder into less than 10 mm size, then dried it with ambient temperature for 2 weeks to obtain identical moisture content. Feed around 20 kgs/day in both digesters.

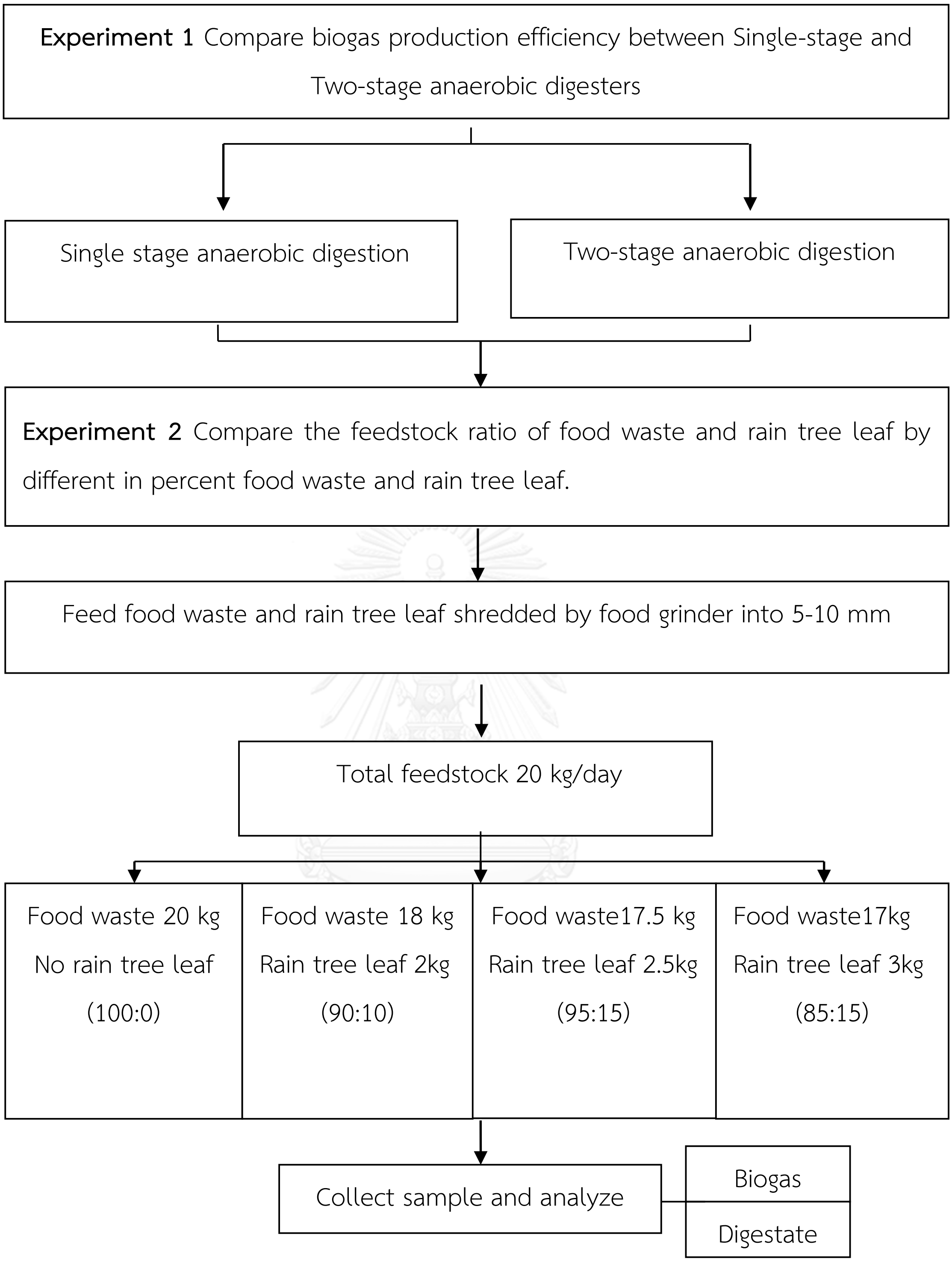


Figure 3.1 Methodology

3.2 Parameters

This experiment compose of 3 parameters which are independent parameter, control parameter, and dependent parameter. First experiment and second experiment study parameter from start up until stationary phase shown in Table 3.1.

Table 3.1 Parameter in this experiment

Experiment	Independent Parameter	Control parameter	Dependent parameter
1	Compare biogas production efficiency between Single-stage and Two-stage anaerobic digesters	- feedstock 20 kg/day - Mixing time 60 min/day	- pH - Temperature - Total solid - Total volatile solid - Volatile fatty acid - Total alkalinity - COD - biogas - biogas composition
2	Compare the feedstock ratio of food waste and rain tree leaf by different in percent food waste and rain tree leaf. Food waste (100 : 0) Food waste : Rain tree leaf (90 : 10) Food waste : Rain tree leaf (95 : 5) Food waste : Rain tree leaf (85 : 15)		

3.3 Material

3.3.1 Anaerobic digesters (pilot scale)

3.3.1.1 Single-stage anaerobic digester

Volume of the single-stage anaerobic digester is 2500 liters with working volume around 1250 liters. A horizontal plug-flow cylinder digester type (Kompogas process) with 1.2 meters in diameter. Feed stock was shared by food then moved into the single-stage anaerobic digester by a screw conveyor. A paddle type mixer was provided for slow mixing at short period after waste feeding to a digester tank. The biogas generated from the anaerobic activity was kept in the biogas holding tank and sent through the gas pipeline for further utilized in canteen.

Figure 3.2 Pilot scale single-stage anaerobic digester

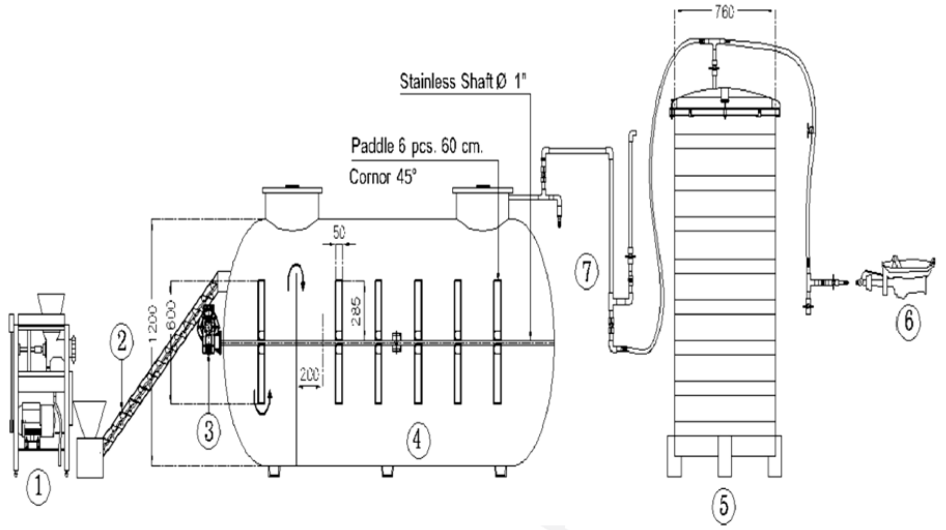


Figure 3.3 Diagram of the prototype single-stage anaerobic digester
(1. Food grinder 2.Screw conveyor 3.Motor 4.Anaerobic digester 5.Biogas holding tank 6.Biogas utilization for canteen 7.Manometer)

3.3.1.2 Two-stage anaerobic digester

Two-stage anaerobic digester consists of two reactors. They are acid reactor and biogas reactor

1) Acid reactor has volume 1,000 liters. A vertical cylinder with height 1.56 meters. Food waste pump was installed inside the reactor to pump food waste into acid tank and sending food waste from acid tank to methane tank

2) Biogas tank with volume 2,500 liters. A vertical cylinder compose of biogas pipe connected from acid tank to biogas tank and biogas holding tank. The upper part installed motor with the 3 mixing paddles to mix food waste

A. Acid reactor B. Biogas reactor

Figure 3.4 Two-stage anaerobic digester consists of acid reactor and biogas reactor

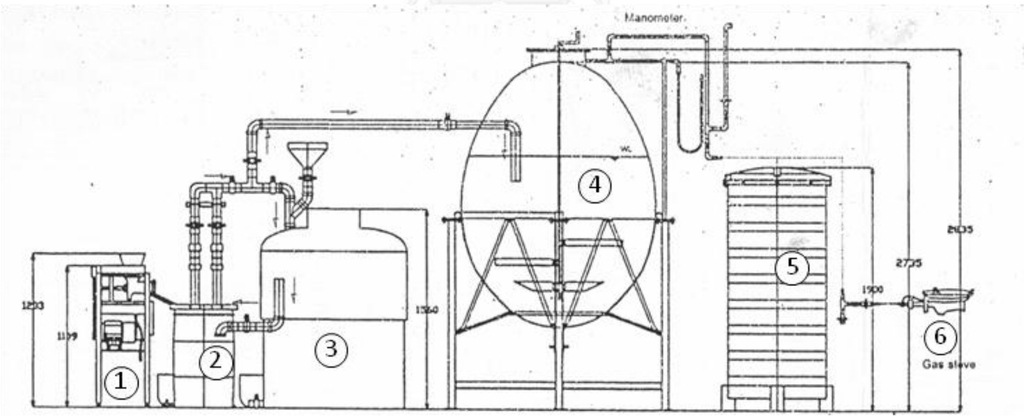


Figure 3.5 Schematic diagram of the prototype two-stage anaerobic digesters (1. Food grinder 2.Food tank 3.Acid Tank 4.Methane Tank 5.Biogas holding tank 6.Biogas utilization for canteen)

3.3.1.3 Biogas holding tank

Biogas from single-stage or two-stage anaerobic digester was sent to keep in biogas holding tank with floating drum type. After the holding tank containing biogas, then it will be rising up. The volume of biogas was measured by gas flow meter connected with anaerobic digester.

Figure 3.6 Gas holding tank (floating drum)

Figure 3.7 Gas flow meter connected anaerobic digester and biogas holding tank

3.3.1.4 Compressed pressure tank

To improve efficiency of using biogas in the canteen. Compressed pressure tank was installed to increase pressure to 2 bar, and then pump biogas to use for cooking.

Figure 3.8 Compressed biogas tank

3.4 Methodology

3.4.1 Food waste preparation

Food waste from the canteens of Chulachakrabongse and Mahitaladhibesra buildings, Chulalongkorn University, which was composed of food residues, vegetables, rice, meat and grease, etc. Plastic, bone and shell which are difficult to degrade were separated. After that food waste was shredded by food grinder into 5-10 mm size and mixed with rain tree leaf. Parameter below were analyzed.

- COD
- Total solid
- Total volatile solid
- C:N ratio
- pH

Figure 3.9 Food waste

Figure 3.10 Food grinder

3.4.2 Rain tree leaf preparation

The rain tree leaf was shredded by a leaf chipper shredder into less than 10 mm size and mixed with food waste then analyze

-

Figure 3.11 Rain tree leaf

3.4.3 Start up anaerobic digestion process

Digestate from Chulalongkorn Dormitory Building and Samyarn Market which was adapt to digest food waste around 150 liter, 10 kilograms of castle manure and dilute with 50 liter of water. Start up around 1 month then start the experiment.

3.4.4 Methodology

3.4.4.1 Experiment 1

1) Compare biogas production efficiency between Single-stage and Two-stage anaerobic digesters. Feedstock was fed to anaerobic digester tank 20 kg/day, 5 day a week. Mixing time was 60 minutes for 28 days.

2) Digestate was collected and analyzed COD, VFA, Total Solid, Total Volatile solid, Alkalinity pH Temperature. Biogas production volume and composition were analyzed.

3.4.4.2 Experiment 2

1) Compare biogas production and degradation of waste within the different ratio of food waste to rain tree leaf. Food waste (100: 0), Food waste: Rain tree leaf (90: 10), Food waste: Rain tree leaf (95: 5), Food waste: Rain tree leaf (85: 15)

2) Digestate was collected and analyzed COD, VFA, Total Solid, Total Volatile solid, Alkalinity pH Temperature. Biogas production volume and composition were analyzed.

3.5 Analytical method

Record daily biogas production volume. Composition of biogas was analyzed by Gas Chromatography. Digestate was collected and analyzed parameter as shown in Table 3.2.

Table 3.2 Analytical frequency and method of digestate and biogas

Parameter	Analytical method	Frequency
Biogas production volume	Gas meter	A
Temperature	Thermometer	A
pH	pH meter	A
COD	Closed Reflux, Titration Method	B
Total solid (TS)	Oven 105 ^{o}CB	B
Total volatile solid (TVS)	Burn in furnace 550 ^{o}C	B
Volatile fatty acid (VFA)	Direct Titration Method	B
Alkalinity	Direct Titration Method	B
% Methane	GC (Gas Chromatography)	C

A analyzes parameter 5 day per week

B analyzes parameter 3 day per week

C analyzes parameter 1 day per week

Standard methods for examination of water and wastewater (American Public Health Association, 1995)

CHAPTER IV

RESULT AND DISCUSSION

4.1 Characterization of food waste and rain tree leaf as feedstock

The physical and chemical characteristics of the feedstock are important information for designing and operating anaerobic digesters, because they affect biogas production and process stability during anaerobic digestion (Zhang et al, 2007). In this experiment feedstock of single-stage and two-stage anaerobic digester were collected from two canteens inside Chulachakrabongse and Mahitaladhibesra Buildings at total solid loading of around 20 kilograms per day.

The different value of the feed stock depends on a variety of food waste from canteen. Table 4.1 shows the characteristic of all co-substrates (food waste (FW) and rain tree leaf (RTL) as feedstock (varying substrate ratios to 85% FW: 15% RTL, 90% FW: 10% RTL, 95% FW: 5% RTL and 100% FW) to be fed in anaerobic co-digestion process. The co-substrate feed had pH 6.22 ± 0.30, 5.57 ± 0.69, 5.78 ± 0.53, and 6.09 ± 0.16 for 85% FW: 15% RTL, 90% FW: 10% RTL, 95% FW: 5% RTL and sole 100% FW, respectively. A high concentration of COD was found to be 131,344 ± 13,589 mg/l, 144,854 ± 16,528 mg/l, 151,534 ± 21,980 mg/l, and 157,067 ± 35,933 mg/l, respectively. The carbon to nitrogen ratio (C/N) was in the range of 17.88 to 21.45. The percentage of volatile solids to total solids ratios (VS/TS) were 90.23 ± 1.44, 93.36 ± 0.39, 94.58 ± 0.75 and 93.94 ± 0.86 respectively, the results show that all feedstock have high in organic content above 90% which were suitable to use as substrate for bacteria growth.

Table 4.1 Characteristics of feedstock

Feedstock	85% FW with 15% Rain tree leaf	90% FW with 10% Rain tree leaf	95% FW with 5% Rain tree leaf	100% Food waste
pH	6.22 ± 0.30	5.57 ± 0.69	5.78 ± 0.53	6.09 ± 0.16
COD (mg/L)	131,344 ± 13,589	144,854 ± 16,528	151,534 ± 21,980	157,067 ± 35,933
C/N ratio	17.88 ± 3.21	19.03 ± 3.79	20.77 ± 4.23	21.45 ± 4.43
TVS (mg/L)	201,119 ± 12,118	204,121 ± 8,586	166,843 ± 13,605	122,463 ±20,683
TS (mg/L)	222,949 ± 15,111	204,121 ± 9,841	176,462 ± 14,934	130,308 ± 21,516
TVS/TS (%)	90.23 ± 1.44	93.36 ± 0.39	94.58 ± 0.75	93.94 ± 0.86

Carbon to nitrogen is an important factor that is reflected by the nutrient levels of feedstock. A high C/N ratio induces a low protein solubilization rate and leads to low total ammonia nitrogen (TAN) and volatile fatty acid (VFA) concentration. Thus ammonia inhibition might not occur through optimizing the C/N ratio in the anaerobic digestion system. However, an excessively high C/N ratio provides insufficient nitrogen to maintain cell biomass, leading to fast nitrogen degradation by microbes and eventually leading to low biogas production. A low C/N ratio increases chance of ammonia inhibition due to high TAN and VFA concentration and this can lead to rising pH which inhibits methane-forming bacteria and possibly causing to insufficient utilization of carbon sources. The optimal C/N ratio for anaerobic digestion has been suggested to be between 20 to 30. (Mao et al, 2015). However, the recommended the C/N ratio for the co-digestion of onion juice and digested sludge should be maintained at 15 (Romano & Zhang, 2008); and for corn stover, inoculated with digested sewage sludge, the digestion process worked well with a C/N ratio between 15 to 18 but failed with a C/N ratio of 21 or higher because the pH dramatically decreased in the first 7 days at 37 ^{o}C.

In this study we found that the C/N ratio of our feed stock was in the range of 17.88 – 21.45, which was optimal for operating the anaerobic co-digestion system. The C/N ratio decreased with an increase in rain tree leaf composition as rain tree leaf has high in nitrogen around 3.12±0.05 percent nitrogen content. The feed mixing ratio of

85% food waste mixed and 15% rain tree leaf has the lowest C/N ratio as shown in Figure 4.1. The C/N ratios of four mixing ratios of food waste and rain tree leaf were 17.88 ± 3.21, 19.06 ± 3.79, 20.77 ± 4.23, and 21.45 ± 4.43 with 85%FW: 15%RTL, 90%FW: 10%RTL, 95%FW: 5%RTL and 100% food waste, respectively.

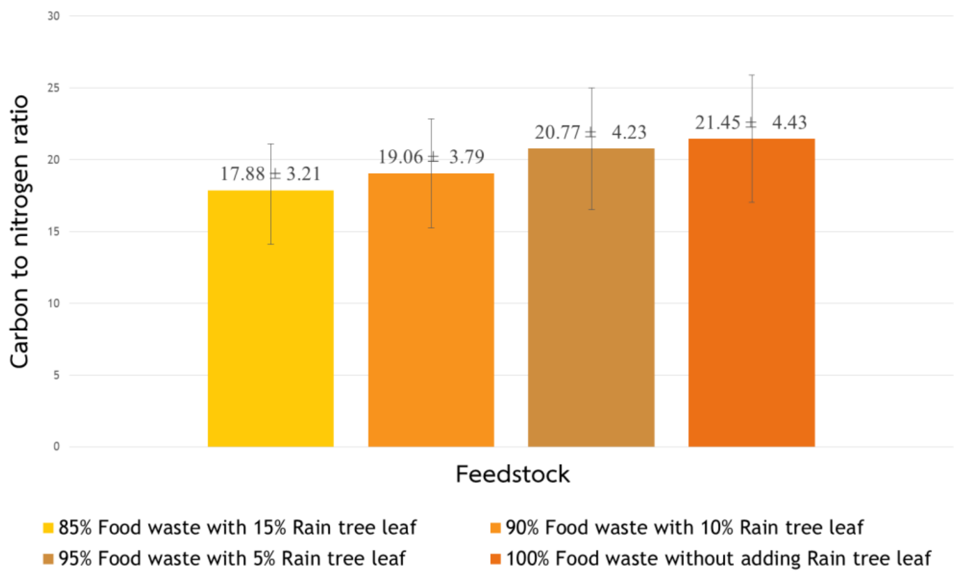


Figure 4.1 C/N ratio of feedstock at different mixing ratios

4.2 Operating condition of anaerobic co-digestion process

4.2.1 Temperature

A pilot scale single-stage and two-stage anaerobic digesters were operated in the same period of time. After the system start-up, four mixing ratios of food waste and rain tree leaf were study. The first phase was carried out with 85%FW 15%RTL during a rainy season (average temperature 29.3±1.0 °C shown in Figure 16). The second and third phase were carried out with the feed mixing ratio of 90%FW 10% RTL and 95%FW 5% RTL from September to October (average temperature 29.9±1.6 °C and 31.2±0.9 °C shown in Figure 4.2). For system operation with 100% FW was also done from November to December with the operating temperature of 28.0±1.3 °C. Although temperatures from different phase are a little bit different but they are still in optimal temperature for microorganism to grow in mesophillic temperature range (25.9 – 32.6 °C).

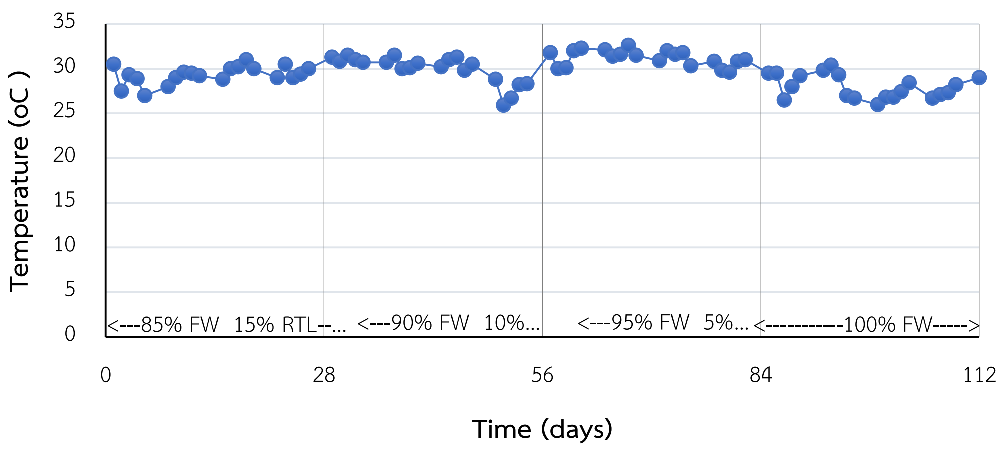


Figure 4.2 Operating temperature for anaerobic co-digestion system

4.2.2 Pressure

Pressure is an important parameter to convert gas to standard temperature and pressure (STP). The gas production volumes from anaerobic digestion should be shown in standard conditions with comparison to sea level standard atmospheric pressure. To analyze the gas production volume as Normal cubic meter will be mentioned in appendix topic. Pressure was measured by barometer every day. The average pressure shown in Figure 4.3 was 1,009.9±2.8 hPa. The first phase was 1009.4±1.1 hPa, the second phase was 1011.5±2.1 hPa, the third phase was 1006.7±2.0 hPa and the last phase which was in winter season of Thailand had the highest pressure 1012.0±2.1 hPa.

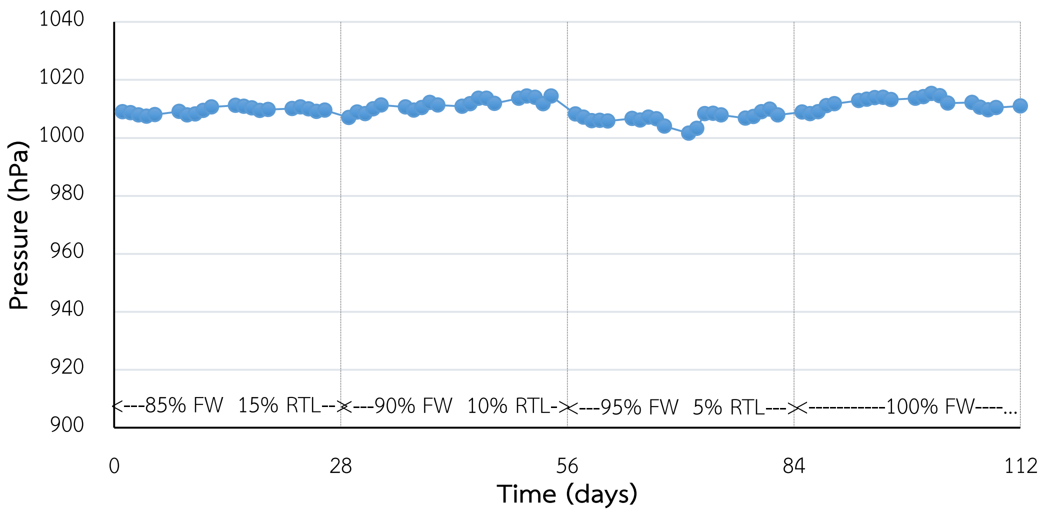


Figure 4.3 Operating pressure

4.3 Performance Evaluation for Anaerobic digesters

4.3.1 Single-stage anaerobic co-digestion of food waste and rain tree leaf

4.3.1.1 pH

It has been recognized that the optimal range of pH for obtaining maximum biogas yield in anaerobic digestion is 6.5–7.8, the range is relatively wide and the optimal value varies with substrate and digestion technique (Liu et al, 2008). The pH value is a function of volatile fatty acid (VFA) concentration, bicarbonate concentration, and alkalinity of the system as well as the fraction of CO_2.

The operating pH in single-stage anaerobic co-digestion was shown in Figure 18. By separating results into different ratios of feedstock as 85%FW15%RTL, 90%FW10%RTL, 95%FW5%RTL, and 100%FW, the obtained pHs were 7.74 ± 0.11, 7.77 ± 0.11, 7.88 ± 0.06 and 7.76 ± 0.16 respectively. Overall operating pHs were suitable for acetogens and methanogenic bacteria growth for the entire experiment.

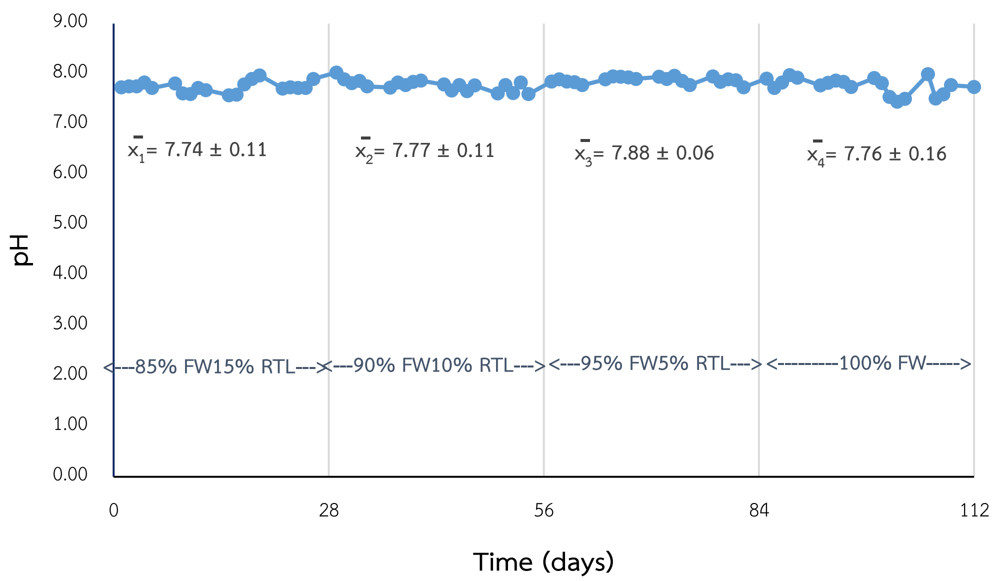


Figure 4.4 Operation pH in single-stage anaerobic co-digestion system

4.3.1.2 Total Chemical Oxygen Demand (COD)

The feedstock put in the reactor was a mixture of food waste from the campus restaurant, consisting of mixed cooked and fresh leftover food, and rain tree leaf. The preparation included homogenization of co-substrate of food waste and rain tree leaf into a food grinder and separated into 2 batch for feed single-stage and two-stage anaerobic digesters. The average COD concentrations of feed stock of 1st phase (85% FW 15% RTL), 2nd phase (90% FW 10% RTF), 3rd phase (95% FW 5% RTF) and 4th phase (100% FW) were 131,344 ± 13,589 mgCOD/l, 144,854 ± 16,528 mgCOD/l, 151,534 ± 21,980 mgCOD/l and 157,067 ± 35,933 mgCOD/l, respectively.

The COD of liquid digestate from the single-stage anaerobic co-digestion is shown in Figure 4.5. The liquid digestate was digested from the reactor having high COD concentration. By separating results into different ratios of feedstock as 85%FW15%RTL, 90%FW10%RTL, 95%FW5%RTL, and 100%FW, the COD concentrations obtained with the liquid digestate were 41,408±2,426 mgCOD/l, 36,525±6,646 mgCOD/l, 36,021±8,625 mgCOD/l, and 27,991±7,184 mgCOD/l, respectively. The result shows that the digestate COD was the lowest with 100%FW, compared to other ratios with co-digestion of FW and RTL. The trend of digestate COD was higher when adding more rain tree leaf. The texture of rain tree leaf helps in mixing and releasing of dissolve gas.

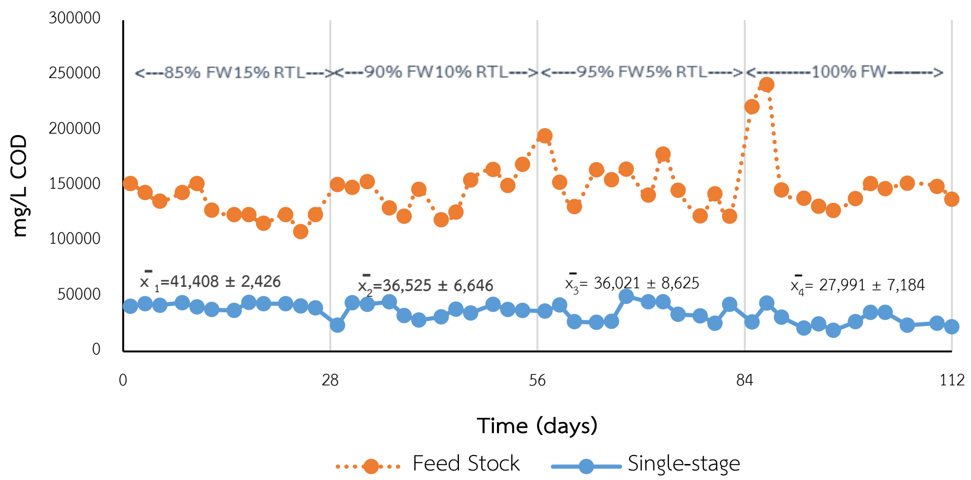


Figure 4.5 Total COD in single-stage anaerobic co-digestion system

4.3.1.3 Volatile fatty acid

Volatile fatty acid is an intermediate substrate for producing methane. If the accumulation of VFA is high can lead to the failure of reactor, Therefore, VFA is an important parameter predicted reactor failure according to the VFA levels on digester (Franke-Whittle et al, 2014) Volatile fatty acid in acetic acid form (CH_3COOH) of the single-stage digester is shown in Figure 4.6 with average concentration of 1,693±432 mgCH_3COOH/l. By separating results into different ratios of feedstock as 85%FW:15%RTL, 90%FW:10%RTL, 95%FW:5%RTL, and 100%FW, the VFA results were 1,583±550 mgCH3COOH/l, 1,956±435 mgCH3COOH/l, 1,699±362 mgCH3COOH/l, and 1,536±288 mgCH3COOH/l, respectively.

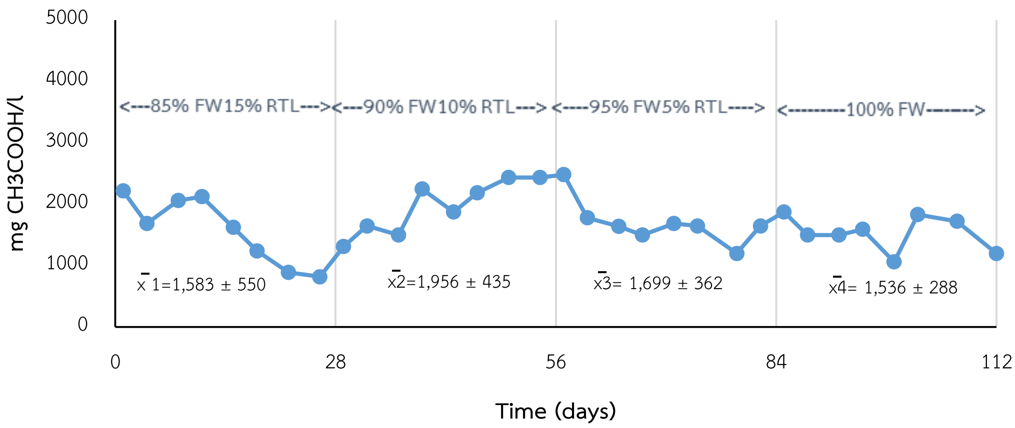


Figure 4.6 Volatile fatty acid in the single-stage anaerobic co-digestion system

4.3.1.4 Volatile fatty acid Composition

Acetic acid serves as an intermediate substrate for methane-forming bacteria. The result shown in Figure 20 indicates that high concentration of acetic acid was found after digesting a complex molecule of FW and RTL to form a simple molecule. The anaerobic co-digestion of FW and RTL were observed with co-substrate ratios of 85%FW:15%RTL, 90%FW:10%RTL, 95%FW:5%RTL and 100%FW, the concentrations of acetic acid in the digester were 1,690±298 mg/l, 2,012±585 mg/l, 728±199 mg/l, and 636±121 mg/l respectively. In 95%FW5%RTL and 100%FW low concentration of acetic acid were achieved.

Propionic acid was used by hydrogen producing acetate-forming bacteria to produce acetate, hydrogen, and carbon dioxide. Single-stage anaerobic digester produces propionic acid in the range of 104.13±342.87mg/l 221.63±104.13mg/l 680.22±196.83 mg/l 326.31±62.11 mg/l 329.31±68.58 mg/l, respectively.

Acetate and propionate concentrations of up to 6,000 and 3,000 mg/L shows no inhibition of the AD process in reactors treating manure (Ahring, 1995). Thus, the high acetate concentrations seen in the reactors under investigation may not have been problematic (Ahring, 1995).

Butyric, Isobutyric, Isovaleric and n-Veleric acid are longer chain fatty acids. They were not much available. The Isocarpronic and N-capronic acid was found a little in a single-stage digester.

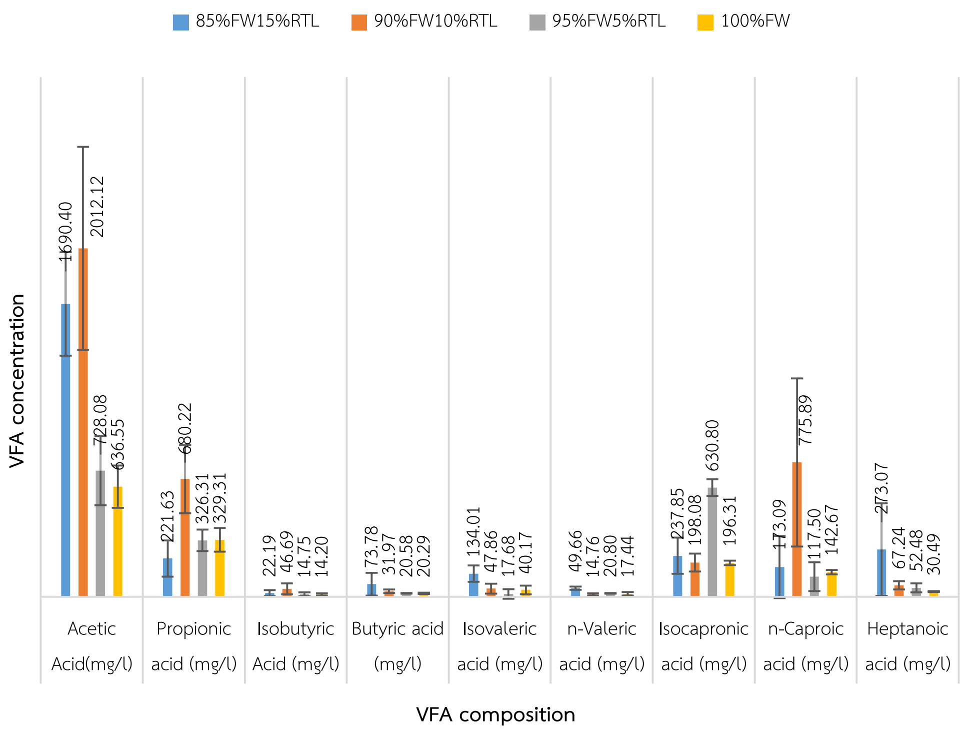


Figure 4.7 Composition of volatile fatty acid in single-stage anaerobic digestion

4.3.1.5 Ratio of propionate to acetate

A rise in the ratio of propionic to acetic acid was observed in the single-stage digester when decreasing the rain tree leaf in feed stock. The propionic to acetic acid ratio was raised from 0.13 in 85%FW:15%RTL to 0.34 in 90%FW:10%RTL, 0.45 in 95%FW: 5%RTF and finally 0.52 in 100%FW.

The ratio of propionic to acetic acid increased, depending on the changes in feed stock as adding of rain tree leaf can effect of more variety of bacterial. The lower value show more stability of the anaerobic digester. The result in every ratio found that propionic to acetic acid were lower than 1.4 which mean the system were stable, It has been recognized that ratio up to 1.4 can cause failure of digestion activity due to high content of propionic to acetic which is more complex for bacterial cell to use (Marchaim & Krause, 1993).

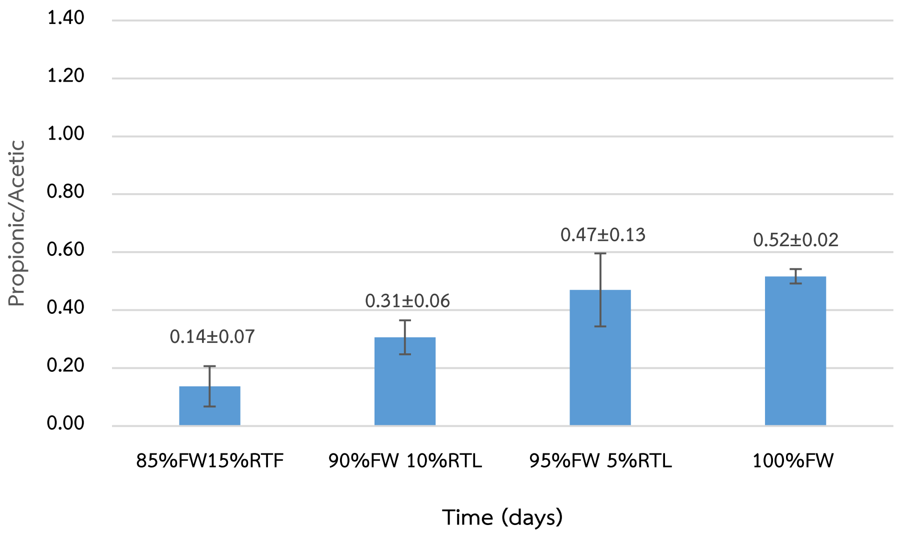


Figure 4.8 Ratio of propionic to acetic acid in single-stage

4.3.1.6 Total alkalinity

This experiment did not control pH by adding any chemicals. Alkalinity balance was achieved by ammonium bicarbonate (NH_4HCO_3) from anaerobic digestion of organic matter to organic acid and ammonium (NH_4^+). After organic acid was used by methanogen to produce methane (CH_4) and carbon dioxide (CO_2). The leftover ammonium reacted with carbon dioxide to form ammonium bicarbonate (NH_4HCO_3). Alkalinity prevents fluctuations of pH in an anaerobic digester. Alkalinity of single-stage anaerobic digester were 14,925±2,251 mg/l$CaCO_3$ with 85%FW:15%RTL, 14,894±1,193

$mg/lCaCO_3$ with 90%FW:10%RTL, 17,897±480 $mg/lCaCO_3$ with 95%FW:5%RTL, and 17,711±731 $mg/lCaCO_3$ with 100%FW. These values were obviously higher than 1,500 mg/L, which is commonly considered as the upper limit for allowing stable operation of biogas digester (Angelidaki I, 2005)

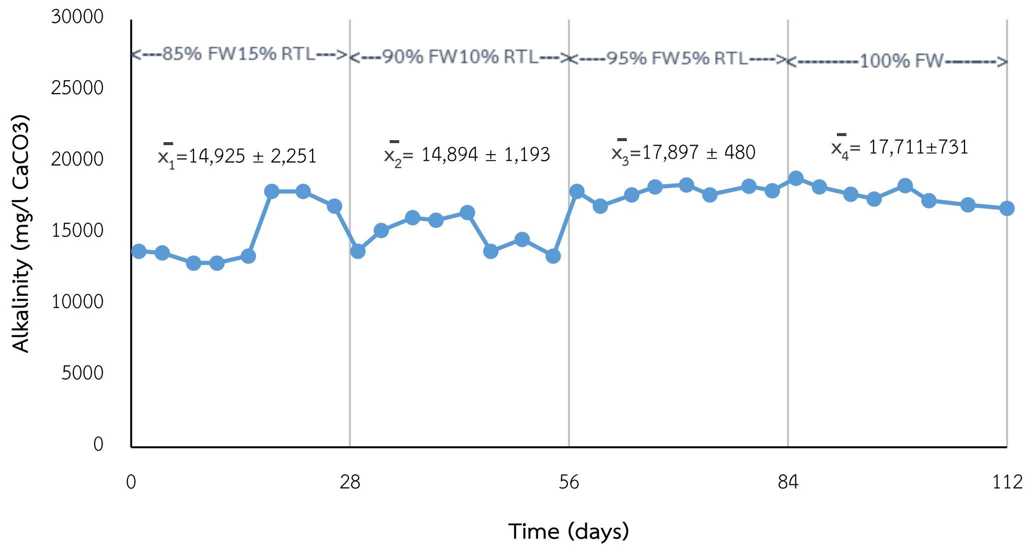


Figure 4.9 Total alkalinity in single-stage anaerobic digesters

4.3.1.7 Buffering capacity (VFA/Alk ratio)

To ensure digester stability in terms of pH, it is important to monitor the relationship between alkalinity and volatile fatty acids. A volatile fatty acid to alkalinity ratio (VFA/Alk) is an effective way to monitor digester stability. The results show that single-stage anaerobic digesters had VFA/Alk ratios lower than 0.4 which means that the systems had strong buffering capacity.

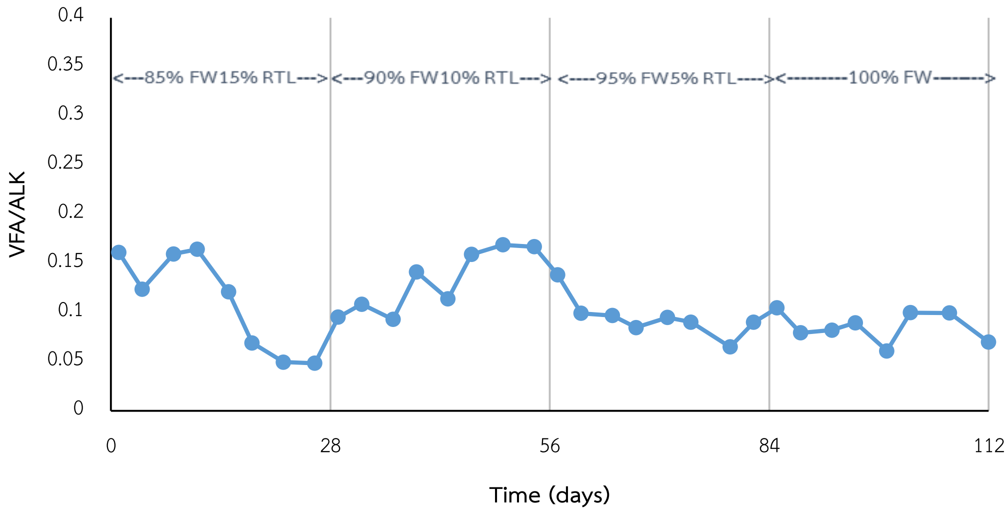


Figure 4.10 Buffering capacity of single-stage anaerobic digester

4.3.1.8 Total Solid

Total solid of feed stock varied by 1st phase (85%FW 15%RTL), 2nd phase (90%FW with 10%RTL), 3rd phase (95%FW with 5%RTL) and 4th phase (100% FW). They were 222,949 ± 15,111, 204,121 ± 9,841, 176,462 ± 14,934, 130,308 ± 21,516 mg/l of total solid, respectively. After anaerobic digestion process took place, the total solid of single- stage was mostly reduced to average values 38,069±2,783 mg/l of total solid, 36,983±3,361 mg/l of total solid, 39,628±2,481 mg/l of total solid, and 37,029±3,366 mg/l of total solid, respectively. The total solid reduction were around 73 – 81%

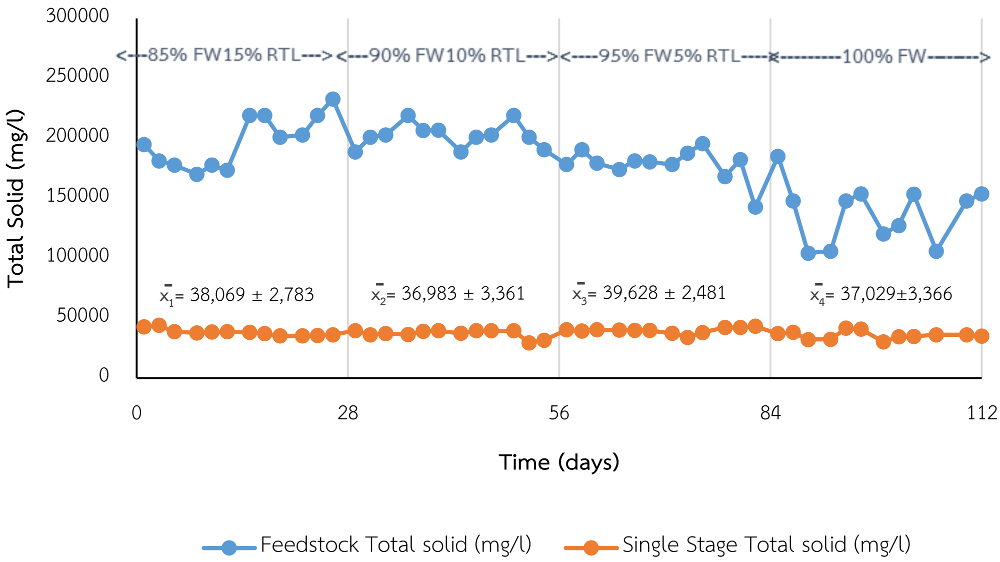


Figure 4.11 Total solid in feedstock and single-stage anaerobic digester

4.3.1.9 Total Volatile Solid

Total volatile solid of feed stock varied by first phase (85% FW with 15% RTL), 2^{nd} phase (90% FW with 10% RTL), 3rd phase (95% FW with 5% RTL) and 4^{th} phase (100%FW). They were 201,119 ± 12,118, 204,121 ± 8,586, 166,843 ± 13,605, and 122,463 ±20,683 mg/l of total volatile solid, respectively. After anaerobic digestion process took place, the total volatile solid of single- stage was mostly reduced to 25,201 ± 1,964 mg/l of total volatile solid, 22,723 ± 2,925 mg/l of total volatile solid, 21,838 ± 2,296 mg/l of total volatile solid, and 18,900 ± 2,596mg/l of total volatile solid, respectively. Calculation of percent reduction of total volatile solid found that 69-89 percentage of total solid were removed in single-stage.

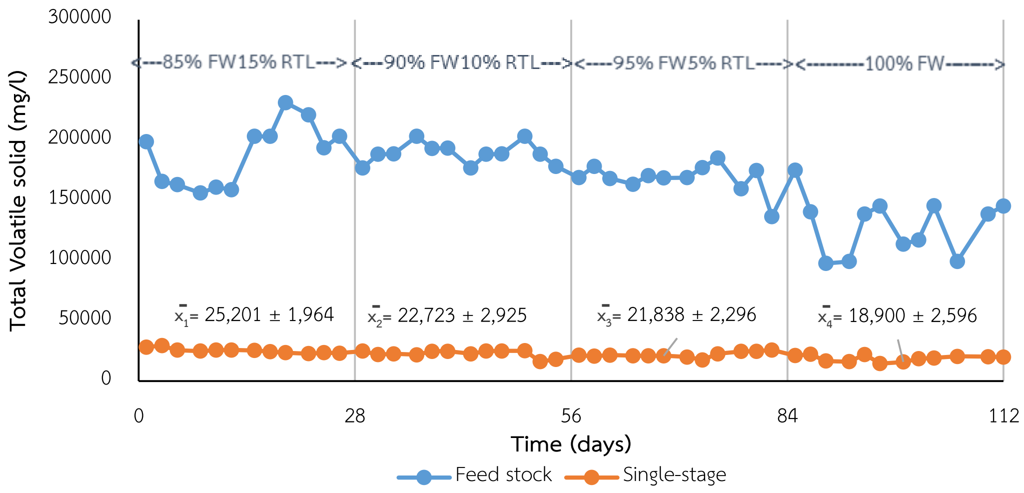


Figure 4.12 Total volatile solid of feedstock and single-stage anaerobic digester

4.3.2 Two-stage anaerobic co-digestion of food waste and rain tree leaf

4.3.2.1 pH

It has been recognized that the optimal range of pH for obtaining maximum biogas yield in anaerobic digestion is 6.5–7.8, the range is relatively wide and the optimal value varies with substrate and digestion technique (Liu et al, 2008). When feed stock enter into the digesters, organic matter was digested into volatile fatty acid (mainly acetic and propionic acids). pH dropped in acid tank of two-stage anaerobic digester as shown in Figure 4.13, pH was averagely around 3.35 ± 0.20 After that, the digested volatile fatty acid entered into methane tank, the methanogenic bacteria plays an important role to degrade acetic acid into methane and carbon dioxide where carbon dioxide was changed to form bicarbonate (HCO_3^-) that buffering pH. The pH was raised up in the methane tank of the two-stage digester as 7.57 ± 0.29 in 85%FW15%RTL, 7.48 ± 0.28 in 90%FW10%RTL, 7.47 ± 0.35 in 95%FW5%RTL and 7.44 ± 0.32 in 100%FW. This pH range was suitable for the growth of acetogenic bacteria and methanogenic bacteria.

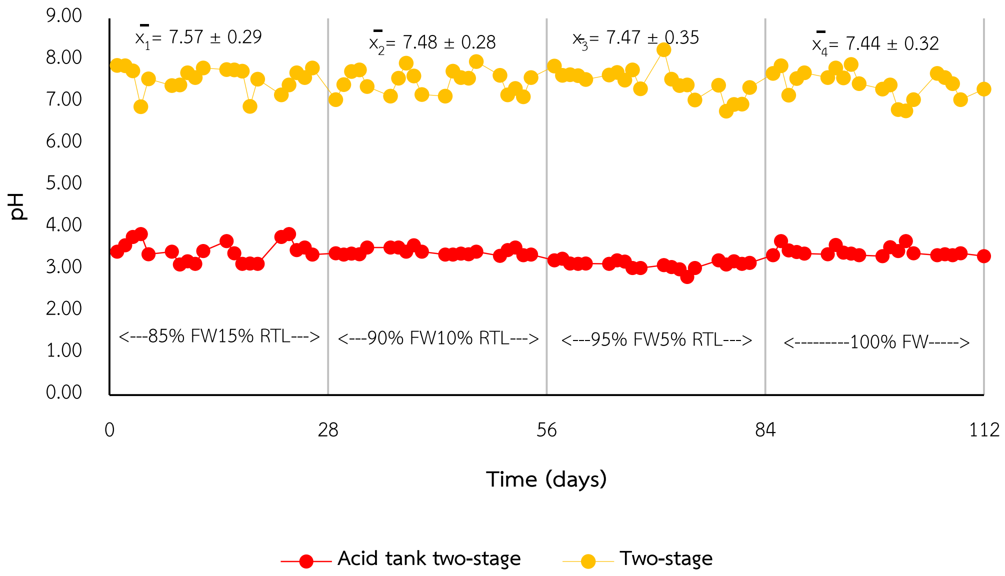


Figure 4.13 Operation pH in acid digester and methane digester of two-stage anaerobic co-digestion system

4.3.2.2 Total Chemical Oxygen Demand (COD)

The feedstock put in the reactor was food waste from the campus restaurant, consisting of mixed cooked and fresh leftover food. The preparation included homogenization of co-substrate of food waste and rain tree leaf into a food grinder and separated into 2 batches for feeding both single-stage and two-stage anaerobic digesters. The average COD concentrations can be seen in feedstock on Table 6.

The average COD concentrations of two-stage anaerobic co-digestion are shown in Figure 4.14 by separating the obtained results into different ratios of feedstock as 85%FW:15%RTL, 90%FW:10%RTL, 95%FW:5%RTL, and 100%FW, the COD concentrations of the liquid digestate were 26,451±3,942 mgCOD/l, 32,387±2,413 mgCOD/l, 33,613±4,137 mgCOD/l, and 38,298±5,787 mgCOD/l, respectively. The trend of COD was lower when adding more rain tree leaf.

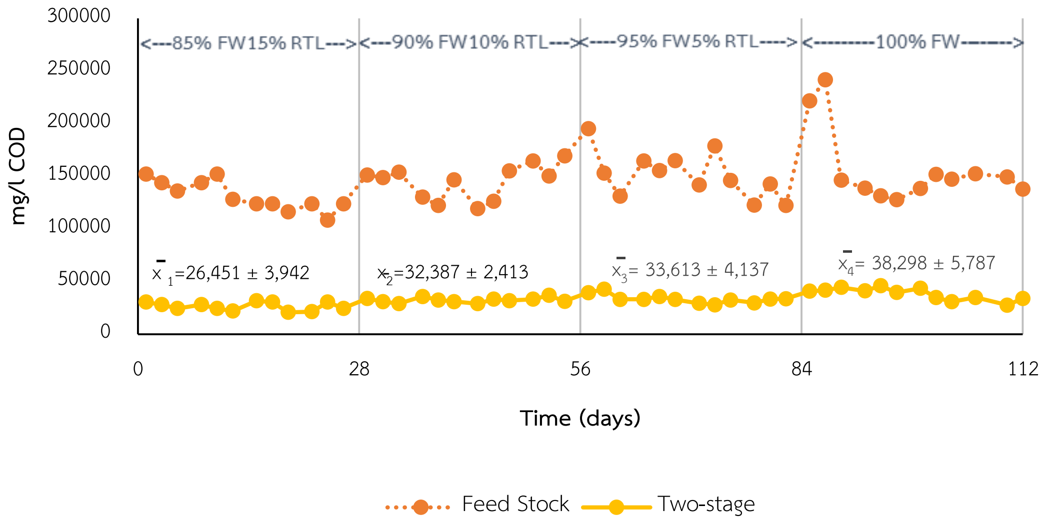


Figure 4.14 Total COD in two-stage anaerobic co-digestion system

4.3.2.3 Volatile fatty acid (VFA)

Volatile fatty acid is an intermediate substrate for producing methane. If the accumulation of VFA is high can lead to the failure of reactor, Therefore, VFA is an important parameter predicted reactor failure according to the VFA levels on digester (Franke-Whittle et al, 2014), Volatile fatty acid in acetic acid form (CH_3COOH) of the two-stage anaerobic digester has lower VFA concentration of average 1,297±436 mgCH_3COOH/l than that of the single-stage. Since bacterial varieties are required in anaerobic digestion. Then, separating reactor in two-stage digester makes each phase in the suitable environmental conditions and higher VFA degradation (Cooney et al, 2007). In this experiment, pH values did not change significantly, and a high bicarbonate and ammonia contents most likely contributed to the buffering of the system.

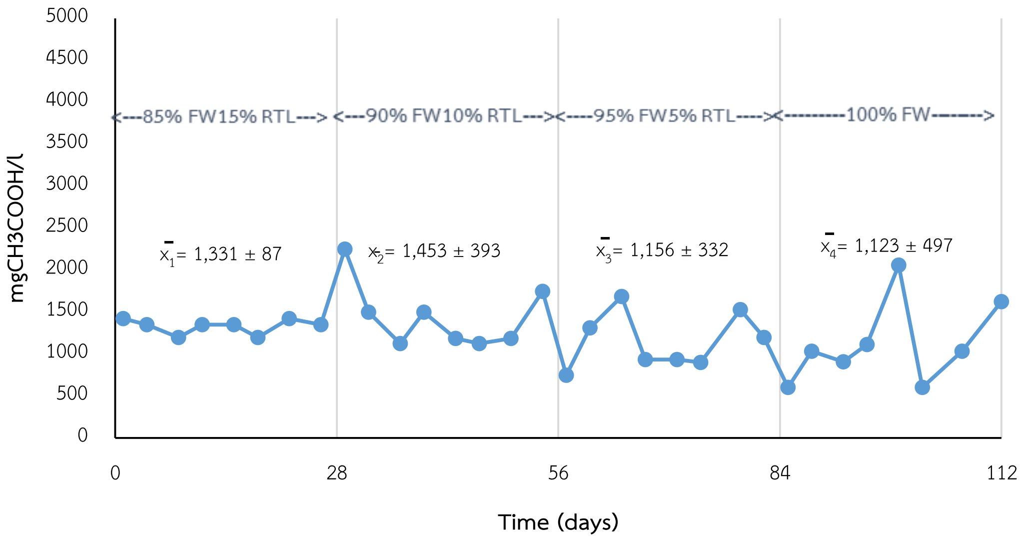


Figure 4.15 Volatile fatty acid in the two-stage anaerobic co-digestion system

4.3.2.4 Volatile fatty acid Composition

Acetate serves as an intermediate substrate for methane-forming bacteria. In acid tank of the two-stage anaerobic digester which acidogenesis and acetogenesis reaction occurred. High concentration of acetic acid content was found around 42,780 to 62,736 mg/l in the acid tank. However, after pumping the digestate from acid-tank to methane digester, Methane-forming bacteria used acetate as a substrate for producing methane. The result shown in Figure 4.16 indicates that acetic acid could be reduced to 4,319±3,574 mg/l in 85%FW15%RTL 1,527±501 mg/l in 90%FW10RTL 1,626±1,145 in 95%FW5%RTL 1,048±338 mg/l in 100%FW of methane tank in two-stage anaerobic digestion. It could say that lower complex molecule like rain tree leaf leads to lower acetic acid left in a digester.

Propionate was used by hydrogen producing acetate-forming bacteria to produce acetate, hydrogen, and carbon dioxide. In acid tank of the two-stage anaerobic digester was found very little of propionate compared with acetate. The concentration of propionic acid was around 5,720 to 8,599 mg/l in acid digester. In methane digester of two-stage anaerobic digester, propionic acid was greatly reduced to be in the range from 2,554±2,435 mg/l in 85%FW15%RTL 628±408 mg/l in

90%FW10%RTL 518±443 mg/l in 95%FW5%RTL, and 355±270.04 mg/l in 100%FW of methane digester.

Acetate and propionate concentrations of up to 6,000 and 3,000 mg/L shows no inhibition of the AD process in reactors treating manure (Ahring, 1995). Thus, the high acetate concentrations seen in the reactors under investigation may not have been problematic (Ahring, 1995).

Butyric, Isobutyric, Isovaleric and n-Veleric acid are longer chain fatty acids. More complex molecules were found in acid tank but after feeding to methane tank, it has been used and almost disappeared or it might not be detected due to pumping digestate from acid tank to methane tank that might dilute the volatile organic acid concentration.

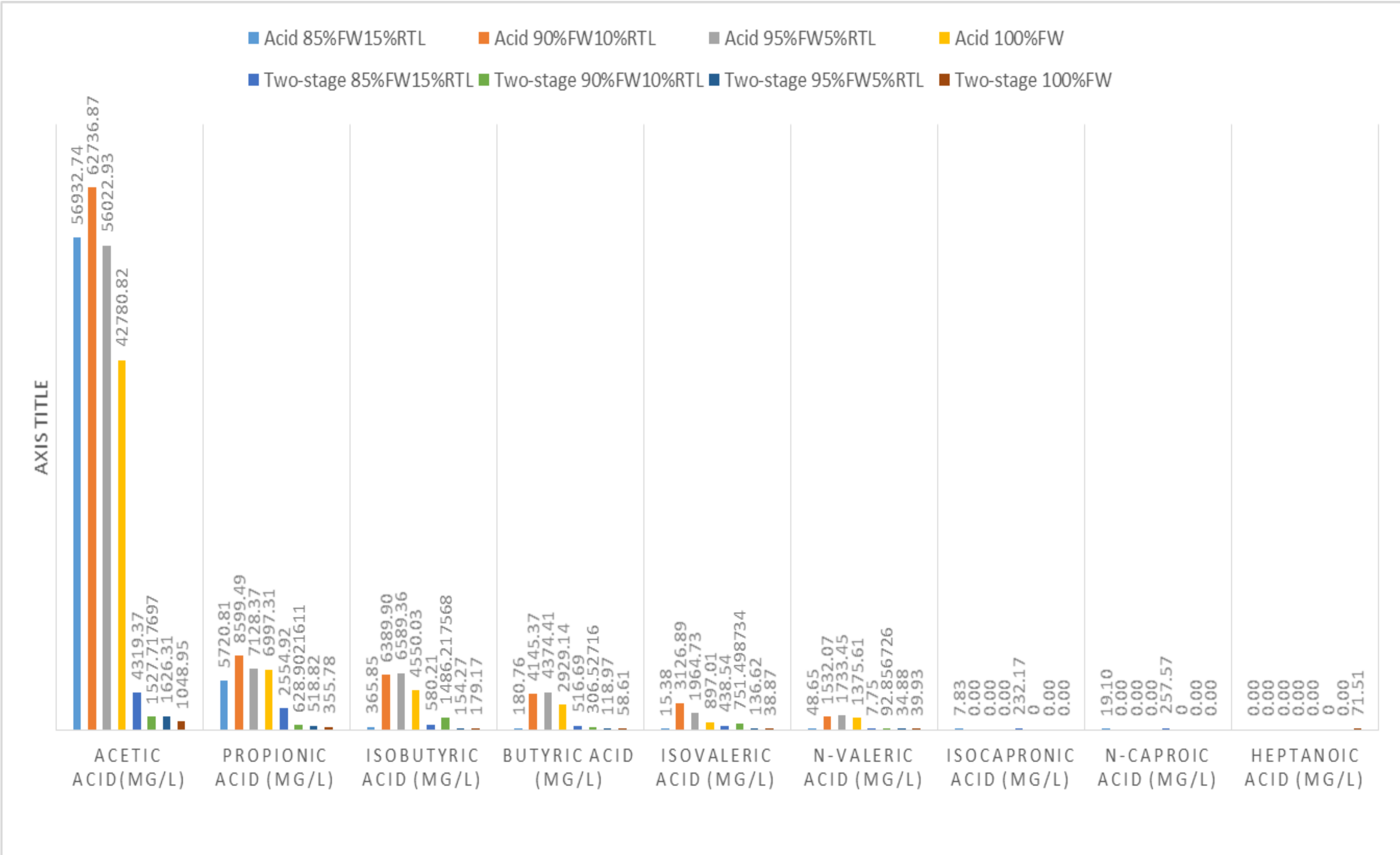


Figure 4.16 Volatile fatty acid in two-stage anaerobic digestion and acid tank

4.3.2.5 Ratio of propionate to acetate

The result in every ratio found that propionic to acetic acid were lower than 1.4, It has been recognized that ratio up to 1.4 can cause failure of digestion activity due to high content of propionic to acetic which is more complex for bacterial cell to use (Marchaim & Krause, 1993). The propionic to acetic acid ratios were 0.54±0.27 in 85%FW15%RTL, 0.38±0.20 in 90%FW10RTL, 0.28±0.10 in 95%FW5%RTL, and 0.32±0.15 in 100%FW. The lower propionic to acetic acid ratio meaning high in methane yield as 95%FW15%RTL and 100%FW have 61.03% and 62.47% methane yield.

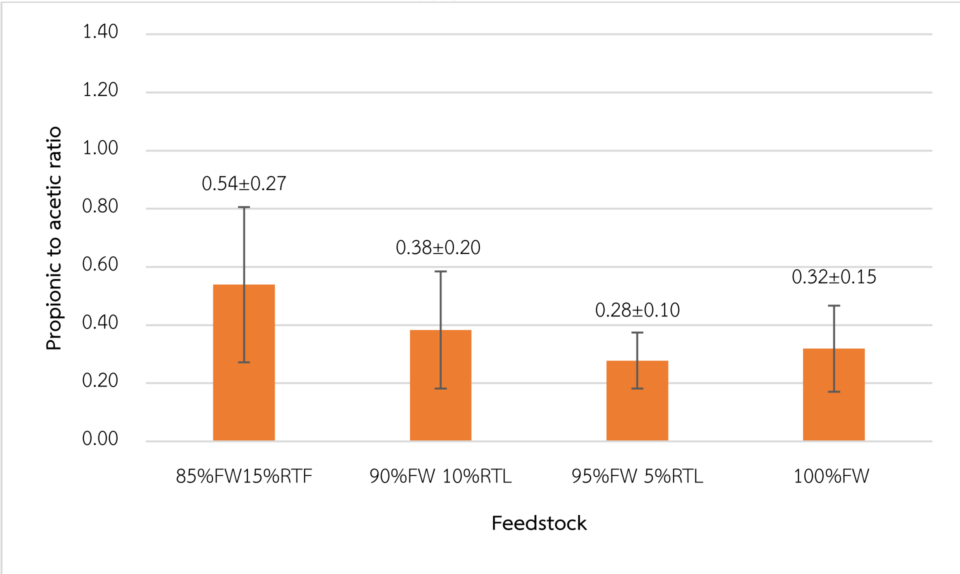


Figure 4.17 Ratio of propionic to acetic acid in two-stage digesters

4.3.2.6 Total alkalinity

This experiment did not control pH by adding any chemicals. Alkalinity balance was achieved by ammonium bicarbonate (NH_4HCO_3) from anaerobic digestion of organic matter to organic acid and ammonium (NH_4^+). After organic acid was used by methanogen to produce methane (CH_4) and carbon dioxide (CO_2). The leftover ammonium reacted with carbon dioxide to form ammonium bicarbonate (NH_4HCO_3). Alkalinity prevents fluctuations of pH in an anaerobic digester. The alkalinity of the two-stage digester dropped from 17,912±516 in 85%FW15%RTL 11,088±2,952 in 90%FW10%RTL, 9,618±654 95%FW15%RTL and 9,514±1,358 mg/L $CaCO_3$ when lower of rain tree leaf in the feed. Therefore, co-digestion of feed thereby leading to better process stability. This is in agreement with (Y. Zhang, 2012) that co-digestion help achieve better process stability. By the way, these values were obviously higher than 1,500 mg/L, which is commonly considered as the upper limit for allowing stable operation of biogas digester (Angelidaki I, 2005)

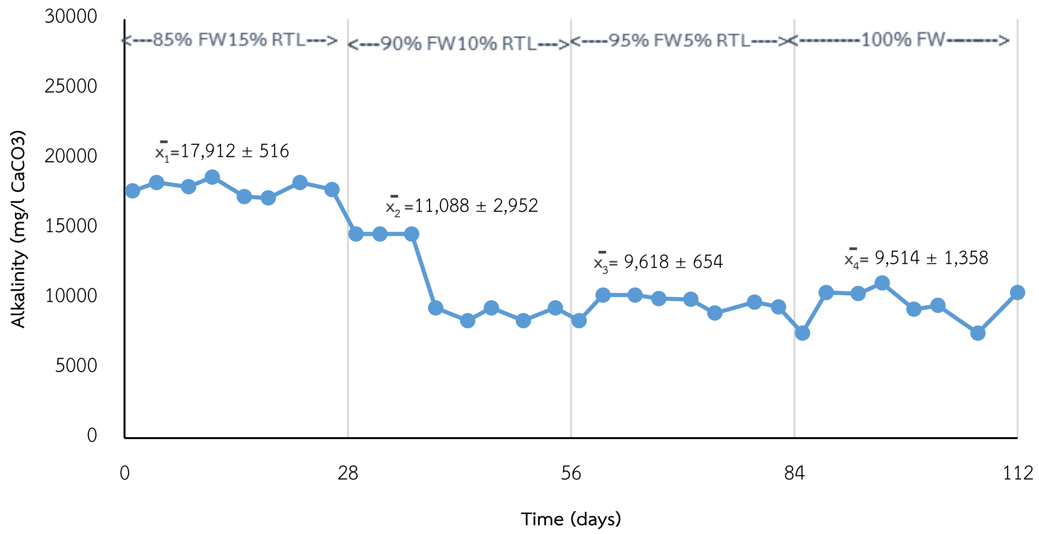


Figure 4.18 Total alkalinity in two-stage anaerobic digesters

4.3.2.7 Buffering capacity (VFA/Alk ratio)

To ensure digester stability in terms of pH, it is important to monitor the relationship between alkalinity and volatile fatty acids. A volatile fatty acid to alkalinity ratio (VFA/Alk) is an effective way to monitor digester stability. The results show that two-stage anaerobic digesters had VFA/Alk ratios lower than 0.4 which means that the systems had strong buffering capacity.

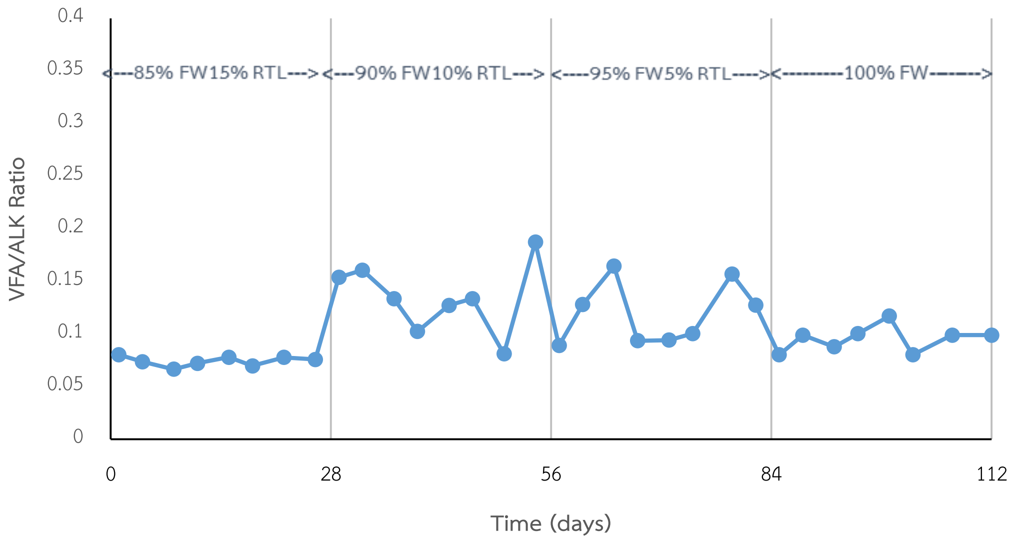


Figure 4.19 Buffering capacity of two-stage anaerobic digester

4.3.2.8 Total solid

Total solid of feed stock varied by 1st phase (85% FW 15% RTL), 2nd phase (90% FW with 10% RTL), 3rd phase (95% FW with 5% RTL) and 4th phase (100% FW). They were 222,949 ± 15,111, 204,121 ± 9,841, 176,462 ± 14,934, 130,308 ± 21,516 mg/l of total solid, respectively. After anaerobic digestion process took place, the total solid of two-stage were mostly reduced to average values of 32,970±2,596 mg/l of total solid, 30,960±8,573 mg/l of total solid, 56,616±8,179 mg/l of total solid and 54,796±7,863 mg/l of total solid, respectively. Calculation of percent total solid reduction were around 59 – 83%. The last two phase of two-stage (95% FW with 5% RTF and 100%FW) the result show in very high total solid content and high of error standard deviation due to the shape of two-stage anaerobic digester was vertical cylinder and installed digestate pipe under the reactor when the sludge precipitate, even mixing, the precipitate still come out lead to the high in total solid content in two-stage anaerobic digester.

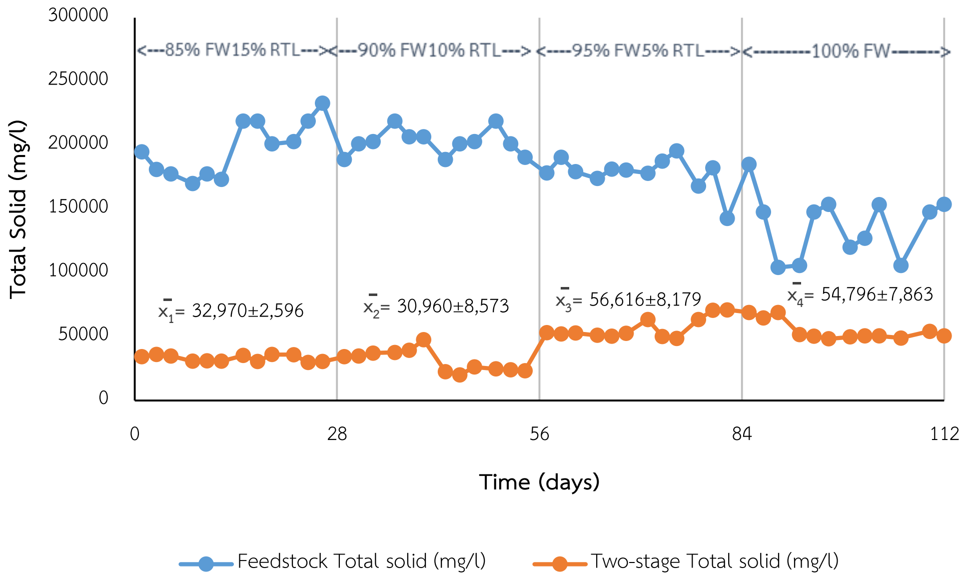


Figure 4.20 Total solid of feedstock and two-stage anaerobic digester

4.3.2.9 Total volatile solid

Total volatile solid of feed stock varied by first phase (85% FW with 15% RTL), 2nd phase (90% FW with 10% RTL), 3rd phase (95% FW with 5% RTL) and 4th phase (100%FW). They were 201,119±12,118, 204,121±8,586, 166,843±13,605, and 122,463±20,683 mg/l of total volatile solid, respectively. After anaerobic digestion process take place, the total volatile solid of two-stage digesters was mostly reduced 22,408±1,673, 20,538±7,730, 38,640±9,594, and 38,664±8,344mg/l of total volatile solid, respectively. The result show in very high total solid content and high of error standard deviation in 90%FW 95%FW and 100%FW due to the shape of two-stage anaerobic digester was vertical cylinder and installed digestate pipe under the reactor when the sludge precipitate, even mixing, the precipitate still come out lead to the high in total volatile solid content in two-stage anaerobic digester. Calculation of percent reduction of total volatile solid found that 85-87 percentage total solid were removed.

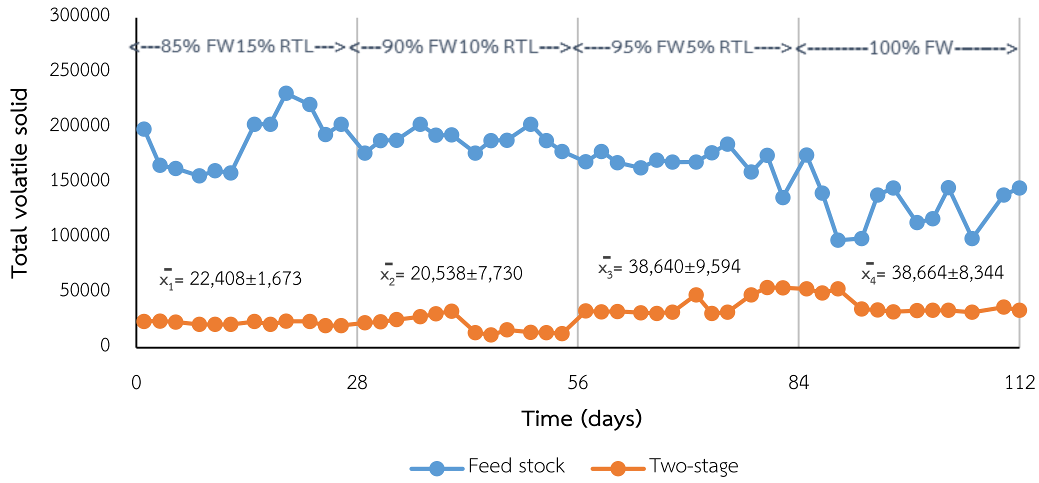


Figure 4.21 Total volatile solid in feedstock and two-stage anaerobic digester

4.4 Comparison of organic degradation and solid reduction between single-stage and two-stage anaerobic co-digestion systems

4.4.1 COD removal efficiency

The conversion rates were calculated based on the direct measurement of the total feed stock COD (tCODf), and the total digestated COD (tCODd). The result in Figure 4.22 shows that two-stage anaerobic digester has higher COD removal efficiency than the single stage anaerobic digester. As can be seen in Figure 4.23, it illustrates that after burning in furnace at 550°C for 20 minutes the digestate of single-stage anaerobic digester has a black ashes (incomplete combustion), which refers to some carbon source available in the digestate. The percent COD removal efficiencies of single-stage digester were 68.15±3.84 in 85%FW:15%RTL, 74.54±2.68 in 90%FW:10%RTL, 75.92±6.17 in 95%FW:5%RTL, and 81.99±3.59 in 100%FW. However, the percent COD removal efficiencies of two-stage digester were 79.67±3.77, 77.36±1.39, 77.34±3.49 and 77.55±6.03, respectively. It might be corrected to indicate that two-stage anaerobic digester can digest most of feedstock to methane gas as it has higher COD removal efficiency.

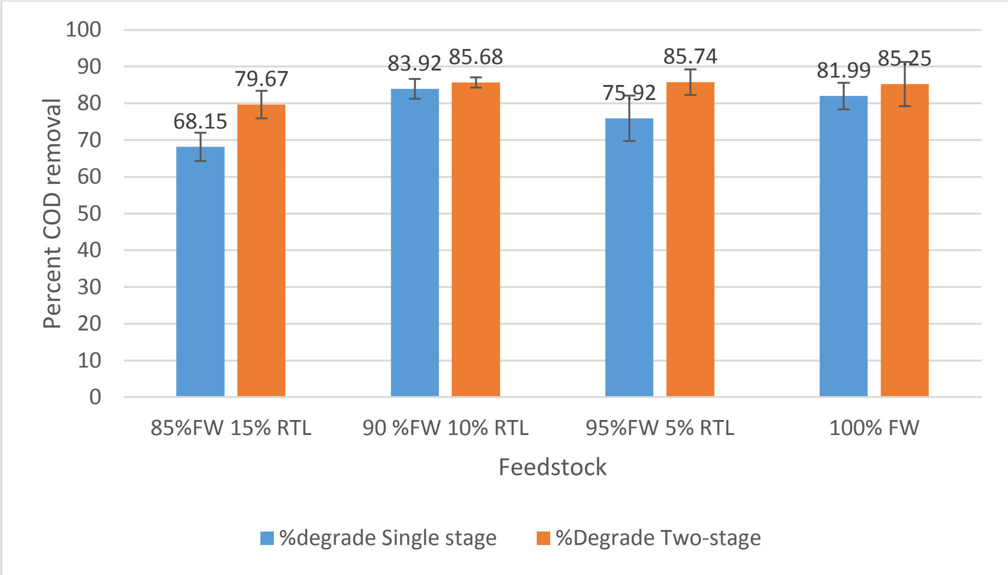


Figure 4.22 COD removal efficiency in single-stage and two-stage anaerobic digesters

Figure 4.23 Single-stage and two-stage digestate after burning in furnace at 550°C for 20 minutes

By comparing COD removal efficiencies of different ratios of co-substrate of food waste and rain tree leaf, the result show that without adding rain tree leaf, COD can achieve better COD removal efficiency.

4.4.2 Total solid reduction

Total solid reduction refers to a reduction of space when input high total solid waste as food waste in the digester and the digestate come out in liquid form. The result shows that total solid reduction percentages of single-stage digester with different co-substrate ratios of 85%FW:15%RTL, 90%FW:10%RTL, 95%FW:5%RTF and 100%FW were 80.44±3.00 ,81.70±1.74 ,77.56±3.078, and 73.54±3.96, respectively. However in two-stage digester, total solid reduction percentages of two-stage digesters were higher with 85%FW:15%RTL, 90%FW:10%RTL, 95%FW:5%RTF and 100%FW were 83.10±2.25, 84.72±4.04, 67.79±7.09 and 59.03±9.69, respectively. For the case of 100% FW, it shows the lowest total solid reduction efficiency. This could be concluded that co-digestion could improve the reduction of solid content. There was an error occurred with 95%FW and 100%FW in two-stage digester due to the design of sampling valve is under the tank so the leak of sludge came out lead to high in solid content.

Forster-Carneiro et al (2008) found that a high total solid (TS) content of feed waste could reduce substrate degradation, resulting in less methanogenic activity. In our study, the result shows similar trend that 85% food waste with 15% rain tree leaf having high total solid content produced less biogas.

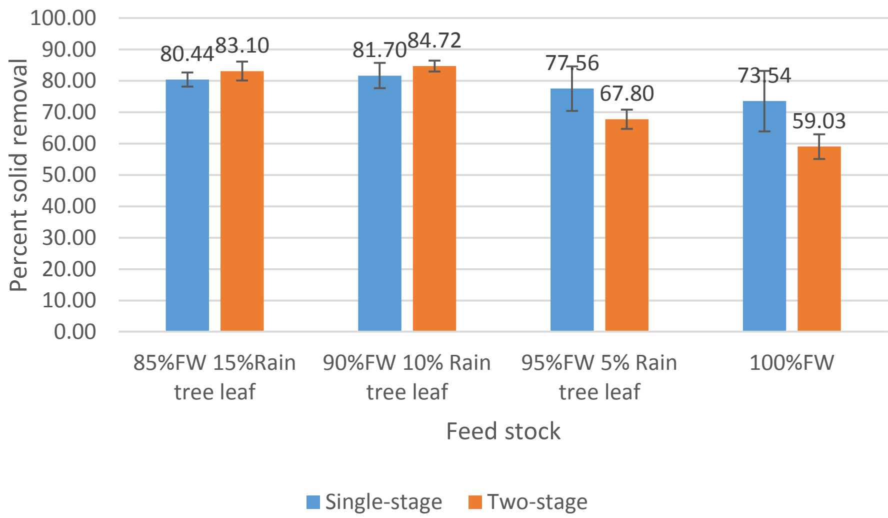


Figure 4.24 Solid removal efficiency of single-stage and two-stage anaerobic co-digestion

4.4.3 Total volatile solid reduction

The result shows that total solid reduction percentages of single-stage digester with 85%FW:15%RTL, 90%FW:10%RTL, 95%FW:5%RTF and 100%FW were 86.30±2.56, 87.96±1.5, 86.90±2.26, and 85.09±2.71, respectively. However in two stage digester, total solid reduction percentages with 85%FW:15%RTL, 90%FW:10%RTL, 95%FW:5%RTF and 100%FW were 87.91±1.71, 89.17±3.90, 76.69±7.29, and 69.33±8.80, respectively. There is no significantly different between single-stage and two-stage digesters for total volatile solid reduction with co-substrate ratios of 85%FW:15%RTL, 90%FW:10%RTL, whereas slightly lower TVS reduction were obtained with the two-stage digester when the higher food waste composition were applied.

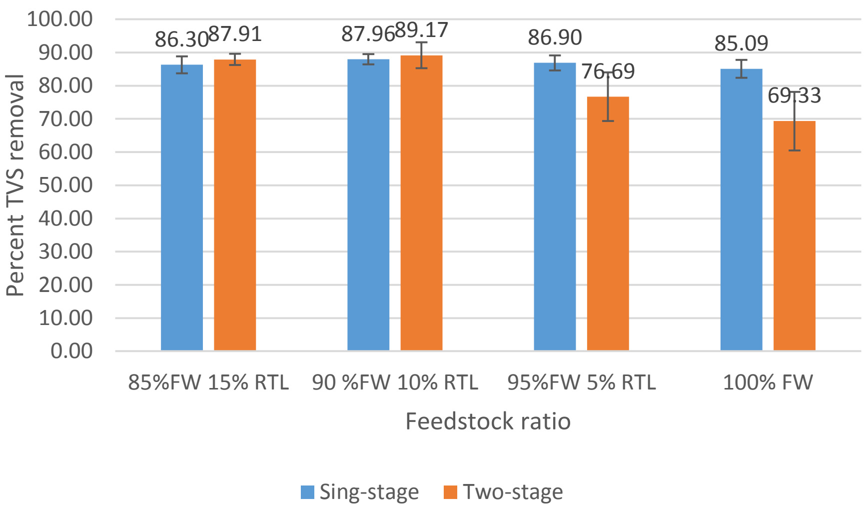


Figure 4.25 Total volatile solid removal efficiency of single-stage and two-stage anaerobic co-digestion

4.5 Biogas production

Biogas production was calculated for conversion of the biogas to Normal conditions as presented below. Fluctuation of room temperature and atmospheric pressure during the measurement of gas can contribute to errors in volume calculations. Therefore, to apply corrections, the record of change of atmospheric pressure and temperature is important.

The daily biogas production observed in this study was found that two-stage system has significantly higher biogas production than that of the single stage system at maximum biogas of 4.213 Nm^3 and 1.965 Nm^3, respectively as shown in Figure 4.26. The average biogas production was 1.216±0.373 Nm^3 in the single-stage system and 1.946±1.016 Nm^3 in two-stage system.

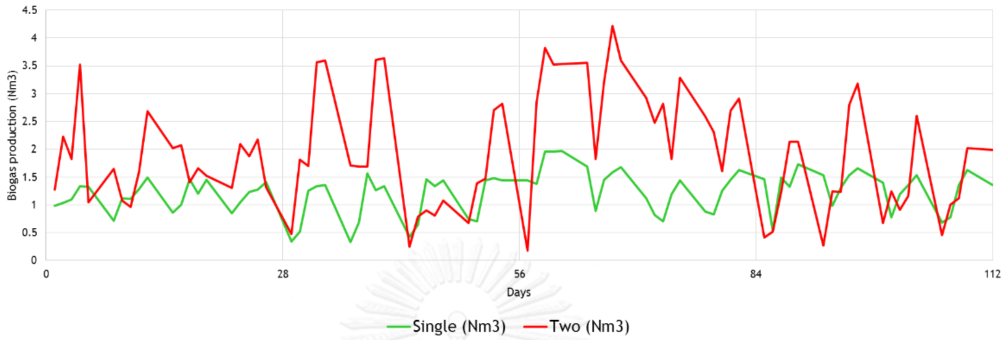


Figure 4.26 Biogas production rate of single-stage and two-stage anaerobic co-digestion

4.5.1 Accumulative biogas production

Accumulation of biogas production was plot in Figure 4.27, the results show that two-stage anaerobic digestion has higher biogas production activity. It can be plot into straight trend line with equation y = 1.4726x - 2.5571. However, single stage anaerobic digestion has lower slope and can also plot in straight trend line with equation y = 0.8771x - 1.1214

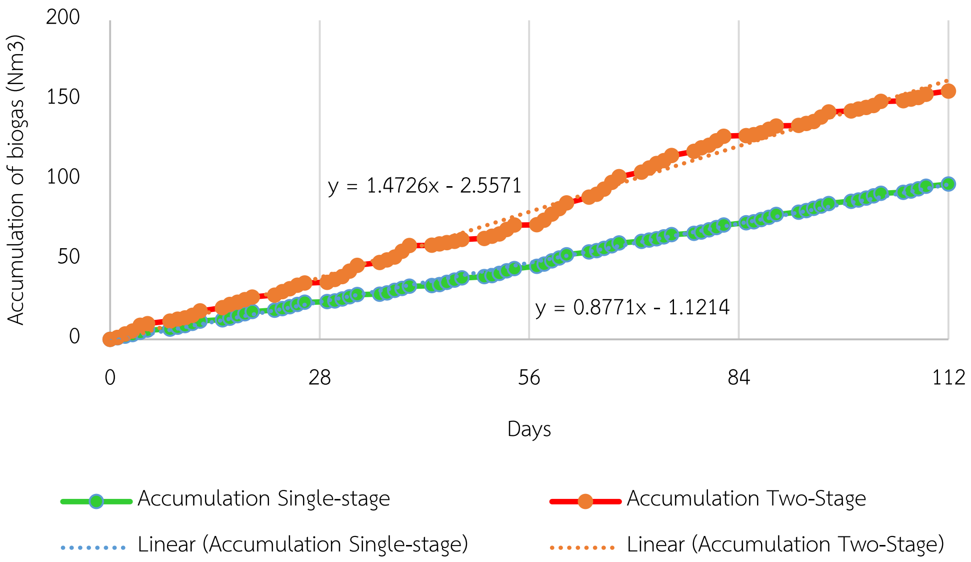


Figure 4.27 Accumulation of Biogas production in single-stage and two-stage anaerobic co-digestion

4.5.2 Comparison of biogas production in different feedstock

4.5.2.1 Biogas production in single-stage with different feedstock

Comparison of biogas production is done as in Figure 4.28 by the variation of co-substrate of food waste and rain tree leaf in a single-stage anaerobic co-digestion. The result was found in Figure 4.29 that the average biogas production in single stage became stationary phase within 2-3 days. Table 4.2 show that feedstock of 95%FW5%RTL has the highest average daily biogas production in stationary phase was 1.612 ± 0.094 Nm^3 ($p<0.05$). In 100%FW, the average daily biogas production in stationary phase was 1.514 ± 0.075 Nm^3. There were no significantly different in biogas production between 85%FW15%RTL and 90%FW10%RTL were 1.347 ± 0.102 Nm^3 and 1.376 ± 0.028 Nm^3. The result shows that co-digestion of food waste and rain tree leaf at 95%FW5%RTL which equal to C/N ratio 20.77 ± 4.23 was the optimal condition for highest biogas production in this experiment.

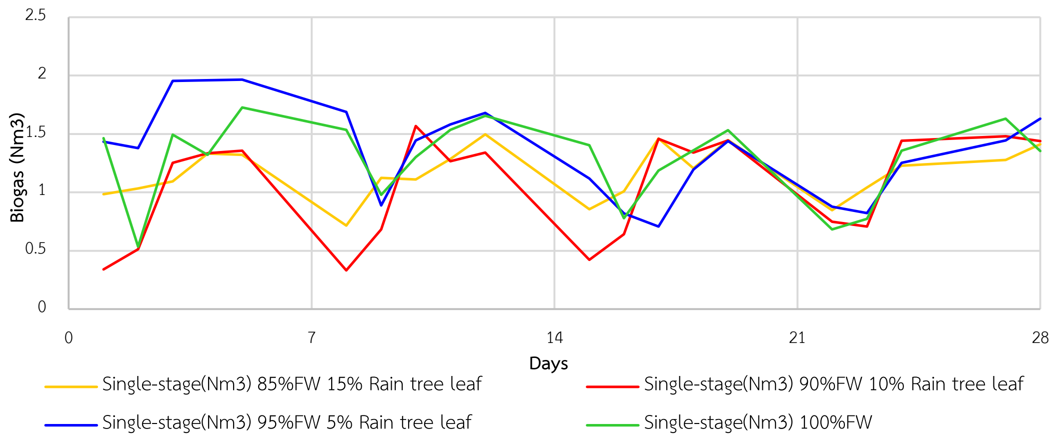


Figure 4.28 Biogas production in single-stage with different feedstock

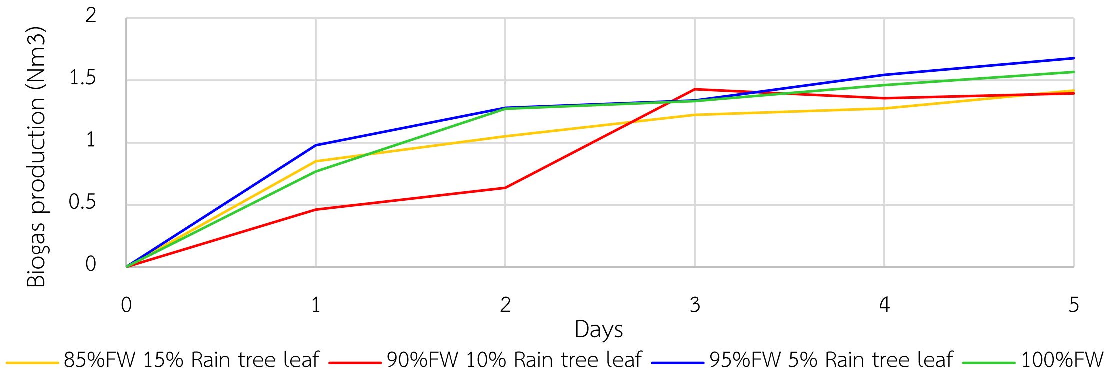


Figure 4.29 Average biogas production in stationary phase of single-stage with different feedstock

Table 4.2 Biogas production rate in single-stage with different feedstock

	85% FW 15% RTL	90%FW 10% RTL	95%FW 5% RTL	100% FW
Biogas production (Nm^3)	1.347 ± 0.102b	1.376 ± 0.028b	1.612 ± 0.094a	1.514 ± 0.075ab

4.5.2.2 Biogas production in two-stage co-digestion with different feedstock

Comparison of biogas production is shown in Figure 4.30 by the variation of co-digestion of food waste and rain tree leaf of a two-stage anaerobic digestion. The result was found in Figure 4.31 that the average biogas production in single stage became stationary phase within 3-4 days. It was found in Table 4.3 that the feedstock with 95%FW5%RTL has highest average daily biogas production in station phase of 3.194±0.189Nm^3. Follow by 90%FW10%RTL which was 2.726±0.077 Nm^3, then 100%FW which was 2.251±0.316 Nm^3. The lowest biogas production was found in the case of 85%FW15%RTL which was 2.051±0.264 Nm^3. The result shows that co-digestion of food waste and rain tree leaf at 95%FW5%RTL which equal to C/N ratio 20.77 ± 4.23 was the optimal condition for highest biogas production in this experiment.

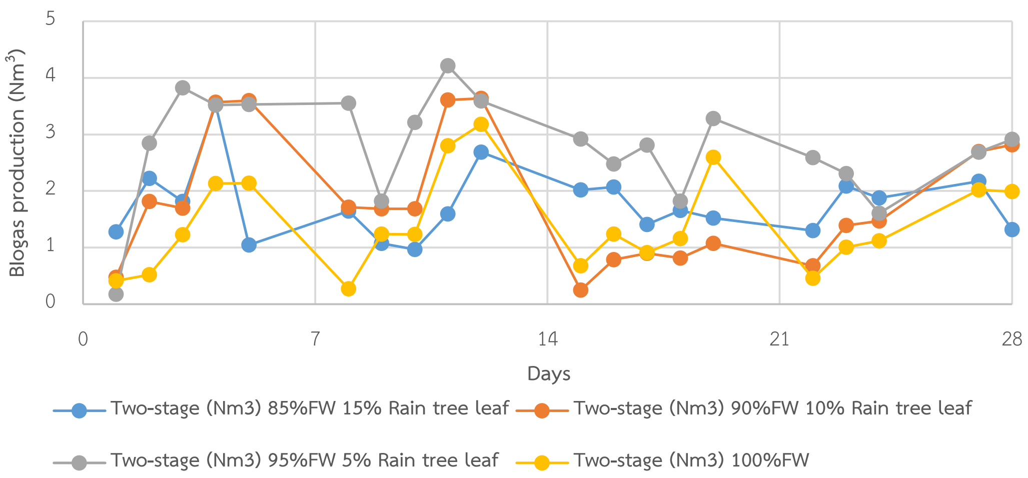


Figure 4.30 Biogas production in two-stage anaerobic digester with different feedstock

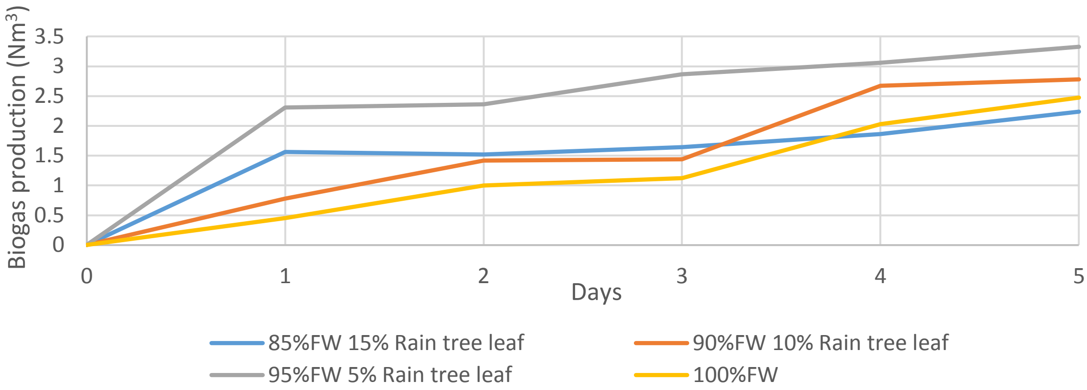


Figure 4.31 Average biogas production in stationary phase of single-stage with different feedstock

Table 4.3 Biogas production in two-stage anaerobic digester with different feedstock

	85%FW 15%RTL	90%FW 10%RTL	95%FW 5% RTL	100% FW
Biogas production (Nm^3)	2.051±0.264c	2.726±0.077ab	3.194±0.189a	2.251±0.316bc

4.5.2.3 Percentage of methane composition

The result shown in Table 4.4 indicates that percent methane composition is almost more than 50% of methane in every experiment. The two-stage anaerobic co-digestion process had higher methane composition which was averagely 57.51±6.95 in two-stage digester and 55.59±7.20 in single-stage digester, it might be that separation of acid reactor and methane reactor can digest most of acetate which is the intermediate substrate of methane-producing bacteria to produce methane. Both single-stage and two-stage co-digestion, the case with 95% to 100% of food waste have the highest methane composition.

Table 4.4 Percent methane composition

Feedstock	Single-stage	Two-stage
85%FW 15% RTL	50.84±2.88	59.71±2.44
90%FW 10% RTL	51.19±0.70	46.83±0.19
95%FW 5% RTL	53.75±2.03	61.03±1.98
100%FW	66.59±4.16	62.47±4.07

4.6 Composition of liquid digestate

The result shown in Table 4.5 presents that the digestate consists of organic matter 2.72 %w/w in single-stage and 2.74 %w/w in two-stage co-digestion process. Nitrogen content was found at 0.54% in single-stage and 0.40% in two-stage co-digestion. It has little content of phosphorus of 0.08% and 0.14%, respectively. Potassium content is at 0.18% and 0.12%, respectively. It has slightly content of sulfur of 0.02% and 0.04%, respectively. Since bacteria can use almost all the nutrient. This digestate can be used as a soil amendment to improve soil by adding bacteria and nutrients to soil.

Table 4.5 Composition of liquid digestate

Parameters	Single-stage	Two-stage
Organic Matter (%w/w)	2.72	2.74
Nitrogen (Total N) (%w/v)	0.54	0.4
Phosphorus (Total P_2O_5) (%w/v)	0.08	0.14
Potassium (K_2O) (%w/v)	0.18	0.12
EC (dS/m)	30.7	20.5
S (%w/v)	0.02	0.04

4.7 Biogas application to LPG

Single-stage digester uses electricity to rotate paddle inside digester around 0.2 kWh. For two-stage anaerobic digester, it needs more electricity to pump digestate from acid reactor to methane reactor and also to rotate paddle 0.3 kWh. 1 unit of electricity = 2.778 THB (Metropolitan-electricity-authority, 2000). If convert biogas into LPG 1 m^3 of biogas is equivalent to 0.45Kg of LPG (R. Ananthakrishnan, 2013) The price of LPG 15kg cylinder container equal to 320 THB

In single-stage digester which can produce less biogas can save money 28,036.38 THB per year. However, two-stage digester can save money 53,511.91 THB/year as can be seen from Table 4.6.

Table 4.6 Electricity consumption and LPG production from anaerobic digester

Detail		Single-stage	Two-stage
Expenditure	Electricity consumption (kWh/day)	0.2	0.3
	Electricity (THB/month)	16.669	25.0029
	Electricity (THB/Year)	200.02	300.04
Save money	Biogas production (m^3/day) 20kgs Food waste	8.06	15.36
	Biogas convert to LPG (kg per day) (1 m^3: 0.45 kg)	3.627	6.912
	LPG (THB/day) (1kg : 21.33THB)	77.36	147.43
	LPG (THB/Month) (1kg : 23.33THB)	2,320.91	4,422.98
	LPG (THB/Year)	28,236.40	53,811.95
Net saving	THB/Year	28,036.38	53,511.91

CHAPTER V

CONCLUSION

This study was done to compare the efficiency of anaerobic co-digestion systems for single-stage and two-stage anaerobic digesters with different feedstock ratios (85% food waste with 15% rain tree leaf, 90% food waste with 10% rain tree leaf, 95% food waste with 5% rain tree leaf and 100% food waste without adding rain tree leaf).

1) To operate the anaerobic co-digestion system that convert organic matters from food waste and rain tree leaf to biogas effectively, many parameters were considered. The different feed stock composition shows differences in C/N ratios which were 17.88 in 85% food waste with 15% rain tree leaf, 19.06 in 90% food waste with 10% rain tree leaf, 20.77 in 95% food waste with 5% rain tree leaf, 21.45 in 100% food waste. The more addition of rain tree leaf could decrease the C/N ratio due to high percent of nitrogen in rain tree leaf.
2) For biogas production efficiency, it was found that 95% food waste with 5% rain tree leaf has the highest biogas production efficacy in both single-stage and two-stage system which were 1.612 ± 0.094 Nm^3 in single stage and 3.194 ± 0.189 Nm^3 in two-stage. The biogas production efficiency could be affected by the different C/N ratio and the result shows that C/N ratio around 20.77 is the optimal condition for production of biogas. Co-digestion of food waste and rain tree leaf at 95%FW5%RTL helps in balance nutrient and also the texture of rain tree leaf in digestate could help in releasing biogas when mixing.

3) In degradation efficiency, the COD removal efficiency of single-stage was in the range of 68.15-81.99%. However, two-stage anaerobic digester has better removal efficiency since it has higher percentage of COD removal efficiency 78.67 – 85.64 %. Two-stage anaerobic co-digestion has higher COD removal efficiency since in anaerobic digestion there are two types of bacteria including in the system acid-forming bacteria and methane-forming bacteria.
4) Due to bacterial varieties is required in anaerobic digestion. To separate reactor in two-stage digester makes each phase in the suitable environmental conditions. In terms of total solid removal effiecieny, it was found that both of single-stage and two-stage has significantly high efficiency in reducing total solid. The result shows that anaerobic digestion could help in reducing waste in the environment.
5) For biogas production efficiency, in this study, two-stage anaerobic digestion system has significantly higher biogas production than that of the single stage system at maximum of 4.213 Nm^3 and and 1.965 Nm^3 with around 1.612 ± 0.094 Nm^3 organic waste-day in two-stage system while in single-stage, average biogas production was 3.194±0.189 Nm^3.
6) The digestate consists of organic matter 2.72 %w/w in single-stage and 2.74 %w/w in two-stage. Nitrogen 0.54% in single-stage and 0.40 in two-stage. It has little of Phosphorus of 0.08% and 0.14%, respectively. Potassium content was 0.18% and 0.12%, respectively. It has slightly content of sulfur of 0.02% and 0.04%, respectively. Since bacteria can use almost all the nutrient. This digestrate is not a liquid organic fertilizer, it can be a soil amendment.

In conclusion, the two-stage anaerobic digestion can achieve more biogas production according to the separation of digesters help acid-forming and methane-forming bacteria were in an optimal pH, higher volatile fatty acid were produced, then digested by methane-forming bacteria to biogas. The carbon to nitrogen ratio were also an important factor that can affect the bacteria growth. To balance nutrient, rain tree leaf were co-digested with food waste at 95%food waste to 5% rain tree leaf or C/N ratio of 20.77 showed the highest biogas production. Co-digestion can increase load of biodegradable organic matter which is good for waste treatment technology to reduce waste storage area. Not only treatment of waste, this study can support biogas for cooking in the canteen as well.

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APPENDIX

Table A1. Temperature and Pressure

Feedstock	Day	Temperature	Pressure
	1	30.5	1009
	2	27.5	1008.7
	3	29.3	1007.9
	4	28.9	1007.5
	5	27	1008
	8	28	1009.1
	9	29	1007.9
	10	29.6	1008.2
	11	29.5	1009.5
	12	29.2	1010.7
	15	28.8	1011.2
	16	30	1010.9
	17	30.2	1010.3
	18	31	1009.5
	19	30	1009.8
	22	29	1010.1
	23	30.5	1010.7
	24	29	1010
	25	29.4	1009.1
85	26	30	1009.6
	29	31.3	1007
	30	30.8	1008.9
	31	31.5	1008.4
	32	31	1010
	33	30.7	1011.3
	36	30.7	1010.7
90	37	31.5	1009.6

	38	30	1010.5
	39	30.1	1012.2
	40	30.6	1011.3
	43	30.2	1010.9
	44	31	1011.8
	45	31.3	1013.7
	46	29.8	1013.7
	47	30.5	1011.9
	50	28.8	1013.6
	51	25.9	1014.4
	52	26.7	1014
	53	28.2	1011.8
	54	28.3	1014.4
	57	31.8	1008.2
	58	30	1007.1
	59	30.1	1005.9
	60	32	1006
	61	32.3	1005.8
	64	32.1	1006.7
	65	31.4	1006.1
	66	31.6	1007.1
	67	32.6	1006.6
	68	31.5	1004
	71	30.9	1001.6
	72	32	1003.3
	73	31.6	1008.3
	74	31.8	1008.5
	75	30.3	1007.9
	78	30.8	1006.8
95	79	29.8	1007.4

	80	29.6	1009
	81	30.8	1009.9
	82	31	1007.9
	85	29.5	1008.9
	86	29.5	1008.4
	87	26.5	1009
	88	28	1011.1
	89	29.2	1011.8
	92	29.8	1012.9
	93	30.4	1013.3
	94	29.3	1013.9
	95	27	1014
	96	26.7	1013.2
	99	26	1013.7
	100	26.8	1014.2
	101	26.8	1015.3
	102	27.4	1014.5
	103	28.4	1012
	106	26.7	1012.2
	107	27.1	1010.6
	108	27.3	1009.7
	109	28.2	1010.4
100	112	29	1011

Table A2. Biogas production rate

Unit	System	85%FW15%RTL	90%FW10%RTL	95%FW5%RTL	100%FW
Nm^3/OLR_{kg}	Single-stage	0.116±0.022	0.106±0.044	0.136±0.040	0.128±0.035
	Two-stage	0.176±0.061	0.182±0.112	0.278±0.093	0.142±0.084
Nm^3/kg_{vs}	Single-stage	0.262±0.049	0.277±0.116	0.409±0.119	0.523±0.142
	Two-stage	0.396±0.138	0.477±0.295	0.833±0.278	0.578±0.343

Conversion procedures of biogas from Normal conditions to Standard conditions are presented below. Fluctuation of room temperature and atmospheric pressure during the measurement of gas can contribute errors in volume calculations. Therefore, to apply corrections, the record of change of atmospheric pressure and temperature is important. The gas pressure inside the tube collected over the liquid solution is the sum of the biogas pressure and the vapor pressure. The pressure of biogas, (Pbio) can be obtained by subtracting the vapor pressure of liquid (Pw) at the temperature of measurement from the pressure of collected moist gas (P).

$$Pbio = Pw - P$$

The produced biogas volume in normal condition can be converted to STP using Combine Gas law:

$$Vo = V \times \frac{To}{T} \times \frac{Pbio}{P}$$

V is the measured gas volume, V_0 is the volume of gas in standard temperature and Pressure, P_0 is the standard pressure, T is gas temperature at the time of measurement, and T_0 is the standard temperature. Modified (Buck, 1981) Equation can be suggested for the calculation of vapor pressure

VITA

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