A Novel Approach for Anaerobic Treatment of Food Waste

under Psychrophilic Temperature

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Abstract

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Food waste (FW) has become a significant concern because it can cause serious environmental pollution and greenhouse gas emissions, and hence investigating advanced methods to recycle food waste into energy and materials is of immense importance. Food waste has a high energy potential and has excellent biodegradability and high-water content. As a result, food waste can be treated by anaerobic technology that allows efficient energy recovery and reduces the environment's carbon footprint. However, in cold regions like Canada, mesophilic anaerobic treatment (30–35°C) can be energy intensive. Therefore, in this study a novel approach has been established to treat food waste at a lower operating temperature.

The objective of the study is to develop a sustainable system to treat food waste at a lower ambient temperature since climatic conditions in Canada are more suited for psychrophilic (1-20°C) temperature rather than mesophilic (30–35°C) anaerobic treatment. Low temperature has a negative impact on cellular processes during anaerobic treatment, making substrates unavailable to microbes. Hence, this study introduced a novel biogas recirculation strategy to overcome the thermodynamic constraints at lower temperatures.

Food waste was collected exclusively from the residential area of downtown Montreal near the Concordia campus for 3 months. The collected food waste sample was tested under anaerobic treatment conditions over a time period of 30 days to determine the biomethane generation potential of food waste at mesophilic (30–35°C) and psychrophilic (<20°C) temperatures. In order to investigate the feasibility of anaerobic treatment potential of food waste under psychrophilic temperature and evaluate the efficiency of biogas recirculation technique for enhanced biomethane production and system stability, batch tests were carried out on food waste in different trials. Anaerobic treatment potential of food waste under mesophilic condition was examined for comparison.

The impact of different substrate-to-inoculum ratios (0.5, 0.75, 1.0, 1.5, 2.0, and 4.0) and six different total solid percentages (5%, 10%, 12%, 15%, 18%, 20%) under mesophilic (30–35°C) and psychrophilic (1-20°C) temperatures were investigated for evaluating the optimum conditions for anaerobic treatment of food waste. Additionally, to evaluate the effect of particle size distribution on methane yield, food waste samples were generated using two different particle size distributions: (1) majority of particle sizes less than 2mm and (2) majority of particle sizes larger than 2mm.

Methane concentrations in the produced biogas were found to be 69%-94% under mesophilic anaerobic treatment, and methane concentration was observed between 68%-93% under psychrophilic anaerobic treatment with methane recirculation which was close to mesophilic treatment. The COD removal efficiency was found to be between 40% and 79% at mesophilic anaerobic treatment, while the removal of COD ranged from 34% to 82% for psychrophilic anaerobic treatment with biogas recirculation. A maximum reduction of 91% in volatile solids (VS) was observed under mesophilic anaerobic treatment with biogas recirculation. Furthermore, a

larger particle diameter has no major impact on methane production. Thus, the biogas recirculation approach eliminates the need for energy intensive mechanical particle size reduction.

In conclusion, the biogas recirculation technique at psychrophilic conditions has demonstrated improved methane generation and resulted in higher VS reduction and COD removal efficiency comparable to the results obtained from mesophilic anaerobic treatment. It is evident that applying anaerobic digestion systems with biogas recirculation under psychrophilic conditions could have significant potential in providing economically feasible and energy-efficient option for treating food waste in cold regions. However, further study will be required to develop a comprehensive guideline in this field.

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I would like to dedicate this thesis to my mother.

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List of Abbreviations

AD	Anaerobic Digestion
BMP	Biochemical Methane Potential
CH ₄	Methane
CO ₂	Carbon dioxide
COD	Chemical Oxygen Demand
FID	Flame Ionization Detector
FW	Food Waste
HRT	Hydraulic Retention Time
MSW	Municipal Solid Waste
OLR	Organic Loading Rate
SGP	Specific Gas Potential
SRT	Solid Retention Time
TCD	Thermal Conductivity Detector
TKN	Total Kjeldahl Nitrogen
TMP	Theoretical Methane Potential

TN	Total Nitrogen
TOC	Total Organic Carbon
TS	Total Solid
TVS	Total Volatile Solid
VFA	Volatile Fatty Acid

CHAPTER 1 INTRODUCTION

1.1 PROBLEM STATEMENT

Food waste (FW) has grown enormously in the past few years due to the global economy and population growth. It is estimated that approximately one-third of the food produced globally for human consumption, worth \$750 billion, is lost or wasted through the food supply chain (Ma & Liu, 2019; Slorach et al., 2019). Food waste (FW) composition and its physicochemical properties vary depending on the country, food consumption pattern, and cultural and economic aspects (Lin et al., 2013). FW contains 70-80% water and is highly biodegradable (Zamanzadeh et al., 2017). However, disposing of FW through landfilling, incineration, or composting can negatively impact the environment and contribute to global warming. In 2012, food waste was the largest contributor to municipal solid waste in the US, making up 21% of landfill waste (US EPA, 2014a). In 2014, the USA Environmental Protection Agency predicted that 12.7% of the waste disposed of is food waste (US EPA, 2014). A corollary drawback of landfilling food waste is that its energy value is lost in proportion to the fugitive emissions that contribute to greenhouse gases. Clercq et al. (2017) showed that considerable amounts of GHG, including methane (CH_4) and carbon dioxide (CO_2) , are produced when FW is disposed of in landfills. They showed that the emission of GHGs into the atmosphere contributes to global warming; methane is a potent GHG with a greenhouse effect that is 25 times more powerful than CO₂. Slorach et al. (2019) reported that global food loss and waste generate 6.7% of total anthropogenic GHG emissions annually. Another major environmental impact of FW is associated with the disturbance of the biogenic phases of phosphorus and nitrogen applied as fertilizers in agriculture (Papargyropoulou et al., 2014). Since food waste is a non-productive use of scarce resources (land, water, and fertilizer) and leads to

significant environmental degradation, appropriate management and treatment of FW have become the major concern of many countries worldwide (Gokarn & Kuthambalayan, 2017).

Anaerobic treatment of food waste can be the most promising cost-effective alternative for renewable energy production, environmental protection, and waste management (Ariunbaatar, 2014; Morales-polo et al., 2018; Posmanik et al., 2017). Several studies have already recognized anaerobic treatment as an easily applicable environmentally friendly technology for converting organic materials into renewable energy (Bachmaier et al., 2010; Ratanatamskul et al., 2014). Food waste can be a promising substrate for anaerobic treatment due to its moisture content and high energy (Cirne et al., 2007; Moriarty, 2013).

Anaerobic treatment is a biochemical process in which diverse communities of microorganisms transform insoluble and complex organic substrates into simple molecules such as CH₄ and CO₂ through numerous heterogeneous reactions and sequences of intermediates (Rivas García et al., 2020). The carbohydrate, protein, and lipid fractions of food waste can be fermented to long-chain fatty acids (LCFAs) and volatile fatty acids (VFAs) that are then converted into acetate and hydrogen gas, the substrates needed by methanogens. In general, there is a high content of proteins present in the household food waste, so usually there is no shortage of ammonium ions during the decomposition of food waste by anaerobic treatment. Although anaerobic technology has many benefits, such as decreased GHG emissions and high-quality renewable fuel production, the drawbacks associated with this process, such as relatively high capital costs, long retention time, and the required control of certain key parameters (e.g., pH, temperature, feed rate, alkalinity) limits its widespread implementation (Ariunbaatar, 2014).

Temperature plays a dominant role in anaerobic treatment because it strongly influences the performance of methanogenic and acid-forming microorganisms. Anaerobic treatment is classified

into three types based on temperature: psychrophilic (1-20°C), mesophilic (30-35°C), and thermophilic (50-60°C) (Zhang et al., 2014). However, several studies (Liu et al., 2009, 2017; Zhang et al., 2007) reported that enhanced methane generation could be achieved at higher temperatures and proposed that the high-temperature process would be the best option for the digestion of food waste (Kim et al., 2006). In major parts of Europe, the USA, and Canada climatic conditions are more suited for psychrophilic (<20 °C) rather than mesophilic (30 - 35°C) and thermophilic $(50 - 60^{\circ}C)$ treatment. Most of the research in anaerobic treatment is carried out either in the mesophilic $(30-35^{\circ}C)$ or thermophilic $(50-60^{\circ}C)$ temperature ranges even though substantial biomethane production occurs under cold conditions ($<20^{\circ}$ C), which is mediated by psychrophilic archaea. Hence, opting for anaerobic treatment under the mesophilic temperature range would require a significant amount of energy to maintain the reactor at high operation temperature, reducing the net energy yield and adding to the operating cost (Lettinga et al., 2001). On the other hand, under these climatic conditions, the psychrophilic anaerobic treatment appears to be an effective method for treating food waste with minimal energy requirements. However, anaerobic conversion of food waste at temperatures below 20 °C has attracted limited attention, and very little to negligible information can be found in the literature (Luo and Wong, 2019) since lower operational temperature reduces the micro-organism growth and methane production which can cause failure of the overall anaerobic system (Tiwari et al., 2021).

In this work, food waste was collected from the residential area within the downtown campus of Concordia University in Montreal. Food waste was subjected to anaerobic treatment under both psychrophilic and mesophilic conditions to assess the anaerobic treatment potential of food waste. In addition, a novel approach was followed to treat food waste under psychrophilic condition that exhibited similar efficiency to that of treatment under mesophilic conditions.

1.2 OBJECTIVES OF THE STUDY

1.2.1 General Objectives

The general objective of the present work is to develop a sustainable process to reduce carbon footprint by bioconversion of food waste into methane using the anaerobic treatment. The methane generated can be utilized as a source of heat, fuel, and electricity and reduce our dependency on conventional energy sources like fossil fuels to meet the ever-increasing energy demand. Food waste is one of the most common biodegradable wastes in Canada and North America. Instead of sending them to the landfill or large lagoons, they could be digested, creating energy-rich biogas that could be burned to produce combined heat and power. The fundamental hypothesis of this study is that treating biodegradable food waste in anaerobic treatment is more advantageous than conventional landfilling or composting. The two significant waste problems can be solved with the same waste-to-energy solution: anaerobic digestion. The research presented in this thesis deals with the anaerobic digestion of food waste produced in the urban environment. The primary purpose of this work is to examine the long-term psychrophilic bioconversion of food waste since it is the more feasible treatment option for a cold climatic region like Canada. This study also demonstrates the advantages deriving from psychrophilic treatment compared to the extensively used mesophilic process.

1.2.2 Specific Objectives

The specific objectives were designed to find the optimum operating conditions for the anaerobic treatment of food waste through experiments under mesophilic and psychrophilic temperatures. The study also aimed to investigate the effectiveness of a novel approach for bioconversion of food waste under psychrophilic condition. The objectives of the study can be summarized as follows.

1) To determine the favourable conditions in which the anaerobes thrive for the successful operation of the anaerobic treatment process at both mesophilic and psychrophilic treatment.

2) To assess food waste's characteristics and biodegradability to enhance the biodegradation process.

3) To evaluate and compare different operational parameters of the anaerobic system under mesophilic and psychrophilic temperatures.

4) To determine methane yield and compare the performance of anaerobic system technology under mesophilic and psychrophilic anaerobic treatment.

5) To investigate the impact of biogas recirculation on the bioconversion of food waste.

1.3 ORGANIZATION OF THE RESEARCH STUDY

This thesis is organized in the following chapters:

Chapter 1: Statement of the problem and objectives of the research.

Chapter 2: Critical review of the literature on the food waste treatment with theoretical background of anaerobic treatment.

Chapter 3: Characteristics of the examined food waste, experimental methodology and analytical methods.

Chapter 4: Presentation of the experimental results obtained under various operating conditions related to the food waste treatment, COD removal and generation of methane, and discussion of the findings.

Chapter 5: Overall conclusions of the research work and recommendations for future work.

Reference: List of references used in this study.

Appendices: Reference curve for the compositional analysis of biogas.

CHAPTER 2 LITERATURE REVIEW

Urban food waste (FW) is of paramount importance because it is generated in significant quantities at a relatively stable rate year-round, thus making it a reliable renewable energy source. According to the Food and Agriculture Organization (FAO, 2011) of the United Nations, currently the global volume of food wastage amount is about 1.6 billion tons per year, with a carbon footprint estimated to be 3.3 billion tons of CO₂ equivalents. Less than three percent of the food waste was separated and treated, primarily through composting, and the rest was disposed of in landfills. Due to increasing needs for renewable energy generation and diversion of organic residuals from landfills to reduce the greenhouse gas emissions and other environmental impacts, treatment of food waste using anaerobic digestion technologies has become a more attractive method for food waste management. Food waste, a major component of municipal solid waste, consists of rice, meat, fruits, vegetables, bones, oil, and inert substances (Hafid et al., 2017). The growth rate of food waste is gradually increasing with the development progress of economic and population (O'Connor et al., 2021). Food waste is a suitable substrate for anaerobic digestion since it is enriched in biomass, carbon, moisture and is generally biodegradable (Ajay et al., 2021). Anaerobic digestion also has a lower global warming footprint than composting and landfilling (Edwards et al., 2018). Moreover, anaerobic digestion is optimal to treat organic waste since it has a low life cycle cost in aspects of economic sustainability (Lee et al., 2020). Therefore, anaerobic digestion treatment of food waste is promising in the context of fossil fuel exhaustion and opportunities for both resource and energy recovery.

2.1 FOOD WASTE DEFINITION AND CHARACTERISTICS

All substances that enter the food supply chain but are not consumed represents food waste, i.e., both edible and inedible materials. Food waste is defined by the European Parliament as: "Food intended for human consumption, either in edible or inedible status, removed from the production or supply chain to be discarded, including at primary production, processing, manufacturing, transportation, storage, retail, and consumer levels" (EU Parliament, 2017). Food loss can occur at all stages of production, from pre-harvest on the farm through to post-harvest losses during processing, distribution, retailing and consumption (Food Waste Reduction Alliance, 2016). By far the largest proportion of this material is generated at the point of consumption, in the home or in cafeterias, canteens and restaurants (Parfitt et al., 2010). Some of this waste is avoidable, but a proportion is unavoidable as it consists of parts of the product that are not edible (such as shells, bones, and peels). The development of food waste hierarchies, where prevention is the main goal and only material that is unsuited for human or animal use becomes waste, is the result of a great understanding of the origins and destinies of unconsumed food. On average, developed countries generate around 100–170 kg of food waste per capita per year, more than twice that of developing countries (Dung et al., 2014). Some developing countries, such as China and India, also have high challenges in food waste disposal due to the large total population. In developing countries, 80-90% of food waste is generated in the early stage of the food supply chain, mainly as pre- and postharvesting residues, and food processing waste (FAO, 2011). However, in developed countries (Europe and North America) more than 40% of the food (about 222 million tons) is lost or squandered during the retail and consumer stages, which is almost as much as the total net food production in Africa (230 million tonnes) (FAO, 2011).



Figure 2.1 Framework defining the food supply chain and food waste destinations (JRC, 2017)

2.1.1 Food Waste Hierarchy and Source Separated Municipal Solid Waste

Food waste reflects socioeconomic conditions as well as other variables. Although it is still imperfectly characterized, the amount of food products in the municipal waste stream can range from 25 to 65%, depending on the region (Banks et al., 2018). Although the appearance of food waste may differ depending on its origin, due to local food preferences and habits, in biochemical terms it is generally very similar. It is readily biodegradable, has a high biochemical methane potential (BMP), and exhibits about the same distribution of proteins, lipids, carbohydrates, and necessary components. In 2010, Parfitt et al. (2010) reported that the composition of food waste and the current global food waste database were both very unknown. By far the largest portion of food waste comes from household consumption. Food waste from food production, food in the food supply chain that has lost all its value, by products, peelings, scrapings from preparing meals,

rotten food, and uneaten leftovers make up about 90% of all food waste (Banks et al., 2018). It is now well recognized that food waste components can be categorized as unavoidable or avoidable, with an additional category of possibly or partly avoidable being used in some circumstances. Residues and by-products from food production, such as inedible peels or seeds, make up the first category of unavoidable or inedible food waste. Unused food, frequently thrown out as a result of passing the "best before" date or overspending, or partially consumed foods, such as leftovers from meals, make up avoidable food waste. The possibly or partly avoidable category has been defined as "food and drink" that some people eat, and others do not (e.g., bread crusts), or that can be eaten when a food is prepared in one way but not in another (e.g., potato skins)" (WRAP, 2009).



Figure 2.2 Food waste hierarchy. Left – based on WRAP (2017), Right – based on US EPA (ND) (2018)

From the viewpoint of renewable energy production through anaerobic digestion of food waste, with beneficial use of the digestate, the most prominent features of the collection system appear to be what it accepts, and what type of container is used for collection (large or small). Small container collections typically have very low levels of contamination, which can reduce the need for pre- and post-processing and the energy consumption associated with it. Even a basic AD plant might be able to generate a high-quality output with source separated collection mechanism (VALORGAS, 2012a). The collection of source separated food waste in paper bags, vacuum transport from the kitchen sink to a central grinder with tanker collection of solids and disposal of the supernatant to the sewer were the four different systems that were compared in a study by Bernstad and la Cour Jansen (2012). Edwards et al. (2016) developed an energy and time model for curbside waste collection, which was verified and used to model a set of scenarios for introduction of source separated food waste collections. The results suggested an increase of up to 60% in fuel consumption depending on the collection system adopted.

2.1.2 Characteristics and Composition of Food Waste

FW is characterized by complex components and organic material. There are several types of FW such as fruit and vegetable waste, household and restaurant FW, brewery waste, and dairy waste (Xu et al., 2018). Food waste can differ significantly in visual appearance, even for materials collected from similar sources in close proximity. Taking source separated domestic food wastes as an example, the composition of FW varies based on geographical changes, seasonal changes, cooking procedures, eating habits, age distributions, family sizes and consumption patterns (Meng et al., 2015; Xu et al., 2018). Coupling these variations with possible differences in attitudes towards how their wastes are managed and in purchasing habits. FW consists of various organic components such as proteins, carbohydrate polymers (starch, cellulose, hemicelluloses, and lignin)

lipids, and organic acids. Fisgativa et al. (2016) studied 102 different FW samples and reported that the characteristics of FW displayed a high coefficient of variance (CV). They indicated that the variations of 24% of the studied characteristics were described by geographical change, seasonal change, and the type of collection source. They observed that FW has an average pH of 5.1 (CV 13.9%), carbon and nitrogen ratio of 18.5% (CV 31.8%), 36% of carbohydrates (CV 57.2%), 26% of protein (CV 62.2%), 15% of fats (CV 52.0%), and biomethane potential of 460.0 mL CH₄/kg VS (CV 19%). FW is a readily biodegradable organic substrate because of its large quantity of moisture content (Zhang et al., 2014). Meng et al. (2015) and Xu et al. (2018) indicated that carbohydrates and proteins have a higher hydrolysis rate due to their rapid degradability compared to lipids. Thus, quickly degradable carbohydrates and lipid-rich food wastes can produce high methane yields. Bong et al. (2018) reported that fruit and vegetable waste has low lipid content but relatively high cellulosic content, whereas FW and kitchen waste have high lipid content because of the presence of animal fat and oil. Studies have reported that fruit and vegetable waste had a lipid content of 11.8%, whereas FW and kitchen waste were reported to have 33.2% and 21.6% of lipid content, respectively (Wang et al., 2014; Yong et al., 2015). However, high lipid contents can cause system failure due to the formation of long-chain fatty acids. This occurs when the mass transformation of soluble organics into bacteria decreases due to the destruction of the cellular membrane (Leung & Wang, 2016). Li et al. (2017a) stated that FW rich in carbohydrate content would affect the carbon and nitrogen ratio (C/N), and thus, nutrient restrictions and quick acidification could occur due to the increased organic matter. The biochemical and physiochemical analysis of source separated food waste found in several studies are summarized in Table 2.1. Figure 2.3 represents the municipal solid waste composition in North America and Figure 2.4 is food waste composition from a typical collection facility in Canada.



Figure 2.3 Annual MSW composition in United States (U.S. Environmental Protection Agency, 2008)



Figure 2.4 FW composition from a typical collection scheme (Canadian Biogas Association, 2018)

Source	North	UK	China	S. Korea	South Asia			
	America							
Basic Characteristics for AD								
pH	5.4 ± 0.5	5.02 + 0.01	4.2 ± 0.2	6.5 + 0.2	5.6 ± 0.6			
Total Solid TS (% fresh	30.90 ± 0.07	25.89+0.01	23.1 ±0.3	18.1 ± 0.6	24 ±1.5			
matter)								
Volatile Solids VS (%	26.35 ± 0.14	24.00+0.03	21.0 ± 0.3	17.1 ±0.6	23 ±0.8			
fresh matter)								
VS (% TS)	85.30 ± 0.65	92.70+0.12	90.9	94 ±1	94			
Total Organic Carbon		48.76+0.87	56.3 ±1.1	-				
TOC (%TS)								
Total Kjeldahl Nitrogen		7.53+0.13	5.31	5.42 ± 0.26				
TKN (g/ kg fresh matter)								
Biochemical analysis on a V	VS basis (g/ kg '	VS)						
Carbohydrate		458+14	420	653.2 ± 36.2	550			
Lipid		149+1	364	136± 3	330			
Protein		197+4	186	192 ±8	172			
Hemi-cellulose		88.6+1.2	-	-				
Cellulose		66.1+0.1	109	-				
Lignin		21.7+0.12	-	-				
Nutrition on a TS basis (g/k	g VS)							
Nitrogen (TN)		29.1+0.5	23 3	29.9 1.4				
Phosphorus(P)	5.2 ± 0.8	2.82+0.13	1.49	8.23 0.50	1.49 ± 0.09			
Potassium(K)	9.0 ±1.1	8.59+0.27	23.0 ±0.4	6.83	2.3 ± 0.04			
Potentially toxic elements of	n a TS basis (m	g/kg TS)						
Cadmium (Cd)	< 3	<0.05	-	0.29				
Chromium (Cr)	10+3	4.21+0.62	<1	2.1	0.17			
Copper (Cu)	100+3	4.69+0.82	3.06	38.3	31			
Mercury (Hg)			-	-				
Nickel (Ni)	$\frac{-}{6\pm 3}$	$\overline{2.8+0.1}$		2.4	2.0			
Lead (Pb)	13 ±10	<0.6		2.3				
Zinc (Zn)	250 ± 70	22.4+0.80	693 ± 130	103	160			
Essential trace elements (m	g/ kg TS)		070-100	100	100			
Cobalt (Co)	<u> </u>	0.15+0.03		<0.4				
Iron (Fe)	2480 ± 1300	111+1	433 ± 100	39.6	766 ± 402			
Manganese (Mn)	190 + 100	86.5+2.5	476+411	12	100-102			
Molybdenum (Mo)	-	2.8+0.6	-	0.31				
Tungsten (W)	_	-	1_	-				
Selenium (Se)	_	0.42+0.20	1_	_				
$\begin{array}{c c c c c c c c c c c c c c c c c c c $								
Nitrogen (N)	3 16 +0 22	2 91+0 05	3 16+	3 54	23+03			
	5.10 -0.22	2.91 0.05	0.22	5.51	2.3 ± 0.3			
Carbon (C)	46.78±1.15	48.8+0.9	56.3±1.1	46.67	56.3±1.1			
Hydrogen (H)	-	6.37+0.19		6.39				
Sulphur (S)	0.81 ±0.03	-	0.33	0.33	8.6			
Oxygen (O)		34.7+0.9	-	36.39				

Table 2.1: Physio-chemical and biochemical analysis of FW reported in the literature.

From VALORGAS (2011); Zhang et al. (2007); Zhang et al. (2013) and Shen et al. (2013); Zhang et al. (2011)

2.1.3Biodegradability, Bioavailability and Bio-accessibility

Biodegradability, bioavailability, and bioaccessibility have all been shown to be crucial factors in defining organic matter biodegradation (Jimenez et al., 2014). Biodegradability is the ability of a substrate to be broken down by a microorganism into simpler compounds, but this biodegradation is limited by molecule's bioavailability, complexity and/or toxicity. According to Aquino et al. (2008), bio-accessibility is the degree to which a molecule may be accessed dependent on a few variables, such as the length of time that the substrate is in touch with bacterium. Thus, there is a concept of physical accessibility, such as when cellulose is protected by lignin or vegetal walls of barrier and needs to be broken down physically or chemically in order for microbes to access it (Motte et al., 2014; Reilly et al., 2015). Consequently, the bioavailable organic matter is included in the bio-accessible fraction as the organic fraction is able to be degraded by secreted exo-cellular enzymes (Jimenez et al., 2014). Due to the rapid expansion of the global economy and population over the past few years, there has been a significant increase in food waste and loss. It is estimated that approximately 33.3% of food produced globally for human consumption is discarded annually, with 76.3% landfilled (Ma and Liu, 2019; Slorach et al., 2019, USEPA, 2016). Due to the population growth and increasing urbanization, it is anticipated to keep expanding. Moreover, FW is generally an acid or sub-acid substrate, which is suitable for biodegradation but sub-optimal for methanogens that operate at a slightly higher pH (6.5–7.2) (Fisgativa et al., 2016). Generally, taking into consideration that moisture plus total solids (TS) contents represent 100% of wet weight (wet basis) (wb), FW moisture content is of 70-80% wb and TS are around 20-30% wb, 90% of which are volatile solids (VS) (Zhang et al., 2014). Organic material from plants and animals used as food is by its very nature easily digestible in the relatively uncomplicated human alimentary canal. Our food contains very little lignin, and much of our fiber intake has been milled

in the food preparation process. After 40 days of retention, Heo et al. (2016) investigated the biodegradability of traditional Korean food waste made up of boiled rice (10%-15%), vegetables (65%–70%), meat and eggs (15%–20%), and methane yield of 0.49 L/g VS at 35°C. Hence, it is expected that food waste from homes and restaurants can be easily digested in an AD treatment plant with only the reduction of particle size as a pre-treatment. In a batch treatment test with thermophilic conditions (50°C) after 28 days, Zhang et al. (2011) examined the nutrient content of food waste from a restaurant and demonstrated that the waste contained the right nutrients for AD microorganisms. They also reported a methane yield of 0.44 L/g VS of waste. FW has a low C/N ratio and is mostly made up of easily digestible carbs (50–60% TS), proteins (15–25% TS), and lipids (13–30% TS) (Zang et al., 2007). Additionally, it lacks trace elements but is abundant in macro- elements (Banks et al., 2018).

2.1.4 Methane Potential of Food Waste from Different Sources

Theoretical Methane Potential (TMP)

Biochemical and elemental compositions can be used as a basis for prediction of the actual and maximum theoretical methane potential of a feedstock, in some cases providing a reality check on quoted methane yields (Weinrich et al., 2018). In particular, these values usually provide a reasonably good estimate of the biochemical methane potential (BMP) for food waste. The elemental composition can be used in conjunction with the Buswell equation (Symons & Buswell, 1933) to calculate the maximum theoretical methane potential of the feedstock as shown in equations 1 and 2, assuming all components are converted at standard temperature and pressure (STP). This provides an upper bound for the methane yield (Angelidaki & Sanders, 2004).

$$C_{n}H_{a}O_{b}N_{c} + \left(n - \frac{a}{4} - \frac{b}{2} + \frac{3c}{4}\right)H_{2}O \rightarrow \left(\frac{n}{2} + \frac{a}{8} - \frac{b}{4} - \frac{3c}{8}\right)$$
$$CH_{4} + \left(\frac{n}{2} - \frac{a}{8} + \frac{b}{4} + \frac{3c}{8}\right)CO_{2} + cNH_{3}$$
(1)
$$TMP\left(\frac{mL CH_{4}}{g VS}\right) = \frac{22.4 \times 1000 \times \left(\frac{n}{2} + \frac{a}{8} - \frac{b}{4} - \frac{3c}{8}\right)}{12n + a + 16b + 14c}$$
(2)

The high biodegradability of food waste means that a relatively high proportion of this theoretical yield can be achieved, and the biogas composition is typically close to the predicted value. Theoretically, lipid-rich waste is considered to be highly attractive for AD due to the methane potential of lipids (1.014 m³/kg VS), which is much higher than that of proteins (0.74 m³/kg VS) and carbohydrates (e.g., 0.37 m³/kg VS for glucose) (Zupancic and Jemec, 2010; Kim et al., 2004). Another high-methane yield substrate is household and restaurant food waste, which has relatively high lipid contents and a balanced nutrient composition as well. Carbohydrates and proteins are generally considered to be rapidly degradable, while lipids have lower hydrolysis rates (Mata-Alvarez et al., 2000). Thus, food wastes rich in lipids (e.g., used oil, ice cream) and easily degradable carbohydrates can achieve high methane yields (Labatut et al., 2011; Meng et al., 2015). In contrast, food waste with a high lignocellulosic fraction and low lipid content, such as fruit and vegetable residues and brewery waste, have lower methane potentials of about 0.16–0.35 m³/kg VS. Table 2.2 provides the theoretical methane yield from different biochemical components of food waste.

Substrate	Typical Composition	Methane Yield [L CH4/g VS]	CH4 [%Vol]
Carbohydrate	C ₆ H ₁₀ O ₅	0.415	50
Simple sugars	C ₆ H ₁₂ O ₆	0.373	50
Lipid	C57H104O6	1.013	70
Protein	C ₅ H ₇ NO ₂	0.495	50
Cellulose	C ₆ H ₁₀ O ₅	0.415	50
Hemicellulose	variable	0.424	50

Table 2.2: Typical methane yields from biochemical component of food waste (Angelidaki and Sanders, 2004)

Biochemical Methane Potential (BMP)

The biochemical methane potential is investigated by a test performed under standardized conditions, which is used to assess the potential biogas of a biomass. It represents the maximum producible biogas independent of the conditions adopted in full-scale applications. Different, approaches have been proposed in the past to measure BMP. Regardless of the method used, all of the tests are quite comparable, allowing for comparison of the outcomes. Tests are generally performed in batch form under mesophilic conditions using an inoculum to avoid inhibition due to VFA accumulation and pH drop. The Hydraulic Retention Time (HRT) can be varied from 12 to 65 days. Data obtained from BMP tests indicated some variations, mostly as a result of various food waste contents. Table 2.3 summarizes the compositions and methane potentials of some typical food waste streams. The highest methane potential of food waste is 0.3–1.1 m³ CH₄/added VS, which is generally higher than other AD substrates such as lignocellulosic biomass, animal manure, and sewage sludge (Mao et al., 2015). The differences reported for BMP within the

investigated FW (Table 2.3) are due to the differences in composition in terms of carbohydrates, protein, and lipids and their relative ratios. Among all types of food waste, fat, oil, and grease (FOG) achieved the highest methane yield.

Source	TS%	VS%	VS/TS %	C/N ratio	рН	Carbohydrate %	Lipid %	Protein %	Methane Yield (ml/g VS)	Reference
KW	24.9	23.1	92.8	18.24	-	49	17.3	23	501	Jiang et al. (2018)
FVW	13.8	12.88	93.4		4.5	7.74	3.28	2.87	516	Edwiges et al. (2018)
FW	20	19.26	96.3	15.5	-	47.6	24.1	28.3	548.1	Li et al. (2018)
FW	10.86	10.22	94	15.18	4.16	5.71	2.29	1.31	460	Xiao et al. (2019)
FW	25.94	24.59	94.7	17.5	-	48	15.1	10.6	346.2	Shi et al. (2018)
FW	10.69	10.06	94	-	4.18	5.69	2.29	1.30	477-459	Xiao et al. (2018)
FW	24.3	22.5	92.6	23.11	5.02	-	3.38	-	386.7- 551.4	Liu et al. (2017)
FW	19.1	18.53	97	17.7		10.8	4	3.37	536.19	Li et al. (2016a)
AFW	41	34.44	84	-		52	12	25	-	Naroznova et al. (2016)
VFW	23	22.32	93	-		53	5	14	425	
KW	19.1	17.80	93.2	14.41	4.4	11.8	2.5	3.5	372.1	Li et al. (2016a)
FW	23.2	21.7	93.5		4.5	13.7	2.9	6.5	425.2	Zhang et al. (2015)
FW	20.05	19.21	95.81	28.4		33.22	14.03	25.25	381	Yong et al. (2015)
FW	29.4	28.01	95.3	14.2	4.1		18.1	19	529	Browne and Murphy (2013)
FW	18.1	17.1	94	13.2	6.5	11.2	3.3	2.3	479.5	Zhang et al. (2011)

Table 2.3: Methane potential of FW reported in the literature.

FW=food waste; VFW= vegetable & fruit waste; KW= kitchen waste; AFW= animal food

2.2 ANAEROBIC TREATMENT

The overall AD microbiology of food waste is similar to that observed during the anaerobic treatment of either industrial wastewater or sewage sludge. The difference lies in the proportion of trophic groups such as hydrolytic, fermentative and acidogenic bacteria, and acetoclastic or hydrogenophilic methanogens. Anaerobic treatment of wastewater became popular in the early 1980's (Mulligan, 2002). In this sense, AD is a metabolic process where organic matter is degraded in the absence of oxygen. The reaction shown in equation 3 is typical of anaerobic digestion:

Organic matter + H₂O \rightarrow CH₄ + CO₂ + biomass.....(3)

2.2.1 Biochemistry and Microbiology

All anaerobic biological treatment processes involve a consortium of bacteria (from 105 to 107 bacteria per ml) and are based on a series of parallel biochemical reactions and compounds serving as electron acceptors, other than oxygen, and namely nitrates, sulfates, and/or carbon dioxide. (Mulligan, 2002). A general formula representing the decomposition of organic acids to methane and carbon dioxide is shown below in equation (4) (Eckenfelder and O'Connor, 1961):

$$C_nH_aO_b + (n - a/4 - b/2)H_2O = (n/2 - 9/8 + b/4)CO_2 + (n/2 - 2/8 - b/4)CH_4.....(4)$$

Anaerobic treatment can be considered as complex biochemical process involving different classes of bacteria and several intermediate steps (Solera et al., 2002). Micro-organisms involved in the anaerobic treatment can be classified as follows:

a) Primary fermenter bacteria hydrolyzing polymers and fermenting products of hydrolysis, producing volatile fatty acid (VFA), carbon dioxide (CO₂), and hydrogen (H₂); (Lackey and Hendrickson, 1957)

b) Acetate, CO₂, and H₂-producing micro-organisms known as obligate hydrogen producing acidogens which use VFA, propionate, butyrate, and some aromatic compounds as substrate; (Tchobanoglous and Burton, 1991)

c) CO₂ and H₂ consuming methanogens known as hydrogenophilic methanogens and acetate consuming methanogens known as acetoclastic methanogens (Eckenfelder and O'Connor, 1961). In the AD process, the proportion of each bacterial group depends on the amount of specific substrate. Bacteria or microbial groups act as biocatalyst for each metabolic reaction. The reaction velocity is limited by the substrate or nutrient concentration rather than the amount of biocatalyst and not all substrates are susceptible to anaerobic degradation.

2.2.2 Steps Associated with Anaerobic Treatment

The overall anaerobic process involves coordinated and combined metabolic activity of various micro-organism. Such a coordinated action represents a typical multi organism system in which reactions occur sequentially. Zhang et al. (2014) proposed a simultaneous, major four-step mechanism for the conversion of particulate organic matter into methane: Hydrolysis, acidogenesis, acetogenesis, and methanogenesis. These stages can be seen in a simplified version (Fig. 2.5):



Figure 2.5 Steps of anaerobic treatment process
In actuality, the process is much more complicated and intermediate products are used for future steps and future products are used for intermediate steps in a feedback loop of molecules and microorganisms as seen in Figure 2.5. The individual stages of the anaerobic digestion process can be explained as follows.

1. Hydrolysis

The hydrolysis stage of the AD process often includes the degradation process and is referred to as the first stage. The complex organic compounds produced by the degradation of input material carbohydrates, proteins, and fats - are broken down into simpler organic constituents – sugars, amino acids, and long-chain fatty acids, respectively. Hydrolytic bacteria secrete enzymes which break down the long-chain compounds into soluble compounds and, in the process, generate hydrogen and CO₂. For complex wastes like food wastes that are very highly biodegradable, it is advisable to separate the hydrolysis phase from the rest of the process as it is often the most volatile, and the acids produced can dramatically affect the pH and therefore can limit the rate of the entire AD process (Kothari et al., 2014; Leung & Wang, 2016; Zhang et al., 2015; Zhang et al., 2014). Polymers are transformed into soluble monomers through enzymatic hydrolysis, as shown in equation (5):

 $n(C_6H_{10}O_5) + nH_2O \rightarrow n(C_6H_{12}O_6)$ (5)

2. Acidogenesis

In this step, the hydrolysis products are metabolized by acidogenic bacteria and converted mostly into volatile fatty acids (acetate, propionate, butyrate, valerate) and alcohols (ethanol, methanol). Additionally, some CO₂, ammonia, and hydrogen are produced which, along with acetate, can be directly consumed by the methanogenic bacteria if the system is already at steady state (Zhang et al., 2015). Reactions (6), (7) and (8) show the conversion process of glucose to acetate, ethanol, and propionate, respectively.

3. Acetogenesis

The remaining VFAs and alcohols with chains longer than acetate are further broken down by acetogenic bacteria into acetic acid, CO₂, and hydrogen so that the methanogenic bacteria can metabolize them (Kothari et al., 2014). However, most of the acetate is created by hydrogen-producing acetogenic bacteria (Angelidaki et al., 2007). An acetogenesis reaction is shown in equation (9):

 $C_6H_{12}O_6 \rightarrow 2C_2H_5OH + 2CO....(9)$

4. Methanogenesis

In the ultimate step of the anaerobic digestion process, methane and carbon dioxide are produced from the three remaining products of the acetogenic phase. Moreover, a minor quantity of hydrogen sulphide (H₂S), ammonia (NH₃), and other gases are also present when biogas is produced (Monnet, 2003). Most methanogenic bacteria require an optimum pH range between 6.5 and 7.5 (Leung and Wang, 2016). According to the type of substrate utilized methanogenesis is divided into two groups:

1. Hydrogenotrophic methanogenesis. Hydrogen and carbon dioxide are converted into methane according to the following reaction shown in equation (10):

 $CO_2 + 4H_2 \rightarrow CH_4 + 2H_2O....(10)$

2. Acetotrophic or acetoclastic methanogenesis. Methane is formed from the conversion of acetate through the following reaction presented in equation (11):

 $CH_3COOH \rightarrow CH_4 + CO_2 \tag{11}$

Approximately 70% of the methane is produced from acetate while the remaining 30% is produced from the reduction of CO_2 by hydrogen and other electron donors (Smith et. al., 1966; Bitton, 2005). Methanogenesis is the slowest of the phases of AD and is the most sensitive to operating conditions such as input composition, organic loading rate, pH, and temperature. Any remaining substrate which cannot be digested by the methanogenic bacteria that make up the digestate. Figure 2.6 provides the metabolic steps in the conversion of organic matter by anaerobic bacteria.



Figure 2.6 Anaerobic decomposition of organic matter resulting in methanogenesis (Mulligan, 2002)

2.2.3 Operating Parameters of AD Technology

There are several important parameters for proper design and operation of an anaerobic digestion system. A description of these parameters can be summarized by Mata-Alvarez et al. (2003).

2.2.3.1 Temperature

Methane production is highly influenced by temperature, which affects bacterial performance within an anaerobic digester. Both methanogenic and volatile acid-forming microorganisms are affected by temperature. Changes in temperature extensively influence the performance of methanogenic microorganisms compared to the operating temperature (Gerardi, 2003). The AD process can take place at various temperatures, which are normally classified into three types, i.e., psychrophilic, mesophilic, and thermophilic temperatures.

Thermophilic (50°C–60°C): Thermophilic digestion systems operate at the highest temperature range. The desirable aspect of this system is the fact that the micro-organisms rapidly break down the organic material and produce largest volumes of biogas. This results in smaller digestion tanks and shorter retention times – often as short as 5-10 days. The drawbacks are that more insulation is necessary to maintain the temperature range, and more energy is needed for heating the system. Additionally, because the process occurs so fast, it is the most unstable and most sensitive to minor changes in the input material. It may be more practical in areas that are warm year-round and have a consistent waste input.

Mesophilic (30°C–35°C): Mesophilic is the most common range of digestion due to the robust and stable nature of the bacteria involved. A longer retention time (at least 15–20 days) is needed to break down the organic matter and produce biogas then in a thermophilic system. This range is most common for farming and agriculture-food systems. Regarding food waste, several studies

have shown that using mesophilic digestion can yield similar amounts of biogas under similar retention times as thermophilic systems while being more stable (House, 2006).

Psychrophilic (1°C–20°C): Psychrophilic digestion occurs at around ambient/room temperature and is a more recent development as an anaerobic producer of methane. The drawback to this technology is that it takes much longer for the methane to be produced unless there is a long term "storage pit" or lagoon set up that doesn't require much heating and can save a large amount of energy.

2.2.3.2 Total Solids (TS)

Total solids are the residue or dry material which is left over after drying the substrate from 12 to 48 hours at 105°C. It is a raw estimation of the amount of organic and inorganic material in the substrate.

2.2.3.3 Total Volatile Solids (TVS)

Total volatile solids are an approximation of the "organic" fraction of the total solids which is determined by heating TS to 550°C for 24 hours. Leftover material is an inert or mineral fraction (inorganic).

2.2.3.4 Chemical Oxygen Demand (COD)

COD is a measure of the oxygen equivalent of organic material in a substrate. It is determined by adding a strong chemical oxidizing agent to the substrate in an acidic medium. It gives an accurate estimate of the amount of organic (degradable) material in a sample.

2.2.3.5 Retention Time

Retention time is one of the major parameters, which needs to be repeatedly observed in the anaerobic digesters. Retention time is the time needed for the complete degradation of organic matter or the average time organic matter remains in a digester (Deepanraj et al., 2014; Mao et al.,

2015). There are two important types of retention times involved in the AD system: solid retention time (SRT) and hydraulic retention time (HRT). SRT is the average time that the bacteria spend in the digester, whereas HRT is the average time that the liquid sludge spends in the digester (Deepanraj et al., 2014). According to Mao et al. (2015), bacterial development rate related to retention time depends on process temperature, substrate composition, and organic loading rate; the shorter the HRT, the higher the value of organic loading rate. A retention time of 10-40 days is necessary to treat organic waste at mesophilic temperature, while a lower retention time could be used at thermophilic temperature (Kothari et al., 2014). Yadvika et al. (2004) described that high capital cost and large reactor volume are the main requirements for a longer HRT. However, shorter HRT offers insufficient time for the optimal degradation of the substrate. Yadvika et al. (2004) noted that HRT varies with climate change. For example, for tropical countries and in cold weather, HRT fluctuates from 30 to 50 days and 100 days, respectively. SRT maintains the bacterial population in the reactor, which could result in waste stabilization (Dobre et al., 2014). Chen et al. (2018) observed that the CH₄ yield reduced with higher SRT and the highest CH₄ yield was achieved at an SRT of 6 days, compared to 7.5 days and 10 days. Fernández-Rodríguez et al. (2014) reported that the maximum CH_4 yield was achieved at an SRT of 5–8 days compared to SRTs of 4 days and 3 days. They also indicated that SRTs lower than 4 days were inappropriate for a single-stage dry AD of organic fraction of municipal solid waste. HRT can be determined by equation (12).

Where V: Reactor Volume [m³] Q: Flow Rate [m³/day]

2.2.3.6 Organic Loading Rate (OLR)

Organic loading rate (OLR) is a significant operational parameter that affects the CH₄ yield. The

amount of organic material added to the reactor in each amount of time, usually a per day flow rate per unit volume of digester capacity (Kothari et al., 2014). If the reactor is overfed beyond the suitable OLR, inhibitory substrates such as fatty acids could be accumulated and CH₄ production could be reduced. This is because micro bacteria cannot survive in an acidic condition in the AD system. System failure can also occur due to overfeeding. This affects CH₄ production rate, which is highly dependent on OLR (Kothari et al., 2014). Hence, it is important to control the OLR of the digester. Leung and Wang (2016) demonstrated that lower OLR and longer HRT might prompt organic overload, and thus, CH₄ production could be reduced. This is likely due to the insufficient buffering capability in the digester. On the other hand, higher OLR causes shorter HRT, which may result in microorganism washout, and this could lead to lower biogas production (Leung and Wang, 2016). Liu et al. (2017) pointed out that the optimal OLR on AD of FW under thermophilic and mesophilic conditions was 2.5 and 1.5 g-VS/L/day, respectively. OLR can be evaluated by equation (13).

Where: OLR: Organic Loading Rate [kg substrate / m³ / day] Q: Flow rate of input [m³/day] S: Concentration of VS in the input [kg/m³] V: Reactor Volume [m³].

2.2.3.7 Specific Gas Production (SGP)

This aspect of the AD process relates to the amount of biogas produced in cubic meters to the amount of volatile solids being digested on a per unit basis (often m^3/kg or ton). When compared with other substrates, it can be used as a guide to biodegradability (higher SGP = higher degradability). SGP can be determined by equation (14).

$$SGP = \frac{Qbiogas}{Q*S}....(14)$$

Where: SGP: Specific gas production [m³biogas / kg substrate] Q_{biogas}: biogas flow rate [m³/day]

Q: input flow rate $[m^3/day]$ S: VS concentration of input $[kg / m^3]$.

2.2.3.8 pH and Volatile Fatty Acids (VFA)

pH is the most significant parameter that affects the performance and stability of an anaerobic digester. Microorganisms are sensitive to pH. This is because every group of bacteria needs a different pH range for their growth (Appels et al., 2008). The ideal pH range for hydrolysis, acetogenesis, and methanogenesis is almost 6.0, 6.0–7.0, and 6.5–7.5, respectively (Leung and Wang, 2016). Gerardi (2003) reported that the pH required for acid-forming bacteria and methane-forming bacteria is>5.0 and 6.2, respectively, for acceptable enzymatic activity. Methanogenic bacteria display better performance in a pH range of 6.8–7.2 (Yadvika et al., 2004). CH4 production was found to be 75% more efficient with a pH of>5.0 (Yadvika et al., 2004). Krishna and Kalamdhad (2014) indicated that other main factors contribute to the fluctuation of pH such as alkalinity, volatile fatty acid (VFA), the quantity of CO₂ production, and the concentration of bicarbonate (HCO₃) during the AD process. They reported that the relationship between VFA and HCO₃ concentrations should be controlled, as it helps in adjusting the optimum pH during the AD process. Volatile fatty acids such as acetic acid, propionic acid, butyric acid, and valeric are the primary intermediate products produced from the AD of FW (Zhang et al., 2014).

2.2.3.9 Carbon and Nitrogen Ratio

The C/N ratio represents the relationship between the quantity of carbon and nitrogen present in FW. An optimum C/N ratio is required for an effective AD process (Kothari et al., 2014). Zhang et al. (2014) reported that the C/N ratio greatly influences the stability of the AD process. This is because the optimal C/N ratio not only helps to maintain a suitable environment, but it also helps to control proper nutrient balance through the development of microorganisms. The microbial population could increase gradually if the quantity of nitrogen is low in the FW, and thus, more

time will be required to decompose the existing FW, resulting in lower CH₄ yield. In contrast, ammonia inhibition could occur, preventing microbial growth, especially if the concentration of nitrogen is more than the microbial necessity (Kothari et al., 2014; Leung and Wang, 2016). It is found that microorganisms use carbon 25–30 (Yadvika et al., 2004), 25–35 (Krishna and Kalamdhad, 2014) or 30–35 (Leung and Wang, 2016) times quicker than nitrogen during the AD process. Therefore, the optimum ratio of C/N in the substrate should be almost 20–30:1 (Yadvika et al., 2004) 25–30:1(Krishna and Kalamdhad, 2014) or 30–35:1 (Leung and Wang, 2016).

2.3 COMPARISON BETWEEN ANAEROBIC TREATMENT AND COMPOSTING

Food waste was typically disposed of in landfills alongside other MSW. With the capacity of landfills gradually filling up and fewer landfills being put into operation, it is critical to look for alternative disposal methods. One possible method is composting, which decomposes organic materials in an aerobic environment and produces humus that can be used as fertiliser or soil conditioner (Cheng et al., 2007). Organic constituents are converted into carbon dioxide, heat, and a stable fertiliser by microorganisms, mostly bacteria. Another alternative approach to diverting this food waste is anaerobic digestion. Anaerobic digestion is a naturally occurring digestive process in which microbes convert organic materials into biogas and neutral digestate sludge in the absence of oxygen (Paola et al., 2007). The methane-rich biogas produced by the process, which typically contains between 57% and 70% methane, can be burned as fuel; it is regarded as a renewable waste-to-energy technology and can offset the need for fossil fuels (Paola et al., 2007). Biogas can be upgraded (purified to almost pure methane) to become "biomethane". Not least is the fact that it is one of the few ways to decarbonize the transportation industry when it is compressed to create bioLNG (Defra UK, 2007). The high moisture content and biodegradability of food waste make it feasible for composting and anaerobic digestion. Therefore, both composting

and anaerobic digestion can divert waste from landfills. The key advantage of using anaerobic digestion in an urban environment to treat organic waste as opposed to composting it is that anaerobic digestion produces biogas with a high percentage of methane, which can be used as fuel, whereas composting produces mostly carbon dioxide, "the primary greenhouse gas emitted through human activities," according to the Environment Protection Act (EPA) (2014). Composting releases the carbon dioxide originally sequestered by the organic material from the atmosphere and as such is considered a "carbon-neutral" process. No energy is available from this process. Organic waste treated anaerobically has lower odour levels than unprocessed waste. An example of this is the comparison between manure spreading and the same material spreading after anaerobic digestion, when it is known as "digestate." It is much less likely to cause environmental pollution than spreading untreated organic waste on land. Anaerobic digestion can reduce the volume of the input material by 50%-80% and assist in reducing the carbon footprint (WRAP, 2010). However, carbon emissions are not reduced in any way by composting. Moreover, AD is better suited to the digestion of cooked and oily food waste than composting. As a matter of fact, adding discarded cooking oil and cooked meats to the AD process causes an increase in biogas production. Most of the methane is produced within 30 days of adding the organic material to the digestion process, whereas in composting, a full year is often required for neutralization. The equipment required to aerate and turn the compost piles consumes a significant amount of energy during the composting process. Anaerobic digestion, in contrast, is more beneficial for "greenness" because it generates its own power to achieve this.



Figure 2.7 Schematic diagram of (a) composting (b) anaerobic digestion (Defra UK, 2007)

2.4 ANAEROBIC TREATMENT OF FOOD WASTE

Landfilling, the most used disposal method for food waste, contributes to releasing high organic load leachates, terrain settlement and greenhouse gases. In the landfill, the degree and rate of organic matter degradation are often assumed to be higher than they are, when after two decades in the landfill, 33% of non-plastic, organic waste is recognizable. Indeed, reducing and diverting this waste in landfills has become a priority, especially considering the negative impact of food waste on global climate change, and associated environmental challenges (Morone et al., 2019). Slorach et al. (2019) reported that global food loss and waste generate 6.7% of total anthropogenic GHG emissions annually. Papargyropoulou et al. (2014) demonstrated another environmental effect associated with FW, which is the disturbance of the biogenic phases of phosphorus and nitrogen, applied as fertilizers. Considering the negative environmental impacts of landfilling, incineration, or composting of food waste (Lin et al., 2013; Posmanik et al., 2017) among biological treatments, anaerobic treatment has been proposed as a relatively cost-effective technology for renewable energy production and nutrients recovery (Xu et al., 2015). In the context of Life Cycle Analysis (LCA), AD offers several exciting features. Anaerobic digestion of food waste reduces carbon emissions through the production of biogas and biodiesel that could replace fossil fuels and is considered a renewable, carbon-neutral source (Sarkar et al., 2021). Moreover,

aerobic treatment inevitably leads to extensive emission of undesirable volatile compounds such as ketones, aldehydes, ammonia and even methane. On the other hand, all gases are contained during AD and can be eliminated by flaring the biogas. One of the main factors promoting the AD of food waste is the practice of source-separated collection; these processes are efficient, with as much as 85% of the degradable material being turned into biogas and a similar percentage conversion of the calorific value of the food waste into a usable energy product since the waste becomes "cleaner," and this facilitates the process of energy and materials recovery. Lissens et al. (2001) observed the trend in the AD of solid waste sorted mechanically in central plants or organic wastes separated at the source (biowaste; mainly paper, vegetables, fruits, food left-over and garden waste). Batch reactors for wet and dry systems were the most common types of reliable anaerobic digesters. The process configuration varied according to needs and included, at times, one- or two-phase digestion systems. Almost all treatment plants include both pre- and posttreatments, depending on several factors: characteristics of the substrate, the type of anaerobic technology, and the final use of the biosolid produced. Pre-treatment steps may include magnetic separation, commuting in a rotating drum or shredder, screening, pulping, gravity separation (dry separation), or pasteurization. In the post-treatment steps, the typical sequence involves mechanical dewatering, aerobic maturation, and water and/or gas treatment (Figure 2.8).



Figure 2.8 Overview of pre- and post-treatment technologies in food waste Lissens et al. (2001).

2.4.1 Operating Conditions of Anaerobic Treatment Process

AD requires a series of biochemical transformations which is 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, commonly used technology relate to one stage, two stage and batch reactor. In one-stage and batch systems, all these reactions take place simultaneously in a single reactor, while in two- or multiple-stage systems, the reactions take place sequentially in at least two reactors. A useful tool in evaluating the biological performance of AD is the maximum sustainable reaction rate expressed as both a rate of substrate utilization or maximum organic loading rate (OLR) expressed as kgVS/m³ per day, under standard pressure and temperature. Hydraulic retention time (HRT) is another parameter affecting reactor performance.

2.4.1.1 Wet and Dry Digestion

The term "solid waste" generally means organic biodegradable waste with over 15% TS. The organic solid waste is diluted with water in wet, thoroughly mixed one-stage systems. The AD process can be categorized as wet or dry digestion based on the total solid concentration in FW. The anaerobic digestion of food waste is termed a dry process if the total solid concentration of the food waste stays between 20 and 40%. In contrast, the anaerobic digestion of food waste is considered a wet process when the total solid concentration of the food waste is <15% (Kothari et al., 2014; Deepanraj et al., 2014). Kothari et al. (2014) noted that most AD plants constructed during the 1980s depended on the wet system, whereas new plants constructed in the last decade are primarily based on the dry process. They also noted that the HRT, OLR, and volatile solid removal rate of the dry process were 14-60 days, 12-15 kg VS/m³/day, and 40%-70%, respectively, whereas the HRT, OLR, and volatile solid destruction rate of the wet process were 25–60 days, <5 kg VS/m³/day, and 40%–75%, respectively. Yin et al. (2016) examined dry (total solid=20%) and wet (total solid=5%) processes of AD during food waste treatment and found that the CH₄ production (0.48 L/g VS) and volatile solid reduction (85.6%) were higher in the dry process compared to the wet process. They also found that the dry process allowed a higher VFA concentration and OLR than the wet process, resulting in a decreased possibility of inhibition of the AD technique. Complete mixing of the waste is not possible in a dry process; thus, the ideal contact of microorganisms and substrate cannot be guaranteed. Conversely, the wet process offered several essential benefits, including greater flexibility over the type of feedstock accepted, dilution of inhibitory substances by process water and the necessity of less sophisticated mechanical equipment.

2.4.1.2 Batch and Continuous System

In batch systems, digesters are filled once with fresh waste, with or without addition of seed material, and allowed to react sequentially either in the dry mode at 30 to 40 % TS, or the wet mode with less than 15% TS. Batch systems are technically simple, the investment costs and maintenance requirements are significantly lower than those continuously fed (Kothari et al., 2014). Biogas production can be kept almost constant and/or continuous due to the constant input of FW. Park et al. (2018) studied the effect of feeding mode for the AD of FW and reported that the continuous feeding of diluted FW yielded constant performance compared to the batch feeding of undiluted FW.

2.4.1.3 Single-stage and Two/Multi-stage Process

About 90% of the current European full-scale plants for AD of OFMSW and biowaste rely on onestage systems, approximately evenly split between wet and dry operating conditions (De Baere, 2000). The single-stage process occurs when four metabolic phases—hydrolysis, acidogenesis, acetogenesis, and methanogenesis—take place in one reactor. Low OLR, long retention time, a pH range between 6 and 7, low CH₄ production, less investment, and less maintenance cost are the significant features of the single-stage process (Xu et al., 2018). The main limitation in the singlestage reactor is the presence of acidogenic microorganisms (i.e., decreased pH) resulting from the quick acidification of FW during the acid formation phase, which disturbs the methanogenic bacterial groups. This is likely due to the lack of an optimum environmental condition inside the single-stage reactor. It is worth noting that approximately 95% of Europe's full-scale plants usually operate in a single-stage process occurs in separate anaerobic reactors. The first reactor is used for hydrolysis, acidogenesis, and acetogenesis, whereas the second is mainly used for methanogenesis. Studies have shown that the acidification stage is evident in operating conditions of low HRT (2–3 days) and acidic pH (5.5–6.5), in which the acidification stage must be maintained, whereas high HRT (20–30 days) and optimum pH (6–8) are required for the development of gradually growing methanogenic bacteria (Xu et al., 2018). Hagos et al. (2017) reported that the two-stage process has several benefits, such as increased CH₄ production, high OLR, improved process stability, higher possibility of managing pathogens, and enhanced rate of volatile solid removal efficiency. They also noted that complex maintenance, high capital, and operational cost were the major drawbacks of the two stages process. Xiao et al. (2018) compared the performance and energy recovery of single-stage and two-stage thermophilic anaerobic digestion (TAD) of FW. Single-stage TAD achieved slightly higher CH₄ yield, and volatile solid removal rate compared to the two-stage TAD. They also noted that single-stage TAD recovered more energy than the two-stage TAD.

2.4.2 Energy Recovery through Biogas and Methane Production

The shortage of fossil fuel and the growing concern of global warming have sparked a great development of renewable biofuel applications worldwide. Biogas contains around 50%–80% by volume of methane (CH₄), 30%–40% of carbon dioxide (CO₂), traces of hydrogen sulfide (H₂S), hydrogen (H₂), the percentages of these compounds vary according to the digested substrate and the applied technology (Møller et al., 2009). Anaerobic digestion, even under mesophilic and thermophilic conditions, produces sufficient methane to justify its conversion into electricity and heat. This biogas is a renewable energy because of its potential used in the treatment facilities, thus reducing, and even avoiding consumption from the municipal supply. AD is more environmentally preferable based on its impacts on climate change. When food waste is used for AD, the biogas yield is affected by any difference in carbohydrate, protein, and lipid contents The biogas produced

from AD can be utilized as distinct types of energy sources, such as heat, and electricity produced from biogas combustion and biogas fuel for vehicle use through biogas upgrading (Lin et al., 2013). Regarding the electricity production, a combined heat and power (CHP) unit is often used in AD facilities to generate both heat and electricity. The electricity generated can be used internally in the AD facility and the excess electricity is often delivered to the public grid. The heat can be used to maintain the temperature of the anaerobic reactor and provide heat for nearby industries, given that the AD facility is connected to the methane level of biogas fuel is generally enriched to over 95 % for vehicle use. Furthermore, Lin et al. (2013) also suggested biogas can be upgraded for household use (i.e., city gas) through injection into the gas grid district heating system (Møller et al., 2009). In order to determine the most environmentally friendly option for biogas utilization, Woon et al. (2016) evaluated the environmental impacts from different biogas utilization methods, namely electricity and heat, city gas, and biogas fuel as a petrol, diesel, and liquefied petroleum gas substitute for vehicle use. It was found that upgrading biogas for vehicle fuel use achieves the greatest avoided emissions, especially when substituting for petrol fuel.

2.5 PSYCHROPHILIC ANAEROBIC TREATMENT OF FOOD WASTE

A remarkable evolution has occurred in the attitude toward the treatment of food waste. Skepticism concerning its viability has given way to a universal agreement that different digester types with differing operation temperatures can operate reliably at full scale. Anaerobic Treatment (AD) under psychrophilic temperature has only recently garnered deserved attention. In frigid countries, such as in Canada, Europe, Russia, the northern parts of the USA, and South Australia, the ambient temperature is expected to be below 20 °C for a significant part of the year. Hence, opting for AD under the mesophilic temperature range would require a significant amount of energy to heat the system, which can reduce the net energy yield as well as add to the cost of operation (Lettinga et

al., 2001). Given these environmental conditions, psychrophilic anaerobic treatment (>20 °C) is preferable to mesophilic (30-35 °C) and thermophilic (50-60 °C) treatment (Bikash et al., 2021). The significant amount of energy needed to maintain the reactor at high temperatures is the fundamental drawback of mesophilic and thermophilic treatment processes (Pramanik et al., 2019). This temperature rise is accompanied by a considerable increase in the operating and maintenance costs. Due to subfreezing winter temperatures in Canada, digesters operated during the winter used most of the gas they produced and sometimes required supplementary heating to maintain the digester temperature (Van Die, 1987). Psychrophilic AD seems to be an efficient alternative for treating food waste with minimal energetic requirements in this scenario. Most of the research in AD is carried out either in the mesophilic (30–35 °C) or thermophilic (50–60 °C) temperature ranges even though substantial biomethane production occurs under cold conditions (>20 °C) and is mediated by psychrophilic archaea. Adsorption of AD at ambient temperature can alleviate the use of external energy input and offer economic benefits (Dhaked et al., 2010). Therefore, it is preferable to seed the biodigester with active inoculum that is preferably low temperature adapted and can continue system operation at the psychrophilic temperature range. This can be followed by regulating operational and design parameters, which are also effective methods for improving psychrophilic AD treatment process. There is sufficient experimental evidence to demonstrate that the anaerobic reactor systems used for low temperature treatment result in high methane generation as well as high COD removal efficiency, which contrasts with the common belief that maximum methane production occurs at mesophilic temperature range. According to several recent studies on anaerobic treatment, methane generation is higher in the psychrophilic reactor (0.800 m³/VS) under certain operating conditions than in the mesophilic system (0.751 m³/VS) (Jiri Rusn et al., 2020).

Furthermore, compared to the mesophilic operation, the psychrophilic process utilized higher proportions of volatile organics contained in the substrate for methane generation. A peculiar increase in biomethanation is observed when microbes acclimatized under psychrophilic conditions are exposed to increased temperatures. This is due to a particular class of microbes known as the psychrotropic, which can survive in cold climates and resist temperature changes from the psychrophilic to mesophilic range (Gianese et al., 2002). However, most of these studies indicate more diverse methanogenic archaea at psychrophilic temperatures than mesophilic ones (Dhaked et al., 2010). It is expected that the use of low temperature acclimated microbial biomass could enhance both organic matter removal as well as reduces the reactor start-up time (Akila and Chandra, 2007). The input energy used for the operation of an AD plant should be lower than the output energy generated. If the process is energy-positive, AD plants will be significantly transformed into amenities that generate multiple benefits in the form of recovered water (agriculture, industry, and drinking water according to the treatment efficiency), nutrients, energy, and other value-added chemicals (Massara et al., 2017). However, the anaerobic conversion of food waste at temperatures below 30 °C has attracted limited attention, and extraordinarily little to negligible information can be found in the literature. Modification and certain alterations to the existing design of AD plant treating the food waste can result in better methane production without any considerable energy input. This can provide a promising solution to the energy crisis and fulfil the larger picture of the circular economy concept with zero-waste generation.

2.6 NEED FOR THE PRESENT RESEARCH

According to a report by Uzea et al. (2013), 40% of food produced in Canada ended up disposed as a waste. This food waste is estimated to be worth \$27 billion, or 2% of Canada's GDP (Gooch et al., 2010). The majority of this food waste finds its way to composting or landfills, resulting in the emission of greenhouse gas. In 2009, landfills in Canada contributed 3% of all greenhouse gas (GHG) emissions and 22% of the nation's methane emissions (Statistics Canada, 2012). Treatment of food waste is an economically challenging task, predominantly in cold regions where temperatures can fall below 20°C. Most anaerobic digestion plant operations are performed at mesophilic (30-35°C), but some new processes have been reported at thermophilic (50-60°C) temperatures for increased methane production and improved sludge digestion. The ambient temperature in cold-weather countries such as Canada and the United States is expected to be below 20°C for the majority of the year. Therefore, both mesophilic and thermophilic anaerobic treatments are limited by the energy required to heat the bioreactors to maintain the required temperatures. In Canada, the majority of the energy produced by AD plants is used for plant operation during the winter, and occasionally even additional heating is needed, which renders the anaerobic plant unprofitable and energy negative. Therefore, an effective method to control organic wastes is essential for the sustainable management of the nation's economy and the environment. However, there is limited information on the successful operation of digesters treating mixed municipal biowaste at temperatures below 20°C. Since, anaerobic technology is very sensitive to temperature, lower operational temperature can be detrimental to microorganism growth and methane production. AD plants at mesophilic and thermophilic temperature ranges are by comparison better-understood technologies, and hence more research efforts are required to better understand the psychrophilic anaerobic treatment of food waste. Rajagopal et al. (2013a)

and Massé et al. (2014) have shown the technical feasibility of psychrophilic anaerobic digestion to treat animal wastes containing excess ammonia without inhibition and had no effect on methane yield. This study aims to use anaerobic technology to treat a scarce resource like food waste at a lower ambient temperature, with the goal of making the AD process more economically feasible and environmentally sustainable. As a result, this research proposes a novel biogas recirculation technique in psychrophilic AD technology for the treatment of food waste that can overcome thermodynamic constraints while maintaining system efficiency comparable to mesophilic and thermophilic treatment.

CHAPTER 3 MATERIALS AND METHODS

3.1 GENERAL REMARKS

This chapter describes in detail the materials and methods used in this study. It includes analytical methods for the determination of chemical oxygen demand (COD), alkalinity, C/N ratio, total nitrogen, volatile fatty acids (VFAs), and carbon dioxide (CO₂) and methane gas (CH₄) content. In addition, methods used to characterize food waste, such as total solids (TS), total volatile solids (VS), etc., are presented. Also, the experimental setup and protocols followed for batch tests will be outlined. The methodology described in this chapter is directed towards developing a psychrophilic anaerobic batch reactor with biogas recirculation into the anaerobic digester to increase methane content in the produced biogas without any additional energy.

3.2 MATERIALS

The utilized material can be divided into food waste, inoculum, and pH adjustment solutions.

3.2.1 Food Waste

The food waste was developed based on weekly food scrap collections from the residential area of Downtown Montreal. Food waste mainly consisted of bread, peas, onions, rice, potatoes, salt, tomato sauce, spinach, beans, coffee grounds, tea bags, eggs, and plastic food wraps. Major portion of food waste was fruit and vegetable waste, then rice, noodles, pasta took up to 23% and 9% of the food waste was non-degradable material. Figure 3.1 shows the approximate compositional analysis of collected food waste sample.



Figure 3.1 Compositional analysis of collected food waste sample

Firstly, the food waste sample was prepared by mixing the whole food scraps by hand, followed by grinding food scraps with 100 mL of water in a food processor, resulting in a paste. Another food waste sample was manually mixed and chopped to reduce particle size, facilitating digestion. Food waste was kept in the fridge at 1 degree Celsius before use. Food wastes and anaerobic inoculum were analysed for total solids (TS), volatile solids (VS), and in duplicate prior to experiments and at the end of the digestion period. Initial and final TS and VS were used to determine solid reduction during digestion. All analyses were performed according to the standard methods (APHA, 2005). Physio-chemical properties of food waste sample used in this study are shown in Table 3.1.

Parameter	Value
pH	5.7-6.1
TS%	27-29
VS%	26.19-28.07
(VS/TS) %	97.42-98.2
Moisture Content (%)	71-73
Alkalinity (g/L HCO ₃)	1.48-1.52
Total COD (TCOD) (mg/L)	179400-181325
Soluble COD (SCOD) (mg/L)	116950-129456
Volatile Fatty Acid, VFA (g/L)	9.1-10.25
Total Nitrogen (mg/L)	1020-1032
Total Carbon (%TS)	52.6-54
C/N ratio	25.41-26.73

 Table 3.1: Physio-chemical properties of food waste sample

3.2.2 Inoculum

The inoculum used in this project was obtained from a benchtop anaerobic reactor treating dairy waste. The collected inoculum was initially kept in the incubator for further acclimation in an anaerobic reactor until biogas from the inoculum ceased altogether. After that the inoculum was kept in 4°C before use. The characteristics of the inoculum are shown in Table 3.2.

Parameter	Value
pH	5.9-6.2
TS%	0.5-1.0
VS%	97-99
Moisture Content (%)	99-99.5
Alkalinity (g/L HCO ₃)	1.04-1.07
Total COD (TCOD) (mg/L)	5000-5500
Volatile Fatty Acid, VFA (g/L)	0.425-0.5
Total Nitrogen (mg/L)	338-342

 Table 3.2: Physio-chemical properties of inoculum

3.2.3 pH Adjustment solution

NaOH (1M) and HCl (1M) solutions were used to adjust the pH of solutions.

3.3 ANALYTICAL METHODS

3.3.1 Chemical Oxygen Demand (COD)

The chemical oxygen demand (20-1500 mg/L) was measured based on the USEPA reactor digestion method (Standard Method 5220 D). In this test, COD test vials (Hach Inc.), a spectrophotometer (Cole Parmer, model DR 2800), and a DRB200 Digital Reactor Block were used. Sample could be diluted with distilled water if COD value exceeded the measuring range.

Test procedure:

- 1. The reactor block was turned on and preheated to 150 °C.
- 2. The cap of the COD reagent was removed, and a 2 ml sample was pipetted into each vial.

The cap was placed, and the vials were inverted gently several times to mix the solution.
 The vials were placed in the preheated reactor block and were heated for two hours.
 The reactor block was turned off after two hours and allowed the vials to cool to 120 °C.
 The COD vials were inverted again and cleaned on the outside.

7. The COD vials were inserted into a rack and then allowed to cool down to room temperature.

8. COD vials were placed in the spectrophotometer and the value was read.

3.3.2 Alkalinity

Alkalinity represents the buffering capacity of solution and assesses the ability of a solution to neutralize acids. Alkalinity was measured by titration based on the method NO. 2320B (Standard Method, 1998). The materials used: Bromocresol green, distilled water, sulfuric acid (0.1 N). Procedure: Bromocresol green solution: 100 mg dry bromocresol green was dissolved in 100 ml distilled water. It changes colour at pH 4.5. Standard sulfuric acid, 0.02N: Diluted 20 ml of 0.1 N standard sulfuric acid into 100 ml by using distilled water. 1 ml of standard sulfuric acid (0.02N) is equivalent to a total alkalinity of 1 ppm calcium carbonate. The last point for the titration test was determined according to equation 15 and the colour change of the solution (blue to pale green).

Where,

A= ml acid used for titration

N= normality of standard acid

3.3.3 Volatile Fatty Acid (VFA) Analysis

The concentration of VFAs was measured by the esterification method, using Volatile Acids TNT plus Reagent purchased from Hach Inc. (Ohio, USA). A spectrophotometer (Cole Parmer, model DR 2800), and a DRB200 Digital Reactor Block were used. (Hach procedure manual, 2009).

Procedure:

1. The reactor block was preheated up to 150 °C.

2. The cap of VFAs reagent vial was removed, and 0.4 ml of solution A sample was pipetted to the vial. Then 0.4 ml of sample was pipetted to the test vial.

3. The cap was placed, and the solution was inverted several times.

4. The vial was put in the preheated reactor block and was heated for 10 minutes.
5. After 10 minutes, the test vial was placed in the rack and was cooled down to the room temperature (15°C-25°C).

6. 0.4 ml of solution B was added to the vial and the cap was replaced. After that the vial was inverted several times.

7. 0.4 ml of solution C was added to the vial. The cap was replaced, and after that the vial was inverted several times.

8. 2 ml of solution D were added to the vial. The cap was replaced, and the vial was inverted several times.

9. The test vial was placed in the spectrophotometer after three minutes and the value represented the volatile fatty acid of the sample.

3.3.4 Total Nitrogen (TN)

Total nitrogen refers to all nitrogen forms. TN was measured by the persulfate digestion method using nitrogen, Total TNT plus Reagent purchased from Hach Inc. A spectrophotometer (Cole Parmer, model DR 2800), and a DRB200 Digital Reactor Block were used.

Procedure:

1. The reactor block was heated up to 100 °C.

2. 0.5 ml of sample, 2.0 ml of solution A and 1 of reagent B tablet were added in quick succession to reaction tube and then closed. The tube immediately was placed in the preheated reactor block and heated for 1 hour.

3. The tube was taken out from the reactor and was cooled down to room temperature.

4. 1 Microcap C was added to the tube.

5. The cap was replaced, and the vial was inverted several times until streaks could not be seen in the tube.

6. 0.5 mL of the solution from the reaction tube was pipetted into a test vial.

7. 0.2 ml of solution D was pipetted into the test vial.

8. Quickly placed the cap and inverted the vial 2-3 times until streaks could not be seen in the vial solution.

10. After 15 minutes, placed the vial in the spectrophotometer and read the value.

3.3.5 Microbial Parameters (TS, VS)

Substrates were further characterized for their composition in total solids (TS) and volatile solids (VS) contents, these properties being the indicators of the potential of any substrate to produce

biogas. Solids analysis is important in the control of biological and physical treatment processes and for assessing compliance with regulatory effluent limitations. The tests were done to measure the biomass concentration in the reactor and also to measure the decrease in the concentration of volatile solids in the effluent in comparison with the influent which is a good method to evaluate the system efficiency. Total Solids (TS) and Volatile Solids (VS) were measured as per the Standard Methods (U.S Environmental Protection Agency, 2000). Each test was carried out at least three times according to the following procedure.

1.Crucibles were used as containers of the samples; empty crucibles were weighed, and their weight was denoted by Wc,

2. When loaded with fresh substrates, their weight was denoted by Ws.

3. The samples were dried in a laboratory drier at 105°C for three days, cooled in a desiccator for 15 minutes prior to weighing.

4. When weighed, the weight was denoted by Wd; the weight of the crucibles and dried substrates. The TS content was calculated using equation (16).

$$TS = \frac{Wd - Wc}{Ws - Wc} \times 100....(16)$$

Where,

TS: total solids content

Wd: weight of the dried sample in g

Wc: weight of empty crucible in g

Ws: weight of fresh sample in g

5. The same dried samples were supplied to a kiln at 550°C for 20 hours to burn to ash.

6. Then, the residue was cooled in the desiccator and weighed denoting the weight by Wash. The results of this process were used to calculate VS content using equation 17.

Where,

VS: volatile solids content

Wd: weight of the dried sample in g

Wc: weight of empty crucible in g

Wash: weight of ash in g

3.3.6 Sample Withdrawal Technique

A 60 ml gas-tight syringe glass and a 10-ml plastic syringe purchased from Fisher Scientific Ltd. (Montreal, Canada) were used for taking gas samples and the liquid, respectively. In batch experiments, liquid sample was withdrawn every day during the treatment process and gas sample was withdrawn once every two days for two weeks in the frequency of 5 days interval till the end of anaerobic treatment. Biogas samples were withdrawn daily from the gas sampling port in the reactor using a 60 mL gas-tight syringe.



Figure 3.2 Biogas Extraction

3.3.7 Volume of Biogas

A Tedlar bag, purchased from Fisher Scientific Ltd., connected to a needle was used to collect the produced gas in the experimental bottles. The volume of collected gas in each bag was subsequently measured by the water displacement method. For this purpose, a flask with one inlet and one outlet in the cap was used. The flask was completely filled with a solution of 0.05 N sulfuric acid. The acidified water prevents the dissolution of biogas in water. As the gas in the bag was introduced in the water, water with the same volume as the biogas was displaced from the flask to a graduated cylinder and its volume was measured.

3.3.8 Purity of the Biogas

The biogas produced during the anaerobic treatment is mainly composed of methane and CO₂ with trace concentrations of other gases. Consequently, the ratio of methane to carbon dioxide demonstrates the composition of produced biogas in each bottle. The composition of the biogas produced was determined by a gas chromatograph (GC, Agilent 7890B with a TCD detector) using CARBOXEN 1010 PLOT (30mm×0.32mm) capillary column from SUPELCO with helium as the carrier gas at 250°C inlet temperature, a TCD detector, column oven temperature of 250°C (ramped at 5°C/min), injection flow of 6.5 ml/min and run time of 5 min. A standard curve (Appendix) was made by preparing gas samples with 20, 40, 60, 80, and 90% (vol./vol.) of methane and recording the associated CH₄/CO₂ ratio. The volumes of methane and carbon dioxide were calculated by multiplying the total volume of biogas, measured by the corresponding percentage of CH₄ or CO₂, indicated by the GC. Calibration curves are illustrated in Appendix.



Figure 3.3 Agilent 7890B gas chromatograph

3.3.9 Methane Yield and System Efficiency

An important parameter for measuring anaerobic digestion system performance is the proportion of methane produced for each unit of COD that has been removed. With this calculation, it is possible to find the amount of biomethane, and by extension caloric energy, that will be produced as from a given COD input into the system. The system efficiency was based on the methane production per amount of COD consumed followed by comparison with the theoretical value (0.35 L methane/gram of COD removed under standard conditions) and COD removal efficiency. The system efficiency and COD removal efficiency can be calculated by equations 18 and 19:

$$\text{COD removal }\% = \frac{\text{COD}_i - \text{COD}_f}{\text{COD}_i} \times 100.$$
 (19)

Where, COD_i = initial COD concentration; COD_f = final COD concentration.

3.3.10 Particle Size Distribution

Particle size distribution of sample food waste was evaluated by Laser Scattering Particle Size Distribution Analyzer LA-950 for the mean particle size less than 2mm. Sieve analysis was performed when the mean particle size was larger than 2mm.

3.3.10.1 Particle Size Distribution by Laser Scattering Particle Size Distribution Analyzer

The LA-950 is a sophisticated tool that uses advanced sizing methods and improvements to analyse both wet and dry samples with sizes ranging from 10 nanometres to 3 millimetres. The principle behind laser scattering is that the angle at which light is scattered by a particle is proportional to the particle's size. The LA-950 adheres to ASTM standards for measuring the statistical percentile values such as D10, D50, and D90 values of materials during testing. Here, D10 represents that 10% of particles in the sample are equal or smaller than this size and D90 means that 90% of total particles are equal or smaller than this size. D50 is the median value of particle size distribution which means 50% of particles are smaller than this size or 50% of particles are larger than this size.

Test procedure:

1. The food waste sample was mixed with water to create a liquid suspension.

2. Liquid suspension of the sample was kept in the reservoir in the equipment.

3. A pump in the analyser was used to recirculate the sample into the measuring unit.

4. The unit used static light scattering (a.k.a. laser diffraction) to calculate particle size using the angle and intensity of scattered light. Larger particles were scattered at small angles, and smaller particles were scattered at wide angles.

5. The group of particles created a pattern of scattered light based on intensity and angle, which can then be transformed into a result indicating the size distribution of the particles.

6. The LA-950 is driven by software that created the particle size distribution curve with the refractive index of sample material.



Figure 3.4 Laser scattering particle size distribution analyser LA-950V2

3.3.10.2 Particle Size Distribution by Sieve Analysis

The sieve analysis test is conducted to calculate the proportion of each particle size present in a sample. The test outcomes can be utilized to construct a curve representing the particle size distribution. The sieve analysis technique is applicable for determining the particle size distribution of samples with diameters larger than 0.075 mm. The sieves utilized in this method have woven wire screens with square-shaped openings. The U.S standard sieve numbers and their related opening sizes can be found in Table 3.3.

Sieve No.	Opening (mm)
3/4"	19
1/2"	12.5
3/8"	9.5
#4	4.75
#8	2.36
#200	0.075

Table 3.3: U.S. Sieve numbers and their related opening sizes

Test Procedure:

1.To perform sieve analysis, 100g of representative food waste sample were collected and its mass $W_0(g)$ was determined.

2. The sieves were thoroughly cleaned before the experiment. If any particles were stuck in the openings, a brush was used to poke them out.

3. The sieves used in the experiment were stacked so those with larger openings (lower numbers) were placed above those with smaller openings (higher numbers). A pan was placed under the last sieve (#200) to collect the portion of sample waste passing through it. The stack should always include the #4 and #200 sieves.

4. The pan and all of the sieves were weighed separately.
5. The sample was poured from above into the stack of sieves and placed the cover. The stack was kept in the sieve shaker, affixed the clamps, and started the shaker.

6. Finally, the mass of each sieve with the retained sample was calculated.



Figure 3.5 Sieves for particle size analysis

3.4 EXPERIMENTAL SETUP

The experiments were performed in batch mode of operation. Pyrex solution bottles (Fisher Scientific Ltd., Montreal) were used as batch reactors. Each bottle had a working volume of 500 mL, and each serum bottle was sealed with a butyl rubber septum and crimped aluminium caps. The cap enabled the bottle to be sealed, prevented the produced biogas from leaking from the bottles, and enabled taking gas and liquid samples using syringes without opening the cap. pH was adjusted in the batch bottle with sodium hydroxide 1M and 1M hydrochloric acid. Batch bottles were kept in an incubator to ensure mesophilic temperature (30-35°C) and atmospheric temperature for psychrophilic (1-20°C) conditions. Each batch test was run in duplicate and carried out for 30 days. The batch setup is shown in Figure 3.6.

3.4.1 Operation

The anaerobic digestion of the food waste in batch reactors was carried out in different trials: (1) First set of batch experiments was performed with six different substrates to inoculum ratios (0.5,0.75, 1.0, 1.5, 2.0, 4.0 weight g VS basis) to determine the optimum ratio for anaerobic treatment and substrate with 10% TS was added into each digester. (2) Next two groups of experiments used six different total solid percentages (5%, 10%, 12%, 15%, 18% and 20%) and two temperature ranges (mesophilic and psychrophilic temperature). Each set of batch trials had the same experimental condition with the substrate-to-inoculum ratio of 1.0, and only the temperature was different. (3) Psychrophilic anaerobic treatment was subdivided into two sets of trials. In the first set, anaerobic treatment was conducted without any biogas recirculation and in the second set psychrophilic anaerobic treatment was experimented with biogas recirculation into the anaerobic digester. The head spaces of the reactors were sparged with ultra-high purity argon gas for 5 minutes to ensure anaerobic conditions. All the reactors were manually mixed once a day for at least 2 minutes every day during the treatment process. Batch reactors were placed in an incubator to provide mesophilic temperature, and the experiments were performed at room temperature (18-22°C) by placing the bottles under the flume hood. In each experiment, blank reactors with only inoculum and tap water were also prepared to correct for the biogas produced from the inoculum. Experiments continued for 30 days until the daily gas production was < 1% of the cumulative gas production. Tap water was used to bring the working volume up to 500 mL working volume. All the reactors were tightly closed with rubber septa and screw caps. The biogas accumulated in the headspace of the serum bottles was sampled regularly, and the CH_4 carbon dioxide (CO_2) concentrations were determined by gas chromatography. Biogas samples were taken every five days intervals till the end of the treatment process. The produced biogas was analysed by

withdrawing the gas via a 60 ml syringe and then introducing the withdrawn sample to the gas analyser. Process parameters, including COD, biogas production, and methane content, were monitored during the experiment. A Tedlar bag with pipe and needle was used to take the gas samples, and a 10 ml sterile plastic syringe was used to take out the liquid sample from the digester.



Figure 3.6 Batch anaerobic reactors

CHAPTER 4 RESULTS AND DISCUSSION

The results of batch tests are reported in this chapter along with a discussion of the findings. The sample analysis was constructed using the conventional techniques described in Chapter 3. The duration of each set of tests was 30 days until no more biogas was produced. The ideal substrate to inoculum ratio was examined in this study for optimal methane production. This chapter compares the wet, semi-dry and dry anaerobic treatments of food waste in depth. Moreover, a detailed comparison between mesophilic and psychrophilic AD treatment was performed. Finally, research into the impact and potential of biogas recirculation on bioenergy production from food waste in a psychrophilic environment was conducted.

4.1 EFFECT OF SUBSTRATE TO INOCULUM RATIO ON METHANE CONTENT

To investigate the effects of substrate-to-inoculum (S/I) ratio on methane production performance, six substrate-to-inoculum (S/I) ratios: 0.5, 0.75, 1.0, 1.5, 2.0 and 4.0 g of weight VS basis were studied. Anaerobic treatment of food waste in batch reactors was carried out in six different trials with 0.5, 0.75, 1.0, 1.5, 2.0 and 4.0 S/I ratios at an average temperature range of 30-35°C and with 10% total solid in the batch experiment. The treatment process was continued for 30 days until biogas production altogether ceased. Sodium hydroxide (NaOH) 1M solution was added to each batch reactor to maintain an initial pH of 7.5. It was necessary to add chemicals (NaOH) for controlling the digester pH when the food waste was digested in a batch digester to prevent digestion failure due to accumulation of VFAs and maintain alkalinity in the system (Chen et al., 2014). Food waste was ground into smaller particle sizes which provided a surface for microbial growth and ensured a homogenous mixture before the treatment.

4.1.1 Variation in CH₄% for 30-day Time Period for Different S/I Ratios

Batch anaerobic treatment of food waste with six different substrates to inoculum ratios was conducted for 30-day time period. The batch reactors were kept under mesophilic conditions of 30 to 35 °C. After 30 days, no significant biogas production was observed. Figure 4.1 shows the variation in methane percentage in biogas during the anaerobic treatment process for different S/I ratios. Determining methane concentration and other biogas constituents are an efficient tool to predict potential stresses on the system. The thermodynamic limitations and accumulation of hydrogen and acetates may cause a reduction or complete inhibition of the activity of the microorganisms involved in the process, especially the archaea methanogenic, consequently causing the methane concentration decrease and reduction in the conversion efficiency of organic matter (Chernicharo, 2007). Studies found stable methane concentration during the anaerobic digestion of food waste with inoculum, which approximately ranged from 65 to 85%, and is compatible with this research's production (Yong et al., 2015). Biogas produced from each batch reactor was analysed by gas chromatography to determine the biogas composition. Methane percentage in biogas was evaluated every 5 days since the initiation of the treatment process. Figure 4.1 showed that the proportion of methane increased with the retention time and then began to decrease with time beyond a particular point in the treatment procedure. After one or two weeks, the microbial populations acclimated to the prevailing conditions, and biogas production commenced, usually with a sharp rise in methane production. A similar trend in methane percentage was observed for all six S/I ratios. From the initiation of the treatment process to up to 10 days of time, methane percentage increased with time. The amount of methane in the biogas gradually decreased after day 10. Methane concentration peaks were 51.5%, 70.5%, 89.2%, 64.5%, 72.7% and 70.6% at S/I ratios of 0.5, 0.75, 1.0, 1.5, 2.0 and 4.0, respectively. A maximum of 89%

methane content has been observed in the biogas composition after 10 days of the treatment process for S/I ratio of 1.0. After 25 days of time, a sharp decrease in methane percentage was noticed and eventually dropped from 10% to 40% after 30 days of treatment. It is because microbial growth and methanogenic activity were higher in the initial stage. The organic content in the reactor started declining with time due to the bioconversion of food waste into methane and carbon dioxide.



Figure 4.1 Variation in CH4 % over a 30-day period for different S/I ratios

4.1.2 Variation in CO₂% with Time for Different S/I Ratios

Carbon dioxide content in biogas composition also fluctuated with time. Figure 4.2 shows the variation in carbon dioxide percentage with time for six different substrate-to-inoculum ratios. Generated biogas was investigated for the compositional analysis to determine the methane and carbon dioxide percentage by gas chromatography at the frequency of 5-day intervals till the end of the treatment process. The obtained results from the experiments demonstrated that the carbon dioxide percentage was relatively higher in produced biogas during the initial phase of the

treatment. After 5 days of the operational period, a drop in carbon dioxide percentage has been observed. At 10 days of treatment, the carbon dioxide percentage was found to be the lowest for every batch reactor. The lowest CO_2 content in the generated biogas was observed for S/I ratio of 1.0 after 10 days of the treatment. CO_2 concentration in the biogas began to rise after 10 days of anaerobic treatment, and at the end of 30 days, it had surpassed the methane content.



Figure 4.2 Variation in CO₂ % over a 30-day period for different S/I ratios

4.1.3 Optimum Substrate to Inoculum Ratio for Anaerobic Treatment of Food Waste

According to Liu et al. (2017) and Zhou et al. (2018), when the S/I ratio is higher than 3.0, monodigestion of a given substrate usually shows lower methane yield. However, good methane production performance could be found at S/I ratio between 1.0 to 1.5 in most mono digesters. Therefore, in this study, to evaluate and compare the influence of S/I ratios on methane content, maximum methane percentages from each S/I ratio were plotted in a graph to determine the optimum ratio for the anaerobic treatment of food waste. The maximum methane concentrations from six S/I ratios were shown in Figure 4.3. The error bars provided in Fig. 4.3 represent the standard deviation of measured methane content of each sample. The maximum methane contents from six batch reactors were found to be 51.5%, 70.5%, 89.2%, 72.7%, 73.2% and 64.5% for S/I ratios of 0.5,0.75,1.0,1.5,2.0 and 4.0, respectively. A study was conducted in Beijing, China using canteen food waste to investigate the characteristics of food waste containing different substrates to inoculum (S/I) ratios. In the experiment, it was observed that there was rapid acidification of the reactor as well as reduced biogas production with a higher S/I ratio (Li et al., 2018). Another study also reported that the more serious acidification occurs with the higher S/I ratio, thus leading to low biogas production (Li et al., 2015). Similarly in this study, methane content gradually increased with the S/I ratio, but after an optimal S/I ratio was reached, subsequent increases in S/I resulted in a decreasing methane concentration. The results obtained from the experiments indicated that a food waste-to-inoculum ratio of 1.0 had a high methane production potential. At the substrate-to-inoculum ratio of 1.0, methane concentration in the biogas significantly increased, reaching as high as 89%, substantially higher than the methane contents found for other S/I ratios.



Figure 4.3 Optimum S/I ratio for maximum methane production

4.2 MESOPHILIC ANAEROBIC TREATMENT OF FOOD WASTE AT DIFFERENT TOTAL SOLID PERCENTAGES

Anaerobic treatment of food waste is an interactive process requiring a coordinated interaction between several groups of microorganisms. This research developed a series of batch tests based on methane production and energy efficiency to assess and compare the anaerobic treatment potential of food waste. Anaerobic treatment of food waste was conducted under three conditions: wet treatment, semi-dry treatment, and dry treatment. Batch tests were examined with six different total solid percentages (5%, 10%, 12%, 15%, 18% and 20%) under mesophilic and psychrophilic temperatures. Batch reactors were fed with source-separated food waste of six different TS% over the course of 30-day time with a substrate-to-inoculum ratio of 1.0. The first batch of experiments was carried out under a mesophilic temperature range (30-35°C) with source-separated food waste and inoculum from a bench-top mesophilic anaerobic reactor. Prior to the treatment procedure, sample food waste was mechanically ground into smaller particles and diluted with distilled water. Particle size reduction of substrate provided more surface for microbial growth and ensured a homogenous mixture.

4.2.1 Particle Size Analysis of Food Waste Sample

The laser scattering particle size distribution analyser LA-950 was used to determine the particle size distribution of the food waste sample. The tests were carried out in a sequential manner a total of three times. Table 4.1 provides a summary of the test findings.

Source	Test	Median	Mean Size	Std. Dev	D60 value	D10 value	D90 Value
		size µm	μm	μm	μm	μm	μm
Food	1	929.353	179.411	380.5885	255	15.2415	929.353
waste							
Food	2	734.942	200.244	297.258	300	13.4879	734.942
waste							
Food	3	744.251	230.34	298.369	286	15.2658	744.251
waste							
Average		802.33	203.32	325.40	280.33	14.66	802.33
Ũ							

Table: 4.1 Summary of test findings from particle size analyzer LA-950

The outcomes of the tests were labelled as Test 1, Test 2, and Test 3. The average value of mean particle size was 203.32 µm with a standard deviation of 325.40 µm. Both particle size distribution curves and cumulative distribution curves for three test runs are shown in Figure 4.4. The red, green, and black line graphs demonstrated the outcomes from Test 1, Test 2, and Test 3, respectively. Particle size distribution curves illustrated the average particle size, the smallest particle size, and the largest particle size of the sample. The particle size distribution curve of Test 1 showed that the mean particle size of food waste sample was 179.42 μ m, the smallest particle size was 1.8 µm, and the largest particle size was 2100 µm. Similarly, for Test 2 the mean particle size of food waste sample was 200.24 μ m, the smallest particle size was 1.8 μ m, and the largest particle size was found to be 1700 μ m. Finally, for Test 3, the mean, the smallest, and the largest particle size were 802.33 µm, 1.78 µm and 1750 µm, respectively. The D10, D60, and D90 values were calculated using the cumulative passing curves for the three test runs shown in Figure 4.4. D10 values from Test 1, Test 2, and Test 3 were 15.2415 µm, 13.4879 µm, and 15.2658 µm, respectively. D60 readings from Test 1, Test 2, and Test 3 were 255 µm, 300 µm, and 286 µm, respectively. Finally, D90 values from Test 1, Test 2, and Test 3 were 929.353 µm, 734.942 µm, and 744.251 µm, respectively. The average D10, D60 and D90 values from three tests were found to be 14.66 μ m, 280.33 μ m, and 802.33 μ m, respectively. The average D10 value indicated that 10% of the sample diameter was equal to or less than 14.6 micrometers, the D60 value indicated that 60% of the sample diameter was equal to or less than 280.33 micrometers and the D90 value represented that 90% of the sample diameter was equal to or less than 802.33 micrometers.



Figure 4.4 Particle size distribution and cumulative frequency curves of food waste sample from three test results

4.2.2 Variation in CH₄% with Respect to Time (Temperature 30-35 °C)

Six batch reactors were fed with six distinct total solid percentages were kept in an anaerobic atmosphere under mesophilic temperature for 30 days until no significant methane production was observed. Here is a chart showing how methane content in the biogas changes over period. The methane percentage was determined at 5-day intervals since the beginning of the treatment

process. The methane content of the biogas generated by each of the six reactors during the whole treatment procedure is shown in Figure. For 5%, 10% and 12% of TS, methane content reached 50% within 5 days of treatment. After more than 15 days of operation, the biogas had a 50% methane concentration, with TS levels of 15%, 18%, and 20%. Therefore, methane peaks were observed after 15 days of operation period as VFA consumption increased with treatment time. The medium's acidification was neutralized by the high alkalinity, leading to a rise in pH and eventually resulted in the degradation of VFA. Methane peaks were reported to be 88%, 81%, 94%, and 89% for 5%, 10%, 12%, and 15% of TS after 15 days of digestion, respectively. After 20 days of operation, however, methane peaks were found to be 72.6% and 69% for 18% and 20% of TS for dry treatment conditions, respectively. In anaerobic digestion of food waste in the prototype anaerobic digester of two phases, it was found the contents of methane between 50.2 and 60.4% (Ratanatamskul et al., 2015). In this study, maximum methane contents ranged between 69% to 94% under mesophilic temperature which is higher than the mentioned study. For instance, to run a generator fuelled with biogas, it is essential to have CH₄ content greater than 50% (Lansing et al., 2008). Biogas, at 64–69% methane, has a total energy potential of 23–25 MJ/m³ (Statistics Canada, 2010). Therefore, methane content remained consistently in the suitable range which demonstrated that the process offered excellent quality of biogas, which remained fairly steady throughout the mesophilic treatment of food waste. Figure 4.5 exhibited that all line graphs provided similar trends. Methane concentration in the biogas showed an initial upward trend, up until the methane peaks, and then steadily decreased with time.



Figure 4.5 Methane percentage variation over a 30-day time period for different TS%

4.2.3 Variation in CO₂% in the Biogas with Time (Temperature 30-35 °C)

Changes in CO₂ concentration during the treatment process were shown in Figure 4.6. During the first several days of the treatment, CO₂ concentration was high in the biogas generated by each reactor. Since CH₄% steadily increased with the anaerobic treatment, the CO₂ concentration in the biogas reduced with time. CO₂ concentration was found to be lowest after 15 days of operation for 5%,10%, 12% and 15% of TS, whereas during dry anaerobic treatment (18% and 20% of TS), CO₂ concentration reached its lowest after 25 days of operation. Subsequent work in laboratory scale digesters has suggested that high ammonia concentrations may cause a shift in the biochemical pathways leading to methane formation (Banks and Zhang, 2010). The pH in the rectors started rising at the end of the treatment which indicated steady increase in the ammonia concentration in the anaerobic reactors. The increase in ammonia concentration eventually led to increase in CO₂ concentration. Therefore, a substantial increase in CO₂% in the biogas was observed for all six reactors at the end of the anaerobic treatment.



Figure 4.6 Variation in CO₂% with time for different TS%

4.2.4 Variation in Methane Contents at Different TS% (Temperature 30-35 °C)

The nature of the raw materials and the operational conditions used during anaerobic digestion, influences the CH₄ concentration and other chemical compositions of the produced biogas (Lombardi et al., 2013). To determine the optimum operational treatment condition for food waste, the maximum methane content of biogas from six different TS% under mesophilic temperature is presented in Figure 4.7. The error bars provided in Fig. 4.7 indicate the standard deviation of measured methane content of each sample According to the results shown in Figure 4.7, the methane content of the generated biogas significantly improved during semi-dry anaerobic treatment conditions. The data indicated that methane content in the biogas initially increased with an increase in total solid percentage, then gradually decreased with a further increase in TS% after the optimal total solid percentage has been reached. This might be caused due to the lower moisture content in waste with higher TS% which resulted in a reduced level of microbial activity such as methanogenesis (Igoni et al., 2007). The highest methane content in the biogas was found at 94.4% at 12% TS, which is almost 30% higher than the typical range of methane (40%-60%) in biogas.

Wet anaerobic treatment at 5% and 10% TS resulted in maximum methane contents of 78% and 81.8%, respectively, and dry anaerobic treatment at 18% and 20% resulted in maximum methane contents of 72.6% and 69.1%, respectively. According to Rajagopal et al. (2016), feed solid content of 10% to 15% helped to maintain good buffering capacity in the anaerobic digester. Good buffering capacity prevents acidification in the digester and results in higher methane production. Previous studies by Itodo and Awulu (1999) showed that substrates of higher TS% were more prone to acidic conditions. Similarly in this study, methane contents found during semi-dry condition (12% and 15%TS) were considerably higher than the wet and dry anaerobic treatment conditions.



Figure 4.7 Variation in CH₄% at different total solid percentages under mesophilic temperature

4.3 PSYCHROPHILIC ANAEROBIC TREATMENT AT DIFFERENT TS% (TEMPERATURE 18-22 °C)

Mesophilic temperature (30-35°C) is considered the ideal temperature for the anaerobic treatment technology. Figure 4.7 shows that the anaerobic treatment of food waste under mesophilic temperature resulted in greater bioconversion and higher methane percentage in the biogas. In

Canada and some portions of the USA, where winter temperature drops under sub-zero temperatures, maintaining a mesophilic condition in the treatment facility is challenging. The largest portion of heating demands in the digester operation is substrate heating to achieve mesophilic or thermophilic condition. It requires over 90 % of all heating demands, and only up to 10 % is required for heat loss compensation (Zupancic and Ros, 2003). Sometimes for mesophilic treatment, it requires a significant amount of energy for additional heating, and considerable energy goes for particle size reduction. Considering this climatic condition, psychrophilic anaerobic treatment can be an effective alternative that can maximize the net energy potential. It is to be noted that the psychrophilic temperature range can have a strong detrimental effect on the rate of hydrolysis of lipids and proteins, and the lipid hydrolysis is reported to commence only after perfect methanogenic conditions are established in the anaerobic digester (Lettinga et al., 2001). All biological processes in the anaerobic treatment are temperaturedependent and a decrease in operating temperature will inevitably affect not only the bacterial metabolism but also the bacterial reaction kinetics (Zhou et al., 2018). Psychrophilic temperature is not favourable for microorganism growth which can lead to the accumulation of VFA and ultimately result in the failure of the process. However, several studies indicate that there are more diverse methanogenic archaea at psychrophilic temperatures than at mesophilic temperatures (Dhaked et al., 2010). To increase the methane concentration in the biogas and enhance system efficiency at a lower temperature, the effect of biogas recirculation on the performance of anaerobic treatment was investigated in this research work. Therefore, two sets of six batch reactors were kept at psychrophilic temperature for 30-day time to assess the feasibility of psychrophilic treatment of food waste and to examine the impact of biogas recirculation on methane production. The first set of psychrophilic batch digesters was operated without biogas

recirculation. Another set of six reactors was evaluated with recirculation of the produced biogas into the anaerobic system. Similar to the previous mesophilic anaerobic treatment, six different TS% were fed into psychrophilic batch reactors while maintaining a substrate-to-inoculum ratio of 1.0. The experiment outcomes showed that biogas recirculation improved methane production and led to a more reliable and energy-efficient system. The amount of energy utilized for particle size reduction was kept to a minimum in order to optimise the net energy gain. No mechanical particle size reduction was used to break down the larger particles. Manual chopping or cutting was applied to eliminate the larger particles and create a uniform mixture. Sample particle diameters were approximately varied between 1.5 mm to 19 mm and the majority of the sample particles was greater than 2mm.

4.3.1 Particle Size Distribution by Sieve Analysis:

Sieve analysis was performed to evaluate the particle size distribution of the food waste sample. Results from the sieve analysis were presented in the semi-logarithmic plots known as particle size distribution curves. Particle diameters were plotted in the logarithmic scale, and the corresponding percent finer was plotted in the linear scale.

Based on the particle size distribution curve, D30, D60 and D90 values were found at 5.4 mm, 7.5 mm, and 10 mm, respectively. D30, D60 and D90 were the particle sizes corresponding to 30%, 60% and 90% finer materials on the cumulative particle size distribution curve, respectively. D30 was 5.4 mm, which meant 30% of the particle diameter in the sample had a size of 5.4mm or less. Similarly, D60 was 7.5mm that represented 60% of the sample particle size less or equal to 7.5 mm. Finally, the D90 result demonstrated that 90% of particles in the sample had a size of 10 mm or less. Sieve analysis is more suitable for the particle sizes greater than 2mm. On the other hand, laser particle diffraction can accurately measure the particles between 0.1 μ m- 2500 μ m. The mean

particle size found from sieve analysis was 6.9mm whereas from the previous particle size distribution test, mean particle size was found 203.32 μ m. The ratio of D90 and D60 values between sieve analysis and LA-950 were 12.5: 1.0 and 26.7:1, respectively. D90 and D60 values from sieve analysis were 12.5 times and 26.7 times higher than those from the prior test, respectively.



Particle Size Diameter (mm)

Figure 4.8 Particle size distribution food waste sample by sieve analysis (ASTM Standard)

4.3.2 Variation in CH₄% with Respect to Time (Temperature 18-22 °C)

The methane percentage was calculated every five days interval until the end of the treatment process. The methane content of the generated biogas from two sets of batch reactors across the treatment process is shown in Figure 4.9. Figure 4.9(a) shows the changes in methane content in the biogas without any recirculation at psychrophilic temperature, and Figure 4.9(b) shows the changes in methane content with time with biogas recirculation into the anaerobic system. The data sets in both figures showed the same trends. Methane concentration in the biogas

progressively began to rise during the treatment process, and then, after reaching the peak value, methane content began to fall as the treatment process continued. The results obtained from the experiments proved that biogas recirculation significantly enhanced the methane production and methane concentration in the generated biogas. With recirculation, the purity of produced methane increased within the first week of the treatment, reaching approximately $50\pm10\%$. On the contrary, Figure 4.9(b) showed that at psychrophilic temperature without biogas recirculation, methane content varied between 10% to 15% within a week of the treatment process. Without recirculation, methane peaks were observed after 25 days of the treatment and maximum methane content was found to be 45% at 12% of TS, which is lower than the typical range. The data from the second experiment illustrated that recirculation of biogas in the anaerobic reactor raised the methane portion in the biogas by more than 50%. During the second set of experiments, methane peaks for 5%, 12%, 15%, 18% and 20% TS were observed after 20 days of operation, whereas at 10% TS the methane peak was found at 15 days. This delay could be related to the time required for the onset of methanogenic reactions, leading to biogas production during anaerobic digestion. That could be the step for breaking complex organic matter into smaller molecules via the enzymes released by the bacteria. Figure 4.9(b) demonstrated that methane content in the biogas initially showed an upward trend with time. For 5%, 10% and 12% TS, methane content reached 50% within five days of the treatment. For 15%, 18% and 20% TS, the process required more than 15 days to reach 50% methane content in the biogas. Subsequently, as volatile fatty acid (VFA) was consumed with the treatment time, methane peaks were observed after 15 days of the operation period with biogas recirculation. The high alkalinity overcame the acidification in the medium, which increased the pH value and eventually resulted in the degradation of VFA. For 5%, 10%, 12% and 15% of TS methane peaks were found at 88%, 81%, 97% and 89% after 15 days of treatment. On the other hand, for dry treatment conditions, 18% and 20% of TS, methane peaks were found to be 72.6% and 69%, respectively, after 20 days of operation.



Figure 4.9 Variation in methane percentage with time under psychrophilic temperature (a) without biogas recirculation and (b) with biogas recirculation

4.3.3 Variation in CO₂% with Time (Temperature range 18-22 °C)

In the first set of experiments, the CO₂ content in the biogas comprised up to 90% of biogas during the initial stage of treatment process (Figure 4.10). However, the typical range of CO₂% in biogas varies between 30%-40%. With the progression of anaerobic treatment, CO₂% started declining with time until 25 days of operation, when the CO₂% was found to be the lowest. The lowest CO₂ content for TS 5%, 10%, 12%, 15%, 18% and 20% were observed at 61.1%, 58.7%, 54.1%, 61.7%, 74.1% and 74.2%, respectively without recirculation. Results from psychrophilic anaerobic treatment without biogas recirculation demonstrated that CO₂ comprised the maximum component in the produced biogas. However, biogas recirculation significantly reduced the CO₂ level in the biogas. Obtained data from both sets of experiments showed a similar pattern. At the initial stage of the treatment process, CO₂% was higher, which eventually started declining with the treatment process. Then again, after reaching a certain point, CO₂% began to rise in the generated biogas. With biogas recirculation, at 5%, 10%, 12%, 15%, 18% and 20% of TS, CO₂% was observed to be the lowest after 20 days of time, and at 10% TS, CO₂% was the lowest after 15 days of time. The lowest CO₂% were observed to be 17.9%, 6.4%,19.2%, 33.1%, 27.3% and 29.6% for 5%, 10%, 12%, 15%, 18% and 20% TS, respectively with biogas recirculation. Therefore, with recirculation, CO₂ content decreased by more than half in the biogas composition.



Figure 4.10 Variation in CO₂% with time for different TS% under psychrophilic temperature (a) without biogas recirculation and (b) with biogas recirculation

4.3.4 Variation in Methane Content at Different TS% with and without Recirculation

(Temperature18-22 °C)

Anaerobic treatment with biogas recirculation substantially increased the methane content in the biogas composition. Maximum methane contents from each TS% were plotted in Figure 4.11 to assess the optimum anaerobic treatment condition under psychrophilic temperature. The error bars provided in Fig. 4.11 represent the standard deviation of maximum methane content of each sample. The results indicated that without biogas recirculation, methane percentage did not differ considerably with the change of TS%. On the other hand, methane content varied between 68%-93% with the change in TS% for the later set of experiments. The results from the two studies showed that the biogas's methane content initially increased; however, after attaining the optimum total solid percentage, methane percentage gradually decreased with an increase in TS%. Without recirculation, methane percentages were found to be 38.9%, 41.3%, 45.8%, 37.2%, 25.5% and 25.9% for 5%, 10%, 12%, 15%, 18% and 20% TS, respectively. At the same treatment condition and temperature with the biogas recirculation technique, methane percentages were found to be 82%, 93.6%, 80.9%, 72.8%, 72.6% and 68.4% for 5%, 10%, 12%, 15%, 18% and 20% of TS respectively. Hence, 52.6%, 55.8%, 43.4%, 48.9%, 64.8% and 62.1% rise in methane content were observed with recirculation for 5%,10%, 12%, 15%, 18% and 20% of TS, respectively. Methane percentage was the highest, 45.8% at 12% TS without recirculation, whereas the highest methane concentration was 93.6% at 10% TS with biogas recirculation. High methane content also suggests that the methanogenic population in the digesters were well developed (Lansing et al., 2008). The methane concentration found from the recirculation technique is substantially greater than the methane contents reported in the literature for psychrophilic AD of food waste (Martí-Herrero et al., 2019; Muñoz et al., 2020). The most vital step of psychrophilic process, containing the highest risk of failure, is initiation of biodigester operation (Dhaked et al., 2010). In this case, the biogas recirculation technique helped to accelerate the initiation time and could alleviate any deterioration in methane production performance caused by the low operation temperature. Hence, this approach eventually led to a stable and energy-efficient anaerobic system.



Figure 4.11 Variation in CH₄% at different total solid% with or without biogas recirculation

4.4 pH TREND DURING ANAEROBIC TREATMENT PROCESS

pH significantly influences the hydrolysis step, which is a rate-limiting step for the anaerobic treatment process. Figures 4.12 and 4.13 represented the analysis of the digester pH during the mesophilic and psychrophilic data collection period, respectively. In anaerobic digestion, optimum pH for the methanogenic microorganisms is between 6.5 and 7.5 (Gregor et al., 2012). Fig. 4.12 showed that mesophilic treatment was typically operating under stable conditions. The initial pHs for all batch experiments were kept the same to evaluate the pH fluctuations during the anaerobic treatment process. During mesophilic treatment, the final pH values from the six reactors were 6.8, 6.3, 6.7, 6.2, 6.9, 7 for 5%, 10%, 12%, 15%, 18% and 20% TS, respectively, which were within a

stable neutral range (Gunaseelan et al., 2004). The pH trend in Fig. 4.13 reflected that psychrophilic anaerobic treatment with biogas recirculation made the system more stable and efficient. During the first set of psychrophilic experiments, the final pH for 5%, 10%, 12%, 15%, 18% and 20% TS were observed at 4.3, 3.9, 4.0, 4.2, 4.0 and 3.8, respectively. The decrease of the pH proved that acid production in the system increased. This could lead to partial or total inhibition of the methanogenic microorganisms, which might cause stresses in the system, especially regarding methane production (Amani et al., 2010). Conversely, with biogas recirculation, the final pH values for 5%, 10%, 12%, 15%, 18% and 20% TS were 6.3, 6.6, 6.5, 6.2, 6.6 and 7, respectively. These pH values were within the optimum limits for methanogens operation which was an indicator of efficient reactor performance (6.0-8.2) (Kondusamy and Kalamdhad, 2014). The initial pH was above 6.5, and a decreasing trend of pH in the process was observed in the first few days of the digestion for both mesophilic and psychrophilic treatment. The pH drop was caused due to intense hydrolysis rate and volatile fatty acid (VFA) formation. Several studies similarly reported that after first week of anaerobic treatment, effluent pH began to gradually decrease indication an accumulation of volatile fatty acids (VFAs) (Anderson et al., 1992, Rao et al., 2000). However, the pH increased to its standard operating value after VFAs metabolism. The above pH profiles indicated that reactors subjected to mesophilic and psychrophilic treatment with biogas recirculation appeared to be operating in a healthy condition, as pH within the range 6.0 to 7.5 was a good indicator of efficient rector performance (Wheatley, 1991).



Figure 4.12 pH trend during mesophilic treatment over a 30-day period



Figure 4.13 pH trend during psychrophilic treatment over a 30-day period

4.5 TEMPERATURE PROFILE FOR MESOPHILIC AND PSYCHROPHILIC TREATMENT CONDITIONS

Batch experiments with six total solid percentages were conducted under mesophilic and psychrophilic temperatures. Batch digesters were placed in the incubator to maintain mesophilic temperature (30-35 °C), and reactors were kept at atmospheric temperature for psychrophilic conditions (1-20°C). Each day, a temperature reading was taken for 30 days to create the temperature profile. The variation in daily temperature during mesophilic and psychrophilic

treatment are presented in Figs. 4.14 and 4.15, respectively. The lowest temperature reading of 30°C was obtained on the 12th and 20th day, while the highest of 35°C was recorded on the 14th day of the mesophilic treatment process. The average temperature of 32.9°C was recorded at the end of the 30-day digestion time. Similarly, the lowest temperature reading of 18°C was obtained while the highest temperature was recorded at 22°C during psychrophilic treatment. The average temperature of 19.8 °C was recorded at the end of the 30-day digestion.



Figure 4.14 Temperature profile during mesophilic treatment process



Figure 4.15 Temperature profile during psychrophilic treatment

4.6 EFFECT OF TEMPERATURE AND BIOGAS RECIRCULATION ON COD REMOVAL AT DIFFERENT TS%

In order to investigate the impact of total solid percentage, temperature, and recirculation of biogas on the removal of COD, the COD was tested under mesophilic and psychrophilic temperatures at TS 5%, 10%, 12%, 15%, 18% and 20% with and without biogas recirculation. The results of COD removal with and without biogas recirculation at different TS values at the mesophilic and psychrophilic temperatures are presented in Figure 4.16. The error bars provided in Fig. 4.16 represent the standard deviation of measured COD removal of each sample. The highest COD removal was observed at 12% TS at psychrophilic temperature, while at mesophilic temperature, maximum COD removal was observed at 15% TS. Without any biogas recirculation, the highest COD removal was found to be 5461 mg/l, while maximum COD removal was increased to 6269 mg/l with biogas recirculation. Wang et al. (2014) and Cakir et al. (2005) reported that COD removal increases with the increase in treatment temperature. Nevertheless, the results show that maximum COD removal at mesophilic temperature was 5923mg/l at 15% TS. Thus, the results

demonstrated that the recirculation of biogas has a considerable impact on increasing the COD removal efficiency for anaerobic treatment. Similar trends were observed whereby COD removal started decreasing after 15% TS and the effect of TS% on COD removal became insignificant during dry anaerobic treatment.



- - - psychrophilic temperature with methane recirculation

Figure 4.16 COD removal after anaerobic treatment of food waste

4.6.1 COD Removal Efficiency

The results of overall COD removal efficiency at different conditions are summarized in Figure 4.17. The error bars provided in Fig. 4.17 denote the standard deviation of measured COD removal efficiency of each sample. The effect of operating temperature, total solid percentage, and biogas recirculation on the COD removal efficiency was investigated. The results illustrated that all samples showed similar efficiency trends for COD removal after 30 days of the treatment process. COD removal was maximum at 10% TS for mesophilic and psychrophilic temperatures with or

without biogas recirculation. The maximum COD removal of 78.9% was achieved under mesophilic anaerobic treatment condition which was comparable to the COD reduction (73.7%) operating the anaerobic digestion of food waste in mesophilic scheme for a hydraulic time (HRT) of 27 days in prototype anaerobic digester (Ratanatamskul et al., 2014). The data showed that COD removal efficiencies have decreased during semi-dry conditions compared to wet digestion. During dry digestion, COD removal efficiency became the lowest in all conditions. A study showed that a cumulative total COD removal efficiency of around 70% was achieved in a hybrid anaerobic reactor operating under psychrophilic temperature (Tiwari et al., 2021). However, in this work, the highest COD removal efficiency was found to be 81.9% at 10%TS under psychrophilic temperature with biogas recirculation, while the minimum was 34% at 18% TS for psychrophilic temperature without any biogas recirculation. In conclusion: firstly, both TS% and temperature impact COD reduction. Secondly, the recirculation of biogas improved the overall COD reduction efficiency for all treatment conditions. Finally, recirculation of biogas under psychrophilic temperature and 10% TS provided the optimum conditions in this study.



Figure 4.17 COD removal efficiency under different operating conditions

4.7 EFFECT OF TEMPERATURE AND BIOGAS RECIRCULATION ON VS

REDUCTION AT DIFFERENT TS%

Volatile solid reduction at various TS percentages with or without biogas recirculation at mesophilic and psychrophilic temperatures are shown in Figure 4.18. The error bars provided in Fig. 4.18 represent the standard deviation of VS reduction of each sample. During mesophilic treatment, VS reduction were observed 76.5%, 86.9%, 91.6%, 90.0%, 76.6%, and 83.7% for 5%, 10%, 12%, 15%, 18% and 20% TS respectively. VS reductions under mesophilic condition were in the range of 73%–99% which is typical for anaerobic digestion of food waste (Kayhanian et al., 1995). Under psychrophilic temperature, VS reduction was 74.9% after 30 days of operation at 5% TS with biogas recirculation, while VS reduction was 52.9% without recirculation. At 10% TS, these values changed to 71.6% and 67.5%. At 12% TS, VS reduction were observed 92.4% and 61.6% with or without recirculation, respectively. Similarly, for 15% TS, 18% TS, and 20% TS,

VS reduction was 87.9%, 76.7% and 77.6%, respectively, with biogas recirculation, while without recirculation, VS reduction for 15%,18% and 20% TS were dropped down to 74.2%, 73.8% and 60.7% respectively. The higher removal efficiency of VS was a very good indication of high uptake rate of the organic fraction of total solids by methanogenic bacteria (Minale et al., 2014). The highest VS reduction of 92.4% was observed at 12% TS for psychrophilic anaerobic treatment with biogas recirculation. A study of psychrophilic anaerobic treatment of food waste reported an 87% reduction in volatile solids (VS) in semi-dry conditions, which is compatible with this research production (Rajagopal et al., 2016). These results consistently showed that biogas recirculation into the anaerobic reactor could improve VS reduction without needing additional heating. The figures showed that higher VS reduction were observed for at 10%, 12% and 15% TS for all treatment conditions.



CH4% at psychrophilic temperature without recirculation



4.8 METHANE PRODUCTION DURING ANAEROBIC TREATMENT OF FOOD WASTE

4.8.1 Effect of Temperature, TS% and Biogas Recirculation on Methane Content in the

Biogas Composition

The purity of produced methane was low during the initial stage of treatment and gradually increased during batch tests. To evaluate and compare the effect of temperature, TS% and biogas recirculation on methane purity in the biogas, maximum methane percentages were plotted against TS%, shown in Figure 4.19. The error bars provided in Fig. 4.19 represent the standard deviation of measured methane content of each sample. Methane contents of the generated gas under mesophilic and psychrophilic temperatures with or without biogas recirculation are shown in Figure 4.19. It demonstrated that maximum methane content was observed at 94.4% at 12%TS under mesophilic treatment. On the other hand, during psychrophilic temperature, maximum methane content was 93.6% at TS 10% with biogas recirculation. The maximum methane percentage of the produced gas was found to be 45.9% without recirculation. Costa et al. (2009) presented that methane generation increased with the increase of temperature. However, the experimental findings proved that with biogas recirculation, methane content in the produced biogas from psychrophilic treatment was almost similar to that of mesophilic treatment. The data showed that the effect of TS% on methane content became insignificant during dry anaerobic treatment condition. Overall, methane concentrations in biogas were highest during wet and semidry digestion.



Figure 4.19 Effect of temperature, total solid% and biogas recirculation on methane content in the biogas

4.8.2 Methane Production Yield (L CH₄/g COD removed) to Evaluate System Efficiency

The experimental methane generation yield for mesophilic anaerobic treatment fluctuated between 0.19 to 0.26 L CH₄/ g COD removed, where maximum and minimum methane yield was observed at 12% and 20% TS respectively (Figure 4.20). The error bars provided in Fig. 4.20 represent the standard deviation of measured methane yield of each sample. During psychrophilic anaerobic treatment, methane yield varied between 0.08 to 0.12 L CH₄/ g COD removed without any biogas recirculation. In comparison, biogas recirculation resulted in a higher methane production, and varied between 0.17 to 0.23 L CH₄/ g COD removed. For example, at psychrophilic environment without biogas recirculation, the minimum methane generation yield for food waste was found to be 0.08 L methane/g COD removed during dry treatment condition, while at mesophilic environment, the maximum methane yield was observed to be 0.26 L methane/g COD removed during semi-dry treatment condition. The theoretical methane yield is 0.35 L/g COD removed (Michaud et al., 2002) at 0 °C and pressure of 1 atm. Because this work used two temperature

ranges, theoretical methane yield needed to be corrected for the applied temperature. Maximum theoretical methane yield for mesophilic temperature (30-35 °C) was obtained at 0.364 L per g COD, and maximum theoretical methane yield for psychrophilic temperature was obtained at 0.378 L per g COD. Though it is not uncommon for actual methane yield to be below the theoretical, this is perhaps indicative of the bench-top reactor's operational limit of efficiency, which may not be the case in a more efficient reactor design such as an expanded bed reactor or commercial anaerobic digester. In addition to that, the lower methane generation yield in the batch anaerobic reactor is related to the condition of biomass used for the treatment process. The food waste sample was used after one month of storage, while in treatment plants, biomass would be more acclimated to the waste and used without storage. Methane generation yield graphs for all treatment conditions demonstrated similar patterns. The maximum methane yield was obtained from the AD of food waste with semi-dry condition (Dhungana and Lohani, 2020). Likewise, in this study maximum methane yields were also observed for semi-dry treatment conditions, while minimum methane yields were found during dry treatment.



Figure 4.20 Effect of temperature, total solid% and biogas recirculation on methane yield

Current Study									Other Studies Related to Food Wate Treatment			
Treatment Temperature range		Mesophilic Anaerobic Treatment		Psychrophilic Anaerobic Treatment without Biogas Recirculation		Psychrophilic Anaerobic Treatment with Biogas Recirculation		Parameter		References		
								CH4% in biogas	Methane yield			
		% in	ield)D rmv	% in	ield)D rmv	% in	ield)D rmv		0.4-0.49 LCH ₄ /VS	Heo et al., 2016		
Anaerobic Treatment Conditions		Max. CH4% biogas	Methane yi LCH4/g CO	Max. CH4% biogas	Methane yi LCH4/g CC	Max. CH4% biogas	Methane yi LCH4/g CC	55-65%	0.44 LCH ₄ /VS	Zhang et al., 2011		
Wet	5% TS	79	0.2	39	0.1	82	0.18	65-85%		Yong et al., 2015		
	10% TS	81.7	0.22	41.3	0.11	93.6	0.2	60-75%		Banks et al., 2010		
Semi	12% TS	94.4	0.26	45.9	0.09	80.8	0.21	40-70%	0.3-0.4 m ³ kg-1 of VS	Zupančič et al., 2012		
Dry	15% TS	89.3	0.25	37.2	0.12	72.8	0.23	40-60%	0.05-0.12 LCH4/g COD rmv	Minale et al., 2014		
Dry	18% TS	72.7	0.22	25.5	0.08	72.6	0.19	60-70%		Chen et al., 2010		
	20% TS	69.2	0.19	25.9	0.08	68.4	0.17	65-75%	0.18-0.52 m ³ kg-1 of VS	Gunaseelan et al., 2004		

Table 4.2 Overall summary of results and comparison to other outcomes reported in literature.

CHAPTER 5 CONCLUSIONS

5.1 CONCLUSIONS

The waste generated by increased urbanization and population will soon become unmanageable. Cities worldwide need more places to send their waste, as landfills are reaching their limits and closing down. The organic fraction of municipal solid waste constitutes the central part of the methane produced from landfilling and is a potent greenhouse gas. Anaerobic treatment could be used to generate heat and electricity from the organic waste in the urban environment and reduce the amount of waste sent to landfills. Although widely used for wastewater treatment, anaerobic digestion technology has yet to be applied much to mixed food waste, even though it is one of the most energy-dense substrates. This thesis aims to present anaerobic treatment as a mean of extracting methane from food waste and using it as renewable energy. Moreover, apart from traditional reactors, this study also investigates a novel strategy for enhanced methane generation at lower temperatures. The following conclusions can be drawn based on this study's results:

1. Anaerobic biological process shows significant potential to produce methane gas through the treatment of food waste at both mesophilic (30–35°C) and psychrophilic (1–20°C) temperatures. According to the study, anaerobic treatment of food waste at psychrophilic temperature exhibited nearly similar efficiency to that at mesophilic temperature in producing methane when subjected to biogas recirculation. Such a biogas recirculation technique is the major contribution of this study since this novel approach could ensure an energy-efficient food waste management system, especially in cold regions.

2. Among three treatment conditions i.e., wet, semi-dry, and dry anaerobic treatment, the semi-dry anaerobic treatment condition shows considerably higher methane concentration in the produced biogas at both mesophilic and psychrophilic temperatures.
3. The maximum methane concentration has been found to be 94% after 15 days of the mesophilic anaerobic treatment process, which is almost 30% higher than the typical range of methane (40%–60%) in the biogas.

4. The maximum methane percentage has been found to be 45.8% after 25 days at psychrophilic temperatures without biogas recirculation. On the other hand, the methane concentration in the biogas increased to 93.6% after 15 days with biogas recirculation at psychrophilic temperature, which is comparable to that of mesophilic anaerobic treatment.

5.Mesophilic anaerobic treatment shows a maximum COD removal efficiency of 78.9%, while the highest COD removal efficiency of 81.9% has been found under psychrophilic anaerobic treatment with biogas recirculation. Therefore, psychrophilic anaerobic treatment with biogas recirculation exhibits similar COD removal efficiency.

6. Mesophilic anaerobic treatment shows a maximum methane yield of 0.26 L CH_4/g COD removed whereas psychrophilic anaerobic treatment results in a maximum methane yield of 0.23 L CH_4/g COD removed.

7. Maximum volatile solid destruction of 92% was achieved with biogas recirculation at psychrophilic temperatures. On the other hand, mesophilic anaerobic treatment shows a maximum reduction of 91%VS.

8. In addition, the biogas recirculation technique eliminates the requirement for energy intensive mechanical particle size reduction.

This study introduces a novel approach of anaerobic treatment of treating food waste to produce methane through biogas recirculation. Therefore, in contrast to the common notion that maximum methane production in anaerobic treatment occurs in the mesophilic temperature range, this study shows experimental evidence proving that substantial biomethane production can also be possible under psychrophilic conditions. The anaerobic treatment of food waste under psychrophilic temperature range is far more energy efficient than mesophilic processes particularly in cold regions. However, further study will be required to have a comprehensive understanding of the proposed approach of food waste treatment.

5.2 RECOMMENDATIONS FOR FUTURE WORK

Anaerobic treatment technologies pose much higher sustainability features than conventional, nonsustainable, energy-intensive treatment systems. Several recommendations are presented below to improve the developed method's efficiency and expand its applications.

1. More study is required using different types of reactors and other food waste composition.

2. More efforts should be made to divulge the limitations of bacterial-mediated processes for psychotropic conditions. The syntropic interaction between various groups of bacteria and archaea should be explored to identify the role of each community in the mixed consortium. (McKeown et al., 2009).

3. Each AD step's kinetic and thermodynamic considerations should be studied. Also, a mathematical model can be incorporated to predict and address issues related to upscaling and optimization.

4. Hence, proper techno-economic studies along with comparative life cycle assessment (LCA) studies and economic feasibility studies should be conducted before upscaling the technology and implementing it in the real world (Garca Sanchez and Güereca, 2019; Li and Yu, 2016).

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Appendix



Figure A 1 Reference curve for methane content of the biogas obtained by gas chromatography.