Treatment of Small-Scale Brewery Wastewater – Anaerobic Biochemical Methane Potential (BMP) Trials and Moving Bed Biofilm Reactor (MBBR) Field Study

by

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Abstract

As the microbrewery industry expands, disposal of brewery wastewater is becoming more of a concern, both for brewery operators and for local municipal wastewater authorities. Brewery wastewater is characterized as containing high strength organics and high variability in both organic and hydraulic loading. This high variability increased the challenge of treating brewery wastewater properly. Therefore, it is significant for optimizing the operation condition for the small-scale wastewater treatment system. This study conducted a batch study and a field study for optimizing a craft brewery on-site wastewater treatment system, which is equipped with two moving bed biofilm reactors (MBBR).

In the batch study, a two-factor Box-Wilson central composite design (CCD) was adopted to find optimum biomethane production conditions for the digestion of brewery wastewater with a dairy manure inoculum. The effects of two major influencing factors of temperature (T) (25-49°C) and brewery wastewater concentration (BWC) (2-9 g VS/L) on biochemical methane potential (BMP) (CH_4 yield) and CH_4 maximum production rate (R_{max}) were evaluated by applying response surface methodology (RSM). All of the trials presented a high organic removal efficiency with volatile solid (VS) 82-91%, soluble chemical oxygen demand (sCOD) 77-88%, and total chemical oxygen demand (tCOD) between 47% -76%. The experiment result suggested that the first order kinetic rate constant and biogas content (methane percentage in the biogas) can be affected by the temperature. The mesophilic regime had the highest average rate constant, and psychrophilic regime rate constant was significantly lower than the mesophilic and thermophile regime. The conditions in the thermophile range present a high variability for the first order rate constant. The methane ratio in the biogas increased and stabilized by the operation time. Mesophilic and thermophilic regime obtained a stabile biogas content around 25 days, and psychrophilic regime spent extra times to stabilized. At the end of the anaerobic digestion, the psychrophilic, mesophilic, and thermophilic regime had an average methane percentage 47%,65%, and 67% respectively. Optimum BMP and R_{max} were achieved under conditions of 49 °C and BWC of 5g VS/L. Correspondingly, the BMP and R_{max} were 141.40 mL CH_4/g VS added and 36.5 mL CH_4/ day, respectively. However, by pursuing a stability the preferable operational condition T=35℃ and

BWC=5 g/L is recommended, at this condition methane yield is 110.07 CH_4/g VS added and maximum methane daily production is 28.06 CH_4/ day, which is similar to the maximum result.

In field study, an on-site brewery wastewater treatment system equipped with two MBBR reactors was evaluated from October 12th, 2018 to February 10th, 2020 in Beau`s All-Natural Brewing Company, Vankleek Hill, Ontario, Canada. The aim of the study was to characterize the wastewater production (flow and organic loading rate), evaluate operating conditions and performance of the MBBR system and recommend improvements. Discharge from the brewery is highly variable for both organic and hydraulic loading with flow balancing recommended. The MBBR full-scale reactors operated at relatively stable conditions at surface area loading rate (SALR) of less than 25 $g/m^{2}\cdot d$ and dissolved oxygen (DO) greater than 2mg/L. Kinetic rate constants for suspended growth and attached growth biomass in the reactors were found to be similar at 0.0764-0.0908 h^{-1}, however, much larger attached growth mass in the reactors suggests that only a fraction of the attached growth biofilm is active. Effluent recycle was shown to be effective at controlling filamentous bacteria (type-0041) sludge bulking, reducing suspended solid concentration, and sCOD concentration.

Acknowledgment

I want to thank my supervisor, Dr Chris Kinsley, who provided me with the opportunity to work on this research project. During my research study, he provided me with excellent academic guidance and sufficient patience. Other special thanks go to Beau's All Natural Brewing Company for the uninhibited access to their on-site wastewater treatment system and the assistance from their staff throughout the project. I would like to thank Dr Robert Delatolla for giving permission for me to use his lab and lab equipment. I would also like to thank the environmental technical officer Patrick M D'Aoust for giving me technical support throughout my experiments.

I want to sincerely thank my family, especially my mom Azhatiguli Airexi, for providing their unconditional love, supporting me financially and emotionally. Thank my friend Guliziba Juma, who always be my side and provide her support and love. Thanks to my friend Shruti Tanga, Richard Hérard, Rafael Garduno and Sanad Nashad for providing help during my research

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List of Abbreviations

Anaerobic/ Anoxic/Oxic	A^2/O
Ammonia	NH_3, NH_4-N
Anaerobic Digestion	AD
Anaerobic Sludge Bed	UASB
Anaerobic Upflow Blanket Filter	AUBF
Attached Growth	AG
Biochemical Methane Potential	BMP
Biochemical Oxygen Demand	BOD
Brewery Wastewater Concentration	BWC
Carbon Dioxide	CO_2
Central Composite Design	CCD
Chemical Oxygen Demand	COD
Clean-In-Place Processes	CIP
Dissolved Oxygen	DO
Hydraulic Retention Time	HRT
Hydrogen	H_2
Hydrogen Sulfide	H_2S
Inoculum to Substrate ratio	I/S
Microbial Fuel Cells	MFC
Mixed Liquor Volatile Suspended Solid	MLSS
Methane	CH_4
Methane maximum production rate	R_{max}

Moving Bed Biofilm Reactor	MBBR
Moving Bed Biofilm Reactor 1	MBBR1
Moving Bed Biofilm Reactor 2	MBBR2
Organic Loading Rate	OLR
Over-Strength Discharge Fees	ODFs
Orthophosphate	SRP
Recycle Strategy 1	S1
Recycle Strategy 2	S2
Response Surface Methodology	RSM
Soluble Chemical Oxygen Demand	sCOD
Surface Organic Loading Rate	SALR
Surface Organic Removal Rate	SARR
Suspended Growth	SG
Temperature	T
Total Chemical Oxygen Demand	tCOD
Total Suspended Solids	TSS
Volatile Fatty Acids	VFA
Volatile Solid	VS
Wastewater Treatment System	WWTS
Water	H_2O

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CHAPTER 1

INTRODUCTION

1.1 Background

The brewing industry is known to be a large consumer of water and energy (Olajire, 2012). Twenty year of innovation improved the current resource requirements of the industry, but additional research is necessary to reduce the consumption to a more lower level (Fillaudeau et al., 2006; Olajire, 2012). From an environmental prospective, one of the main problems the industry is facing are currently the low water use efficiency, resulting in a low beer to water ratio and large quantities of wastewater produced (Russ & Schnappinger, 2007). Typical beer to water ratio for modern breweries is in the range of 1:6, ratios as high as 1:10 has been reported for breweries using more primitive operations (Brewers Assiociation, 2013; Brewers Association, 2011; Van der Merwe & Friend, 2002). This low efficiency is pressuring breweries operating regions stressed with water availability (European Commission, 2006; Olajire, 2012; Simate et al., 2011). Large quantities of organic rich wastewater are produced during the brewing process (Brito et al., 2007). Due to the high organic content of brewery wastewater, it is necessary to manage wastewater properly as disposal of untreated or partially treated wastewater can result in severe environmental degradation of receiving water bodies (Eyvaz, 2016; Simate et al., 2011). From an economic perspective, excessive use of water will erode profit. An additional cost to many small-scale brewing operations is over-strength discharge fees (ODFs) resulting from the discharge of strong wastewaters. ODFs are calculated based on the type and concentration of that pollutant exceeding the by-law sewer limits (Stantec Ltd., 2012). To encourage breweries to reduce the concentration of the wastewater discharged in municipal systems, municipalities are issuing more demanding ODFs. In recent years, the increase in environmental awareness among the consumers has also pressured business to decrease their environmental footprint. This social trend encourages breweries adapt more sustainable methods to reduce their environmental impacts (Couillard, 2019).

1.2 Study Objectives

The objective of this study is to provide practical strategies to improve the treatment efficiency of small-scale brewery wastewater treatment systems. This study is separated into two different sections, a laboratory jar study to determine optimum conditions for treating high strength brewery wastewater using anaerobic digestion, and a field research study to evaluate the actual operating conditions that influence treatment for an on-site MBBR treatment system in addition to solving actual operational problems. The specific objectives of this research are as follows:

- Evaluate the effects of temperature, and organic loading rate on the anaerobic digestion of high strength brewery sludge in terms of methane production, maximum methane production rate and COD removal using dairy manure digested inoculum.
- Characterize the wastewater production at Beau's brewery in terms of flow and organic loading and recommend system design improvements in terms of flow balancing.
- Characterize and evaluate the full-scale treatment of brewery wastewater with an MBBR system in terms of organic loading, DO and temperature.
- Evaluate the effect of effluent recycling on filamentous bulking and treatment efficiency.

1.3 Thesis Organization

Chapter 1 presents background information about the brewery industry, the characteristics of the brewery wastewater, and the challenges that the brewery industry is facing. Furthermore, chapter 1 presents the significance of the research and the objectives of the study. The chapter 2 presents a literature review of the brewery industry, the brewing process, the brewery wastewater characteristics, some challenges that brewery's face, the introduction of basic brewery wastewater treatment methods, and advanced treatment technologies for treating brewery wastewater.

Chapter 3 presents the result of the batch study of using anaerobic condition to digest high strength brewery wastewater using a dairy manure innoculum. The batch test was conducted based on the central composite design and used two different operation condition (temperature and brewery wastewater concentration)—the optimized condition obtained from biochemical methane potential.

Chapter 4 presents the result from the field study for a craft brewery located at Vankleek Hill, Ontario, Canada, for optimizing the operation condition and increasing the organic removal rate by monitoring the wastewater compositions and flow rate. Meanwhile, a new equalization tanks volume and a recycle strategy suggested for controlling the sludge bulking and increase the treatment efficiency is proposed.

Finally, Chapter 5 presents all conclusions resulting from this study and recommendations for future research in the area as well as for the full-scale operation.

1.4 Contribution of Authors

Based on this research study the following two manuscripts described below with the contributed authors.

Article 1 (Chapter 3):

A. Wusiman and C. Kinsley

Optimizing operational parameters for anaerobic co-digestion of brewery wastewater with dairy manure.

A. Wusiman: Conducted literature review, developed and conducted experimental procedure, analyzed results and wrote manuscript.

C. Kinsley: Provided supervision in the development of experimental procedure, analysis of results and reviewed manuscript.

Article 2 (Chapter 4):

A. Wusiman and C. Kinsley

Optimization of Operational Conditions for Brewery Wastewater Treatment System Operated as Moving Bed biofilm Reactor (MBBR).

A. Wusiman: Conducted literature review, developed and conducted experimental procedure, analyzed results and wrote manuscript.

C. Kinsley: Provided supervision in the development of experimental procedure, analysis of results and reviewed manuscript.

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CHAPTER 2

LITERATURE REVIEW

2.1 Brewery industry

In terms of popularity, beer is the fifth most consumed beverage and is number one in terms of alcoholic beverages (OCDE, 2005). The domestic beer industry has seen a decrease in production of 5.74% between 2013 and 2018 but also increasing revenues (Couillard, 2019). This is credited to the growing craft brew industry that is representing a larger portion of beer consumption and is generally more expensive. At the start of the decade, there were 318 breweries operating in Canada and by 2018 this number had increased to 995. The increase is primary due to the start-up of many new craft breweries. In 2018, 93% of the breweries in Canada were small-scale, consisting of craft brew, microbrewery, nanobrewery and brewpubs ("Beer Canada," 2019). Ontario has the largest number of breweries in Canada (330 of total 995 in 2018) ("Beer Canada," 2019).

2.2 Beau`s All Natural Brewing Company

Beau` s All Natural Brewing Company was established in 2006 in Vankleek hill, Ontario, Canada. Beau`s Brewing use 100% organic ingredient and local spring water to brew their beer. During the nearly 15 years of operation, Beau`s improved their brewing process to become an environmentally friendly and sustainable brewery. With a collaboration with Bullfrog Power, a Canadian green power company, Beau's was able to eliminate their carbon footprint and becoming the first Canadian brewery to operate on 100% sustainable energy (Couillard, 2019). Since 2014, this collaboration has reduced Beau's CO_2 emissions by 110 tonnes per year. In 2016, to reduce their environmental impact even further, they constructed an on-site wastewater treatment system. This system treats their brewery wastewater before discharging to the municipal sewer system. Additionally, the spent grains are diverted for use as animal feed and high strength wastes including spent yeast and waste beer are diverted from the wastewater stream and sent to a local farm which operates an anerobic digester producing renewable electricity (Beau`s All Natural Brewing Company, n.d.).

2.3 The beer brewing processes

Beer is produced by the fermentation process of raw material including hops, grains, water and yeast (Briggs et al., 2000; Simate et al., 2011). Additionally, some breweries use additional ingredients such as sugar and flavouring ingredients to enhance the flavor of the beer (Parawira et al., 2005). Beer production will always include the three-fundamental processes of brewing, packaging and cleaning (Simate et al., 2011). The brewing and cleaning processes are responsible for the production of significant amounts of waste and wastewater (Van der Merwe & Friend, 2002). The main brewing processes used by all breweries are chemical and biochemical operations, and multiple solid-liquid separations (Ambrosi et al., 2014; Fillaudeau et al., 2006; Olajire, 2012), as show in Figure 2-1. The chemical and biochemical operations include mashing, boiling, fermentation and maturation. The solid-liquid separations include wort separation, wort clarification, yeast removal, and beer clarification (Ambrosi et al., 2014; Fillaudeau et al., 2006; Olajire, 2012).

A challenge for breweries is the disposal of by-products produced during the separation processes (Enitan et al., 2015). These can be classified into solids (e.g. spent grains), high strength wastewater and slurries (e.g. spent yeast, first rinse from fermenters, waste beer) and low strength wastewater (e.g. Clean-in-place (CIP) processes, bottle washing). Beau`s sells their organic spent grains to a local farm (Beau`s All Natural Brewing Company, n.d.;) for increase profit from waste which can be an ideal feed for livestock (McManus, 2011). The high strength wastewaters are hauled to a local farm’s anaerobic digester for co-digestion with dairy manure and electricity production. The major source of wastewater during beer production are the CIP processes. Large amounts of wastewater are produced during the cleaning process which contain caustic soda, phosphoric acid, nitric acid and many other chemicals. The cleaning process is the main source and cause of variability in the effluent flowrates and chemical composition (Fillaudeau et al., 2006; Olajire, 2012; Van der Merwe & Friend, 2002). Although most breweries use the same brewing processes, the quantity of water used and wastewater produced can vary depending on the brewery scale, type of beer, cleaning, packaging, sanitizing process, the age and type of the equipment, and operating systems (Fillaudeau et al., 2006).

Based on operational condition, the water utilization efficiency can vary widely between breweries. Modern breweries are operating at an efficiency of 5 liters of water per liter of beer produced

(Brewers Association, 2011; Van der Merwe & Friend, 2002). A portion of the water used will be captured in the final product, a small portion lost to evaporation during the brewing operation, but the majority will be disposed as wastewater (Driessen & Vereijken, 2003; Olajire, 2012; Simate et al., 2011). The discharge from breweries usually varies significantly due to the combination of effluent from different stream. For instance, the bottle washing process can contribute high volumes of wastewater with low organic content. In contrast, fermentation and filtering processes discharge small amounts of wastewater with high organic content (Fillaudeau et al., 2006; Simate et al., 2011).

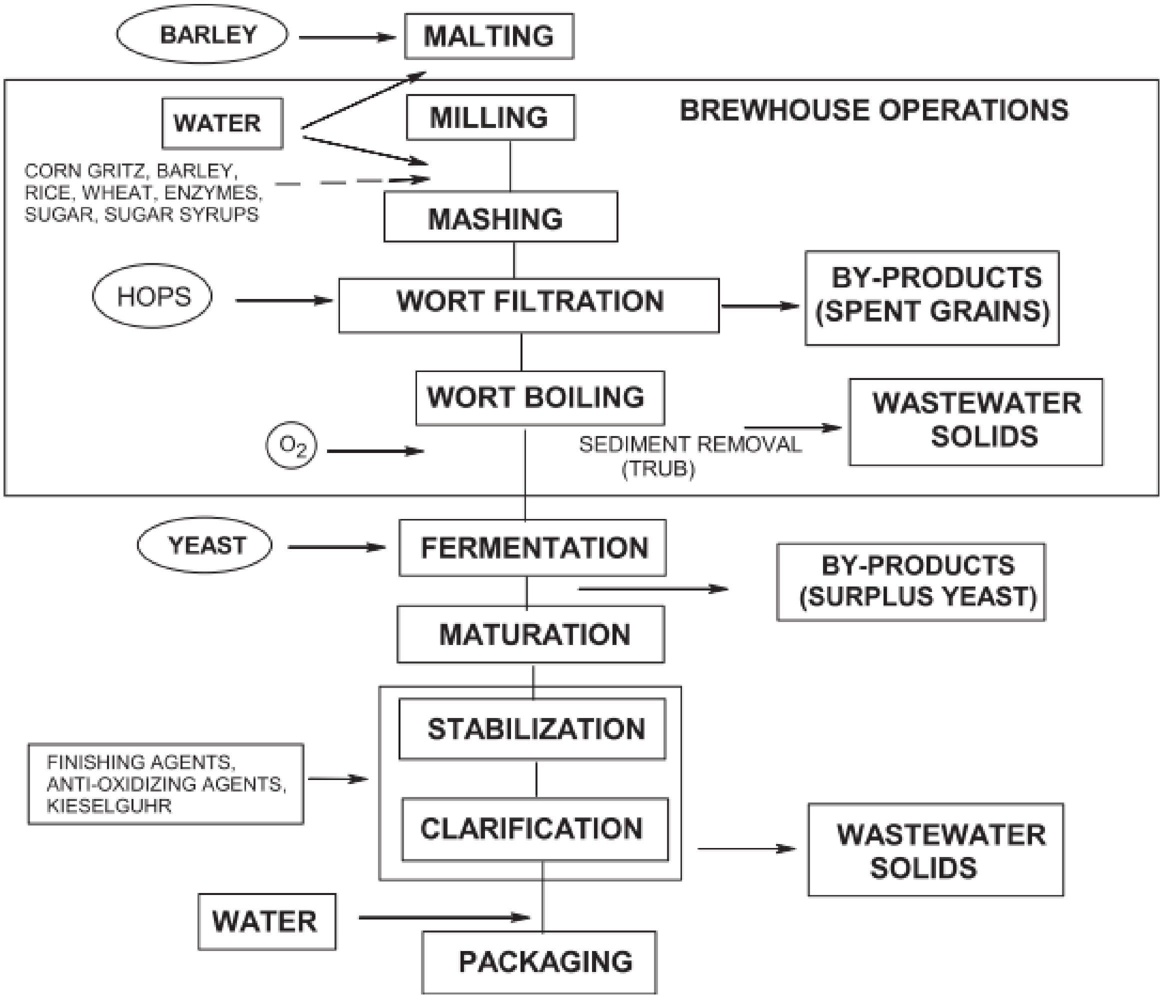

Figure 2- 1. Stages for beer production (Olajire, 2012)

2.4 Brewery wastewater characterization

The wastewater effluent from breweries are generally highly variable in both organic and hydraulic loading. Most of the organics in the effluent are easily biodegradable including sugars, soluble starches, ethanol and volatile fatty acids (Brito et al., 2007). This results in a biochemical oxygen demand (BOD) to chemical oxygen demand (COD) ratio of approximately 0.6 to 0.7 (Brito et al., 2007; Fillaudeau et al., 2006; Rao et al., 2007). The solids in the wastewater consist of spent grains, kieselguhr (diatomaceous earth filter media), waste yeast and trub (organic sludge), and are generally characterised as total suspended solids (TSS)(Brito et al., 2007; Brewers Association, 2011). The nitrogen and phosphorous level in the wastewater are a function of the amount and type of raw material in the wastewater such as spent yeast, and the cleaning products used for CIP processes (Brito et al., 2007). The pH of the brewery wastewater depends on the type of cleaning product and ratio of water to cleaning product used in the process, the typical range of pH for brewery wastewater is between 4.5 to 12 (Brito et al., 2007). Temperature for brewery wastewater can vary depending on the brewing process. Wastewater temperatures can reach as high as 80°C for certain procedures, but the typical range is around 18°C to 35°C (António G. Brito et al., 2007; Saila & Hasan, 2017; Simate et al., 2011). The wastewater characteristics of a typical brewery and two microbreweries in Ontario is summarized in Table 2-1.

As shown, there are large fluctuations in brewery effluent composition, especially for the microbreweries. Compared to the typical range, microbreweries have a broader range and greater extremes of wastewater composition. This increases the challenges for breweries to treat their wastewater. Breweries are challenged to meet consistent effluent objectives with highly variable wastewater strength and flow and many are faced with increasing municipal sewer discharge fees. To achieve higher efficiency and lower cost, the brewery wastewater should be stabilized. Table 2-1 demonstrates the highly variable wastewater composition obtained during our project with Beau`s brewery.

Table 2- 1

Characteristics of typical brewery wastewater with two small-scale breweries

PARAMETERS	**BREWERY WASTEWATER COMPOSITION**		
	Typical (Brito et al., 2007; Driessen & Vereijken, 2003)	Wellington Brewery (Swain, 2019)	This Study Beau`s Brewery
COD (mg/L)	2,000-6,000	3,700-30,000	130-11,040
BOD (mg/L)	1,200-3,600	n/a	1,554-6,210
TSS (mg/L)	200-1,000	10-28,367	60-11,260
T (°C)	18-20	n/a	15.9-30.9[a]
pH	4.5-12	4.99-7.62	3-14
TN(mg/L)	25-80	78.3-80.9	10-100
TP (mg/L)	10-50	30-2,335	50-1,330

Note:

n/a is referred as not applicable

[a] Temperature of wastewater in the first aerobic reactor

2.5 Brewery water and wastewater management

Due to increasing popularity, the number of microbreweries has increased in recent years and many of them are located in smaller communities (Beer Canada, 2019). Brewery wastewater contains high strength organics as well as significant concentrations of both nitrogen and phosphorous, requiring treatment prior to discharge to either a municipal sewer or to a receiving water body (Brewers Assiociation, 2013). High levels of organic matter in the wastewater can reduce dissolved oxygen (DO) in the receiving waters causing hypoxic conditions which can result in fish kills, excess nutrients can increase eutrophication and the extreme pH of wastewater can be detrimental to aquatic life. Many small municipalities with older wastewater treatment plants are not equipped to treat the extra organic and/or hydraulic loads from a new brewery in the community (Brewers

Association, 2013; Olajire, 2012; Simate et al., 2011). Governmental environmental regulations prohibit the discharge of untreated wastewaters and municipal Over-strength Discharge Fees (ODFs) provide a financial incentive for breweries to pre-treat their effluent to the municipal sewer use standard.

Table 2-2 presents the European (EU Council Directive 1991) and Canadian limit for discharging directly to waterbodies and the discharge limit for discharging to the municipal sewer system in the City of Ottawa and Town of Vankleek Hill, ON. Different discharge limits apply in different jurisdictions and limits for direct discharge to water bodies are evidently more stringent compared to discharge to municipal sewer systems (Driessen & Vereijken, 2003).

Table 2- 2

Indicative discharge standard for different region

	Discharge Limits for Waterbody		Discharge Limits for Sewer		Ottawa ODFs change by the time	
Parameters	EU (Driessen & Vereijken, 2003)	Canada (MWWE, 2005)	Ottawa (By-law No. 2003-514)	Vankleek Hill – Township of Champlain (By-law No. 2007-28)	2012 (Paulo, 2014)	2019
BOD_5 (mg/L)	25	20	300	300	1.44 ($/kg)	1.66 ($/kg)
TSS (mg/L)	35	25	300	350	0.77 ($/kg)	0.88 ($/kg)
TAN (mg/L)	10-15	1-3	100	50	5.75 ($/kg)	6.6 ($/kg)
TP (mg/L)	1-2	1.0	10	10	2.31 ($/kg)	2.66 ($/kg)

Current ODFs can add a significant operating cost to breweries and it is likely that ODF's will increase over time as wastewater treatment plant discharge limits become more stringent. It is thus advantageous for the breweries to reduce the strength of their discharge to meet municipal discharge limits. To decrease the strength and variability of their discharge, breweries require robust wastewater treatment systems.

2.6 Brewery wastewater treatment processes

In order to either meet environmental discharge criteria or avoid ODF fees, breweries should operate an on-site wastewater treatment system (WWTS). There are three basic types of unit processes (physical, chemical, and biological) for treating wastewater. To achieve their desired contaminant removal, a brewery WWTS combines several unit processes to achieve the desired level of treatment for wastewater constituents of concern: organic matter, suspended solids, N, P, and pH.

The WWTS for the brewery will depend on the individual plant operation. However, every WWTS is a combination of basic types of unit processes (Metcalf & Eddy, 2014). For brewery WWTS, physical treatment primarily removes solids material in the wastewaters. Typical physical treatment found in brewery WWTS includes screens that remove coarser solids such as the spent grains and sedimentation which is used to remove some of the remaining suspended solids by means of gravity. Another important physical treatment is the flow equalization of the influent flow. Flow equalization evens out the flow in the treatment trains allowing constant flow through the system. The most frequently used chemical treatment processes for brewery WWTS are pH adjustment and coagulation and flocculation. Brewery wastewater pH can be variable, but it is necessary to keep wastewater pH between 6 and 9 to protect the microorganisms which carry out the biological treatment. (Metcalf & Eddy, 2014; Simate et al., 2011; Werkneh, Beyene, & Osunkunle, 2019).

After brewery wastewater undergoes physical and chemical treatment, it is essential to treat by biological treatment to reduce the quantity of dissolved organic matter in the wastewater. Biological treatment for wastewater can be aerobic (with air/ oxygen supply) or anaerobic (without oxygen). As shown in Figure 2-2, aerobic treatment uses aerobic microorganisms (predominantly bacteria) that utilize organic matter and oxygen in the water to produce biomass while producing inorganic end products such as carbon dioxide (CO_2), ammonia (NH_3) and water (H_2O). During the aerobic process, a significant amount of heat is released. A sedimentation basin typically follows an aerobic biological treatment cell to remove excess cell biomass produced. Anaerobic treatment is an alternative biological treatment process that produces biogas and significantly less biomass without the use of oxygen as an electron accepter. The process produces less heat but more energy dense biogas. Biogas produced by anaerobic treatment usually contains 55-75 vol%

methane (CH_4), 25% to 40 vol% CO_2 and traces amounts of hydrogen sulfide (H_2S) (Simate et al., 2011).

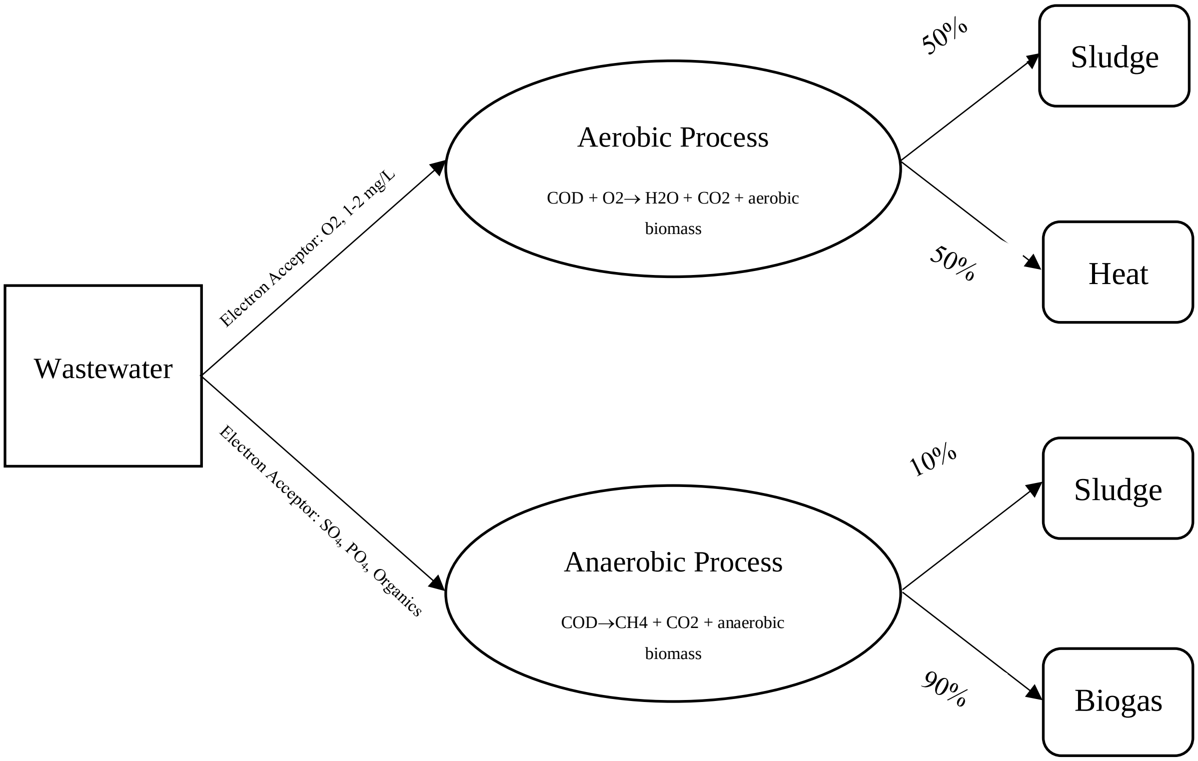

Figure 2- 2. Aerobic and anerobic process (Driessen & Vereijken, 2003; Simate et al., 2011)

Table 2-3 compares aerobic and anaerobic treatment. Aerobic treatment is commonly used for wastewater containing soluble chemical oxygen demand (sCOD) lower than 2000 mg/L, while anaerobic treatment is used for wastewater with sCOD greater than 2000 mg/L(Metcalf & Eddy, 2014). Biogas produced from anaerobic treatment can be used to generate heat or electricity, meanwhile aerobic treatment requires extra energy for the aeration system. Another advantage for anaerobic treatment is that it produces considerably less sludge which can reduce sludge management costs. However, anaerobic treatment requires a longer hydraulic retention time (HRT) than aerobic treatment. While breweries can choose aerobic or anaerobic treatment to treat their wastewater, a combination of the two systems also is a favored design (Driessen & Vereijken, 2003; Metcalf & Eddy, 2014; Simate et al., 2011; Werkneh et al., 2019).

Furthermore, for both environmental and economic reason, new brewery WWTS should possess higher removal efficiency and resource (water and energy) recovery potentials (Simate et al., 2011). For pre-existing breweries, lowering the water usage ratio and maximize the removal efficiency of their current operations is necessary. Additionally, upgrading or building energy recovery systems would benefit the breweries. For breweries operating in water-scare countries, reducing the water usage ratio through process optimization or water reuse will reduce water input as well as wastewater treatment costs (Götz et al., 2014).

Table 2- 3

Comparison of aerobic and anaerobic treatment (C.P. Leslie et al, 1999; Metcalf & Eddy, 2014; Yeoh, 1995)

Feature	Aerobic	Anaerobic
Organic removal efficiency	High	High
Effluent quality	Excellent	Moderate to poor
Organic loading rate	Moderate	High
Sludge production	High	Low
Nutrient requirement	High	Low
Alkalinity requirement	Low	High for certain industrial waste
Energy requirement	High	Low to moderate
Temperature sensitivity	Low	High
Start-up time	2-4 weeks	2-4 months
Odor	Less opportunity for odors	Potential odor problems
Bioenergy and nutrient recovery	No	Yes
Mode of treatment	Total (depending on feedstock characteristics)	Essentially pre-treatment

2.6.1 Biomethane- Anaerobic Treatment

Brewery wastewater is characterized as medium to high strength wastewater (Parawira et al., 2005). Use of aerobic treatment for this level of organic wastewater requires a significant amount of electricity for aeration. By comparison, anaerobic treatment requires considerably less electricity for operation and can produce methane for energy recovery (Chong et al., 2012; Metcalf & Eddy, 2014). Therefore, in the brewery industry, as with the food and beverage sector in general, anaerobic digestion is the most widely used wastewater treatment system (Metcalf & Eddy, 2014).

Anaerobic treatment has a long history that can track back to the 18th century and even earlier (Abbasi et al., 2011). Conventional anaerobic reactors, such as the floating drum digester, fixed dome digester, and inflatable tubular digester, are very simple to build and operate (Bond & Templeton, 2011; Haandel & Lettinga, 1994; Nzila et al., 2012; Rajendran et al., 2012). To increase organic removal efficiency and biomethane yield, the design and operations of the anaerobic reactors have improved over time (Neshat et al., 2017). Currently, there are advanced high rate reactors including the upflow anaerobic sludge bed (UASB), expanded granule sludge bed (EGSB), fluidized bed reactor, and more (Neshat et al., 2017). Anaerobic systems have been further modified to incorporate in vessel filters such as the anaerobic upflow blanket filter (AUBF), AnMBR and combination of EGSB and anaerobic filter (EGSB-AF) (Connaughton et al., 2006; Han et al., 2015; Ince et al., 2000; Neshat et al., 2017; Yu & Gu, 1996).

The key operating parameters which must be controlled for successful anaerobic digestion are: pH, temperature, carbon to nitrogen (C/N) ratio, OLR, HRT, alkalinity, and concentration of volatile fatty acids (VFA) (Metcalf & Eddy, 2014; Neshat et al., 2017; Rajendran et al., 2012). The optimum pH range for CH_4 production is from 6.5-8.0 and it should be recognised that the production of VFAs can drop the pH below the favorable level (Kato, Rebac, & Lettinga, 1999). Therefore, alkalinity can be added as a buffer for maintaining an acceptable level of pH to keep microorganisms active (Han et al., 2015; Ince et al., 2000; Yu & Gu, 1996; Zoutberg & Rob, 1996). The optimum temperature for anaerobic digestion is 25 °C to 45 °C (Saila & Hasan, 2017). The presence of high ammonia can inhibit the activity of methanogens (Farrow, 2016). For operating more effectively, higher OLR with shorter HRT is preferred (Connaughton et al., 2006).

Although the brewery wastewater contains high strength organics, there is no toxic substrate (Driessen & Vereijken, 2003; Olajire, 2012; Simate et al., 2011). Additionally, the temperature of the brewery effluent usually is higher than 20°C (Driessen & Vereijken, 2003; Olajire, 2012; Simate et al., 2011). Thus, anaerobic treatment can be operating more effectively with less concern and electricity. Especially in the tropical and subtropical areas, the anaerobic reactor needs less or even no additional heat to maintain the temperature (Haandel & Lettinga, 1994). Table 2-40 summarizes research studies treating brewery wastewater with different types of anaerobic high rate reactors. The table includes operating parameters, effluent results and biomethane production rates. The maximum or optimized result applied in the table and parameters used in the research is also summarized. The summarized studies maintained system pH at around 7.

Table 2- 4

Anaerobic high rate reactor that treated actual brewery wastewater

Reference	Reactor type	Reactor size L	Inoculum/ Seed	Temp °C	OLR kg COD/m^{3}·d	HRT hour	COD removal %	Biogas yield m^{3} CH4/kg COD removed
(Yu & Gu, 1996)	AUBF	11.32	Digested sludge from MWWTP	20-29	1.42-3.21	4	above 90	0.29-3.33
(Han et al., 2015)	AnMBR	15	n/a	35	10	n/a	98	0.53±0.015(biogas)[a]
(Ince et al., 2000)	AnMBR	120	Digested sludge from MWWTP	36±1	28.5	60-100.8	97	0.28
(Zupančič et al., 2007)[b]	ASBR[c]	30	n/a	55	3.23	13.5-26	88.9	n/a
(Shao et al., 2008)	ASBR	45	Full-scale UASB reactor	33 ±1	1.5-5	24	above 90	0.48
(Rao et al., 2007)	UASB	25	Anaerobic lagoon	37±2	23.1	2	96	0.32
(Cronin & Lo, 1998)	UASB	16	Aerobic activated sludge	19-24	0.25	18	91	n/a
(Parawira et al., 2005)	UASB	500000	Aerobic activated sludge	25-35	6	24	57	n/a
(Zoutberg & Rob, 1996)	EGSB	220000	n/a	25-30	17	1.8	above 98%	n/a
(Kato et al., 1999)	EGSB	225.5	Fine GS[d] from full scale UASB reactor	20	7.5-12.6	2.1-1.2	80-86	n/a

(Connaughton et al., 2006)[e]	EGSB-AF	3.38	From mesophilic anaerobic sludge reactor	15	1.62-4.02	48-18	92.6-88.7	0.15-0.28
	EGSB-AF	3.38	from mesophilic anaerobic sludge reactor	37	1.62-4.02	48-18	87.1-85.5	0.28-0.27

a. In this study, methane occupied 59% of total biogas
b. For substrate used brewery slurry
c. Anaerobic sequencing batch reactor (ASBR)
d. Fine Granule sludge (GS): granules with diameter below 0.8 mm
e. In this study, the research strategy is to increase OLR and to lower HRT

As mentioned previously, breweries discharge warm, non-toxic, high strength wastewater and easily degradable wastewaters (Driessen & Vereijken, 2003; Olajire, 2012; Simate et al., 2011). Those properties make it an ideal substrate for anaerobic treatment. However, brewery wastewater has a high C/N ratio and low alkalinity which can inhibit single substrate digestion. On the other hand, animal manure has a low C/N ratio, which affects digestion performance (Tufaner & Avşar, 2016). Animal manure also generally possesses a high alkalinity and can work as an ideal buffer solution for the system (Cheng & Zhong, 2014). Therefore, co-digested brewery wastewater with animal manure is a favorable solution for compensate the disadvantages of both substances.

Anaerobic co-digestion is a process that aims to obtain optimized biogas production by digesting various substrates together (Alatriste-Mondragón et al., 2006). Co-digestion can optimize the AD process by compensating the various substrates' deficiencies and balancing the system's parameters, such as the C/N ratio, nutrient composition, and alkalinity. Co-digestion can increase biogas yield and system stability and create opportunities to digest specific substrates that could not be efficiently digested as a single substrate (Misi & Forster, 2001). Co-digesting brewery wastewater with animal manure can balance the C/N ratio in the system and increase the buffering capacity for neutralizing the VFAs. Furthermore, co-digestion can dilute potential toxic contents, increase the bacteria diversity, and utilize the nutrients (Ebner et al., 2016; Hashimoto, 1983; Hills &

Roberts, 1981). However, there is not much research reported about the co-digestion of brewery wastewater with animal manure. Olatunde (2016) co-digested brewery wastewater and cow dung at a different ratio. The research conduct at a batch size (1.5L) anaerobic digestion reactor and temperature maintained at 35±0.5 °C. The maximum methane production rate (0.92 L/d) and methane yield (0.287 m3 CH4/kg VS added) obtained at cow dung to brewery wastewater ratio 70:30.

2.6.2 Future energy Recovery Systems

At the end of 2019, world energy production was still dependent on fossil fuels (Ritchie & Max, 2020). Fossil fuels have negative impacts on the environment from mining and combustion: the mining process of fossil fuels can damage the surrounding area and emissions from burning fossil fuel is the leading cause of global warming (Barbir et al., 1990). Because of scarcity of this natural resource and greenhouse gas emissions there is a necessity to find a substitute for fossil fuels. The brewery industry produces high strength organic waste as a by-product of beer production. Therefore, biogas production from brewery wastewater can effectively treat the wastewater while producing green energy (Brunner & Rechberger, 2015). Energy recovery from the brewery wastewater is not limited to the biomethane, and can also produce biohydrogen or direct electricity, which even cleaner than the biomethane. In this section, various methods that can produce energy from brewery wastewater when simultaneously treat wastewater are discussed.

2.6.2.1 Biohydrogen Production

Hydrogen (H_2) has been widely used in different applications. The most significant use of hydrogen is energy resource (Marchetti et al., 1995; Ramachandran & Menon, 1998). Hydrogen is characterized as a non-polluting clean energy source and possesses high conversion efficiency and able to be recycled (Suzuki, 1982). Consequently, generating hydrogen efficiently at low cost is becoming a broad interest in the energy sector. There are different methods for generating hydrogen, such as thermochemical, electrochemical, biological, etc. Biological treatment is getting attention because it is environmentally friendly and requires less energy than other options.

Biohydrogen can be produce by a multiple a different process. The four most commonly used method for biohydrogen production are (Das & Veziroğlu, 2001):

a) Produce by algae and cyanobacteria biophotolysis the water
b) Organic compounds photocomposed by the photosynthetic bacteria
c) Organic matter fermentation
d) Hybrid systems, which is a combination of photosynthetic and fermentative bacteria

Brewery wastewater can be an ideal recourse for the photosynthetic and/or fermentative bacteria to produce biohydrogen because the brewery wastewater contains high organic matter and is not toxic to the microorganisms (Shi et al., 2010; Vijayaraghavan et al., 2007). The photosynthetic biohydrogen production method have high theoretical conversion yields and can operate at a wide spectrum of the light (Das & Veziroğlu, 2001). For this advantage photosynthetic biohydrogen production is getting more attention. Fermentative bacteria have a high reproductive rate, high growth rate, and are capable of producing hydrogen constantly even in dark condition. Unfortunately, fermentative bacteria are not getting enough attention yet(Das & Veziroğlu, 2001).

There is some research about optimizing operational conditions when using brewery wastewater to produce biohydrogen. Vijayaragharan et al. (2007) studied the influence of HRT on biohydrogen production. In this study, the inoculum bacteria culture was separated from the cow dung and applied into brewery wastewater at the anaerobic condition, which contains 2470 mg/L COD. HRTs of 1-day and 7-days were studied and obtain 69% and 95.5% COD removal, 6.7 L and 9.7 L biohydrogen production, and hydrogen concentrations representing 60% and 62% of total biogas produced, respectively. Seifert et al. (2010) obtained biogas containing 90% hydrogen when using a waste to media percentage of 10% by volume. The wastewater was filtered and then sterilised at 120°C for 20 minutes before being added to the media. The inoculum in this study contained *Rhodobacter sphaeroides O.U. 001* (concentration of inoculum 0.36 g dry wt/L) and an applied illumination intensity of 116 W/m^2. This study also demonstrates that sterilization can affect the H_2 production yield (no sterilization 0.76 L H_2/ L medium, after sterilization 2.2 L H_2/L medium). Shi et al. (2010) obtained optimized pH, temperature, and brewery wastewater concentration as 35.9 °C, 5.95, and 6.05 mg/L, respectively. At this condition, H_2 yield and maximum hydrogen production rate is 149.6 mL H_2/g COD and 53.6 mL/h.

2.6.2.2 Microbial fuel cells (MFC)

The MFC is a new alluring technology, since it is capable of producing electricity directly from organic matter in the wastewater and simultaneously treating the wastewater (Feng et al., 2008). Figure 2-3 shows the basic mechanism of the MFC technology (Timmis, 1995). The MFC systems function in a three-step process. The first step, on the anode side, the microorganisms will produce electrons, protons and CO_2 from the organic matter (wastewater or added nutrients). The second step, the electrons flow through the flow meter to the cathode and produce electricity. At the last step, on the cathode side, the microorganisms will convert oxygen to water in the aerobic condition, or under the anaerobic condition will convert nitrate to nitrite or N_2 and convert CO_2 to acetate (Timmis, 1995).

The significant advantage of MCFs is it capability of generating energy directly from wastewater, while anaerobic treatment requires an intermediate gas phase for electricity conversion (Kaewkannetra et al., 2011). Plus, the MCF can reduce CO_2 emissions (Werkneh et al., 2019). The brewery wastewater is an ideal source for the MCFs since brewery wastewater contains high concentrations of soluble COD and less ammonia-nitrogen (Wang, Feng, & Lee, 2008). Unfortunately, research for the MFCs to treat brewery industry discharge water are still at lab-scale, there are no actual full-scale WWTS operated as MFC in the brewery industry.

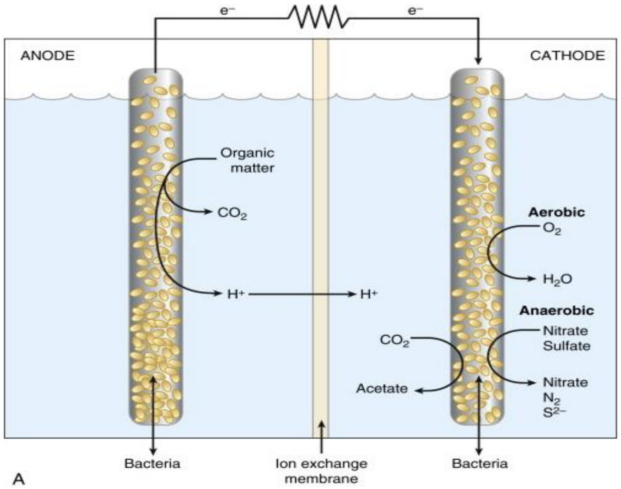

Figure 2- 3.Schematic figure for the cathodic and anodic chambers of a MFC (Timmis, 1995)

2.6.3 Moving bed biofilm reactor (MBBR)

The MBBR process is a growing technology developed in 1989 in Norway (Metcalf & Eddy, 2014). In the MBBR tanks, microorganisms will attach to the plastic media and grow as a biofilm (Ødegaard, 1999). The biofilm attachment can be encouraged by the agitation in the reactor, aeration in the aerobic systems, and mechanical mixing in the anaerobic and anoxic systems (Figure 2-5) (Metcalf & Eddy, 2014). The carriers are usually made from polyethylene with a density designed to be close to 1 g/cm^3 (Ødegaard et al., 2000), which allows the carries to move freely in the reactor. Consequently, the MBBR tank can be filled up to 70% by volume with the carriers (Metcalf & Eddy, 2014; Ødegaard, 1999; Ødegaard et al., 2000). Because the whole reactor is completely mixed the active reactor volume is almost the same as actual volume; which is an advantage compared to the other biofilm technologies. The carriers can provide a large surface area for biofilm growth and react with substances in the water (Figure 2-4) (Ødegaard et al., 2000). Consequently, MBBR technology is capable of accomplishing various treatment purposes for both municipal and industrial wastewater, such as BOD/COD removal, nitrification, and denitrification (Metcalf & Eddy, 2014; Ødegaard, 1999; Ødegaard et al., 2000).

In an MBBR system, various treatment levels can be achieved by combining different basic MBBR units in series (Metcalf & Eddy, 2014). Figure 2-5 shows the basic MBBR system schematics for aerobic and anaerobic/anoxic conditions. MBBR systems tend to produce substantially less sludge that suspended growth systems as the biofilm remains mostly attached to the carriers, producing TSS concentrations ranging from 10-800 mg/L (Bassin et al., 2016; Johnson et al., 2000; Metcalf & Eddy, 2014; Ødegaard, 1999; Rusten et al., 1992).

MBBR systems compared to the traditional WWTS, can provide a larger surface area for reaction, lower hydraulic retention time (HRT), continuous operation, no sludge recycles, and low sludge production (Aygun et al., 2008; Ibrahim et al., 2016; Ibrahim et al., 2012; Metcalf & Eddy, 2014; Ødegaard, 1999; Ødegaard et al., 2000). Due to the parameters mentioned above, the MBBR systems require a relatively small footprint (Ødegaard, 1999; Ødegaard et al., 2000), which can be advantageous in a brewery. The MBBR systems are easy to operate but are difficult to maintain the reactor tank due to the carriers. A significant advantage of MBBR systems compared to other biofilm treatment technologies is not requiring backwash (McQuarrie & Boltz, 2011). Additionally, MBBR systems are capable of nitrification and denitrification. Unfortunately, most

MBBR systems are unable to remove phosphorous efficiently. The most common solution for phosphorus removal is chemical coagulation (Metcalf & Eddy, 2014).

The MBBR system is widely used for domestic wastewater treatment applications, however, there has been limited application to brewery wastewater. Manyuchi and Chikwama (2016) used Mutag biochip (carrier surface area 3000 m2/m3) for treating brewery wastewater aerobically, which contains COD of 673 mg/L and obtained 87.9%, 93.4% and 48% removal for COD, BOD, and TSS respectively, at 12 hours HRT. Biase et al. (2016) conducted a brewery wastewater lab-scale study using anaerobic MBBR for optimizing operational parameters, such as carrier filling percentage and surface organic loading rate (SALR). For filling percentage, the research operated under 35°C at 25% and 35% carrier filling percentage (volume) and obtain 80% removal of COD. The study also achieved a CH_4 production yield of 0.36±0.06 m^3 CH_4/kg sCOD at 25 % filling and 0.39±0.03 m^3 CH_4/kg sCOD at 35% filling. For optimization of SALR, this study operated parallel reactors with different HRT ranging from 6-24 h. The best result was found at 18h HRT, which obtained 88±8.5% sCOD removal with CH_4 yield at 0.34±0.06 m^3 CH_4/kg sCOD. At this condition, the calculated SALR is equal to 13±1 g sCOD/m^2·d. Boyle (2019) researched optimizing operational parameters for treating brewery wastewater via aerobic MBBR. Two different types of carriers and suspended growth were used for determining optimized SALR (10-55 g sCOD/m^2·d) at 12 h HRT. As a result, no significant difference in the result were observed between the carriers and suspended growth system for all three tests, except at the highest SALR (55 g sCOD/m^2·d), where the carries presented lower suspended solids (MLSS) concentration compared to the suspended growth system. In this study, the effect of the HRT was also determined. As well, at fixed SALR (40 g sCOD/m^2·d), when HRT decreased from 12 h to 3 h. there is a significant shift from suspended growth to attached growth, and organic removal rate for the carriers was maintained at 95% at 4 h HRT and decreased to 73% at 3 h HRT. For the suspended growth system, the organic removal rate dropped to 88% at 4 h HRT and 61% at 3 h HRT. The MBBR technology is widely operated in the food and beverage industry (Boyle, 2019), even though we have found limited references to MBBR systems applied to the brewery industry. Due to these advantages, the MBBR system has the potential to become an effective treatment option for low mid-strength brewery wastewaters.

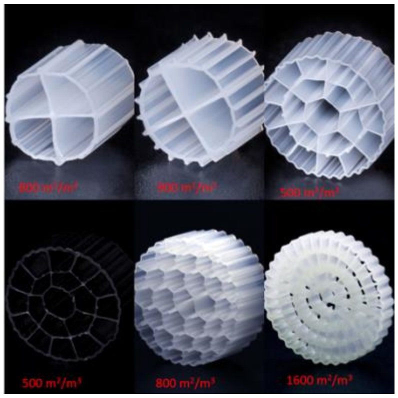

Figure 2- 4. Different types of carrier used for MBBR system

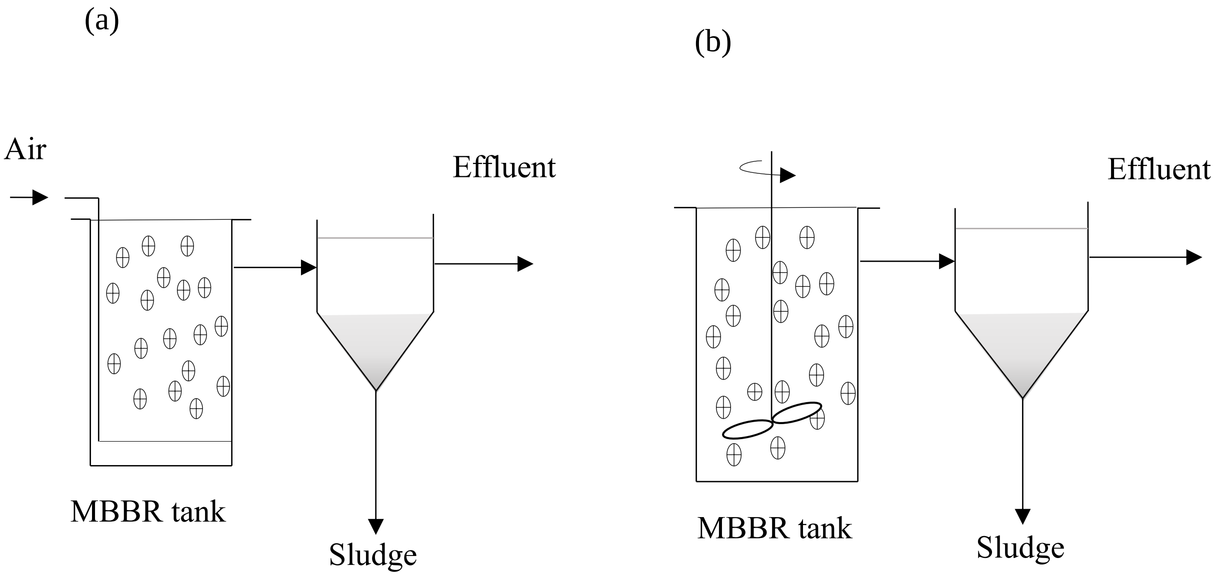

Figure 2- 5. MBBR systems basic schematic, (a) aerobic and (b) anaerobic/anoxic (Metcalf & Eddy, 2014)

2.6.4 *Membrane reactors and reuse*

As mentioned previously, the brewing industry utilises a large quantity of water (Ambrosi et al., 2014). Only a small amount of the total water used is economically beneficial to the brewery (as beer), the remaining becomes wastewater which needs to be treated (Fillaudeau et al., 2006; Han et al., 2015). Generally, the wastewater is discharged into designated systems after being biologically treated. However, there are different potential options to further treat the water to produce useful regenerated water. It must be noted that regenerated water may not achieve drinking water quality (Braeken, Van Der Bruggen, & Vandecasteele, 2004), but recycling wastewater can bring benefits. In this section, advanced biological WWTS and various other methods that are used to treat the brewery wastewaters to reusable quality is discussed.

2.6.4.1 Membrane filtrations (MF)

Membrane filtration technology uses a membrane as a barrier to allow selected constituents to pass through the membrane while the remaining constituents remains in the retentate (Metcalf & Eddy, 2014). MF operations generally use porous membranes for separation, depending on the membrane pore size membranes can be divided into microfiltration, ultrafiltration, nanofiltration and hyperfiltration (Usually known as reverse osmosis) (Metcalf & Eddy, 2014; Simate et al., 2011). Table 2-5 summarizes the essential characteristics for these processes, such as membrane pore size, operational range, and removal ability.

As Figure 2-6 shows, MF can be distinguished as two different types base on their operational conditions: dead-end filtration and crossflow filtration (Ambrosi et al., 2014; Meshksar et al., 2020; Metcalf & Eddy, 2014). In a dead -end system, all the feed water must flow through the membrane to produce permeate while the constituents are retained by the membrane. When the mixed liquor has low concentration or accumulation do not hinder the flow significantly, dead-end filtration will operate at high efficiency. In crossflow operation, the main flow direction is tangential to the membrane surface producing shear forces maintaining a cleaner membrane surface. The water that does not permeate through the membrane returns to the feed tank.

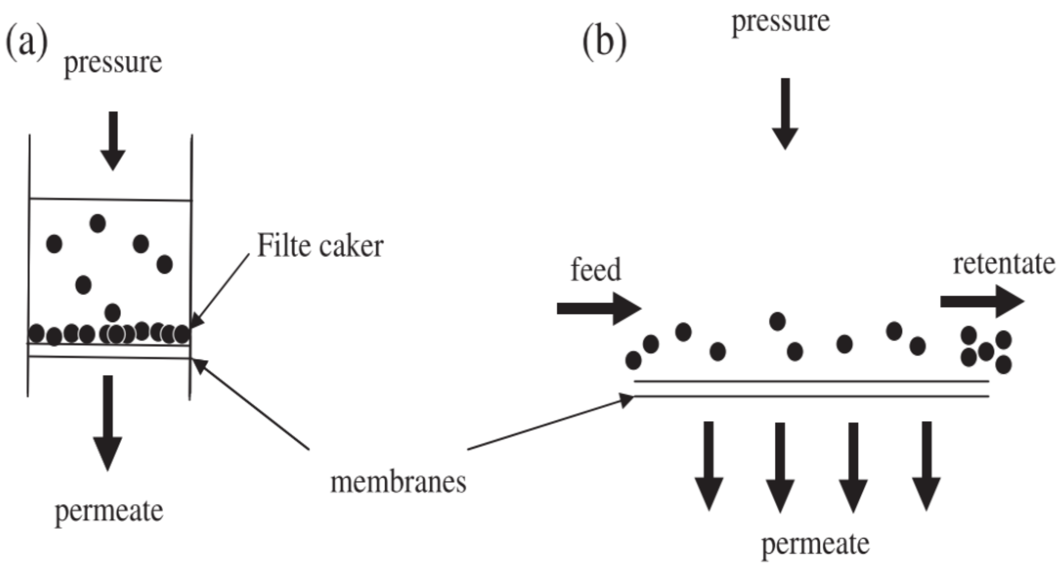

Figure 2- 6. Two types of MF operational mode, (a) dead-end and (b) crossflow (Simate et al., 2011)

MF technology has been successfully used in both drinking water and wastewater applications (Gupta & Suhas, 2009). Compared to traditional filtration technology, MF is more flexible and easier to scale up. Furthermore, the permeate quality of MF is not influenced by temperature. However, membrane fouling affects significantly the performance of MF. Another factor to consider is the quality of the membrane as higher quality products will have a longer operating life. In addition, excessively high flowrates can be destructive when operating some sensitive membranes. High pressure can increase the risk of explosion. Furthermore, the cost for high-quality membranes is one of disadvantage for MF technology (Fillaudeau et al., 2006; Götz et al., 2014; Metcalf & Eddy, 2014; Simate et al., 2011; Werkneh et al., 2019).

In the brewery industry, using membrane filtration for treating wastewater is practiced. Sawadogo (2018) achieved 99% COD removal by nanofiltration, while Braeken et al. (2004) achieved 100% removal, also by nanofiltration. The most significant advantage of membrane filtration technology is it capability of regenerating the wastewater. The highest quality water is obtained when operating nanofiltration or reverse osmosis systems (Simate et al., 2011; Werkneh et al., 2019). There are already full-scale, mature on-site brewery WWTS operating MF systems, especially in water-scare countries; brewery water internal recirculation can bring significant benefits (Götz et al., 2014). Furthermore, membrane filtration, especially nanofiltration and reverse osmose, can be used as the last step combined with other treatment systems to effectively achieve water recovery.

Table 2- 5

Typical characterization of membrane process (Gregory, 2005; Metcalf & Eddy, 2014)

Membrane process	**Membrane dividing force**	**Separation mechanism**	**Typical pore size (nm)**	**Typical operating Range (µm)**	**Typical operating pressure (bar)**	**Smallest items removed**
Microfiltration	Hydrostatic pressure difference or vacuum in open vessels	Sieve	Macropores (>50 nm)	0.07-2.0	<4	Colloids, bacteria
Ultrafiltration	Hydrostatic pressure difference or vacuum in open vessels	Sieve	Mesopores (2-50 nm)	0.008-0.2	2-10	Viruses, large organic molecules
Nanofiltration	Hydrostatic pressure difference or vacuum in closed vessels	Sieve + solution / diffusion + exclusion	Micropores (<2 nm)	0.0009-0.001	5-40	Divalent ions, small organic molecules
Reverse Osmosis (Hyperfiltration)	Hydrostatic pressure difference or vacuum in closed vessels	Solution / diffusion + exclusion	Dense (<2 nm)	0.0001-0.002	15-150	All dissolved martial

2.6.4.2 Membrane bioreactor (MBR)

MBR technologies are a growing component in wastewater treatment (Islam et al., 2008; Simate et al., 2011; Visvanathan & Pokhrel, 2003). It combines biological treatment (aerobic or anaerobic) with membrane filtration technology to achieve liquid-solid separation (Metcalf & Eddy, 2014; Werkneh et al., 2019). Based on the location of the MBR membrane module in the system, they can be classified as side-stream MBR and submerged MBR (Metcalf & Eddy, 2014). Figure 2-7 shows the process configuration for these two types of MBRs. Generally, submerged MBR is more favourable since it does not need extra energy to pump mixed liquor to the membrane chamber (Jeison, 2007). The MBR compared to conventional activated sludge (AS) reactor can achieve lower TSS values even when treating higher concentrations of mixed liquor in a relatively small operating area (Holbrook et al., 2005). Additionally, the MBR operation does not have to be concern about filamentous bacteria bulking problem (Metcalf & Eddy, 2014). However, the major challenge affecting reactor performance is membrane fouling as similar to MF (Simate et al., 2011). Energy cost to produce the vacuum and membrane replacement are extra operational costs compare to AS.

MBR technology has been applied in the brewery industry and received good results. Zhang et al. (2004) used a submerge MBR to treat 0.27g/(g·d) brewery wastewater and achieved above 90% removal for both COD and NH_4. Sawadogo et al. (2018) reported 95% removal when treating 0.7-10.6 g COD/L brewery wastewater with an MBR. Han et al. (2015) used an anaerobic membrane bioreactor (AnMBR) technology for wastewater containing 3.5-11.5 g COD/(L·d) at 35°C to remove above 98% COD and produced biogas at a rate of 0.53 ± 0.015 m^3/kg COD reduced. By combining up-flow anaerobic sludge bed (UASB) with an MBR, Dai and Yang (Dai et al., 2010) removed 96% of COD. MBR technology is effective, especially combined with anaerobic treatment, which can accomplish both water and energy recovery.

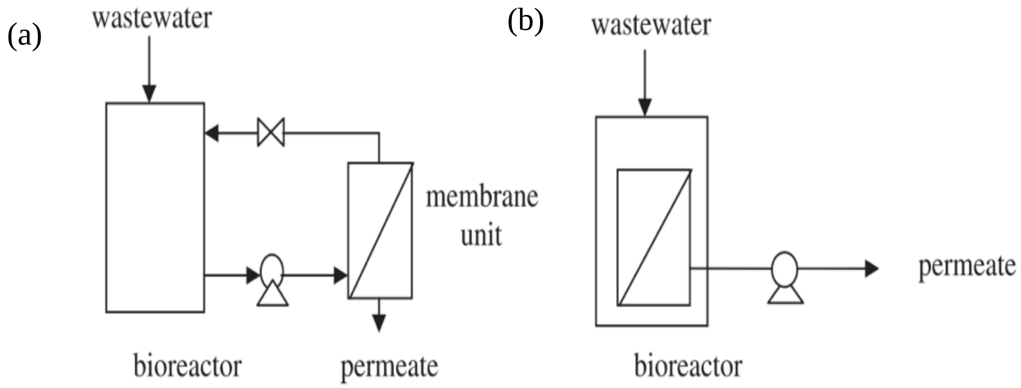

Figure 2- 7. Process configurations for MBR, (a) side-stream MBR and (b) submerged MBR (Simate et al., 2011)

2.6.5 Aerobic +Anaerobic Combined System

As mentioned previously, biological treatment can be divided into aerobic and anaerobic technologies. In general, aerobic treatment is suitable for low strength wastewater (sCOD <2000 mg/L) and anaerobic treatment is suitable for high strength wastewater (sCOD >2000 mg/L) (Metcalf & Eddy, 2014). As described in Table 2-3, the significant advantages of anaerobic treatment are low energy consumption and the potential for both energy and nutrients recovery, while the primary advantages of aerobic treatment are higher removal efficiency and ease of operation (C.P. Leslie et al., 1999). Therefore, a pre-anaerobic treatment with post aerobic treatment can obtain better performance by integrating advantages from both technologies (Driessen & Vereijken, 2003; N.P, 2005)(N.P, 2005). A/O systems can achieve high removal efficiencies with low energy consumption, low sludge production, short HRT, potential energy recovery, and small footprints. Therefore, A/O systems can provide both economical and environmental benefits to the processor (Chan et al., 2009; Gašpariková et al., 2005; Swain, 2019; Willie et al., 2000).

A/O systems according to the operating reactor type can be divide into three different categories: conventional anaerobic-aerobic system, anaerobic-aerobic system using high rate bioreactors, and integrated anaerobic- aerobic reactor (Figure 2-9). The conventional anaerobic-aerobic system is the most straightforward operation of A/O system, usually using a conventional system such as a lagoon, artificial or natural wetlands and aerated stabilizing tank. In the system, the large ponds connected as series, anaerobic treatment usually operates on the bottom of the ponds while aerobic processes occur in the upper layer. Conventional anaerobic- aerobic treatment are easy to operate, but they have long HRTs, large space requirements and cannot treat high organic loading rates (OLRs). The space constraints of these technologies remove them from consideration for breweries in urban areas. To overcome these constraints, A/O systems have been developed which use high rate bioreactors, such as UASB, FBR, MBBR, filter bioreactor, and various membrane reactors (Gašpariková et al., 2005). These systems achieved both resource recovery and high removal efficiencies, which can meet the discharge limits. As different from the conventional A/O system, using high rate reactors can operate across a wide range of wastewater strengths. However, the key to obtaining high removal efficiency and high resource recovery at low HRT is to choose the right combination sequence of the high rate reactors. The right high rate reactors in the A/O system, cannot just reduce HRT and operating footprint, but can also achieve both energy and water recovery. In recent years, the compact high-rate reactor is starting to obtain more attention, which integrates aerobic and anaerobic treatment into one reactor. This provides advantages including a reduced footprint, odour control and lower sludge production. There are four different types of Integrated anaerobic- aerobic bioreactors system shown in Figure 2-8. Integrated systems are more beneficial for economic reason, but the operation and design are still at the beginning stage (Chan et al., 2009).

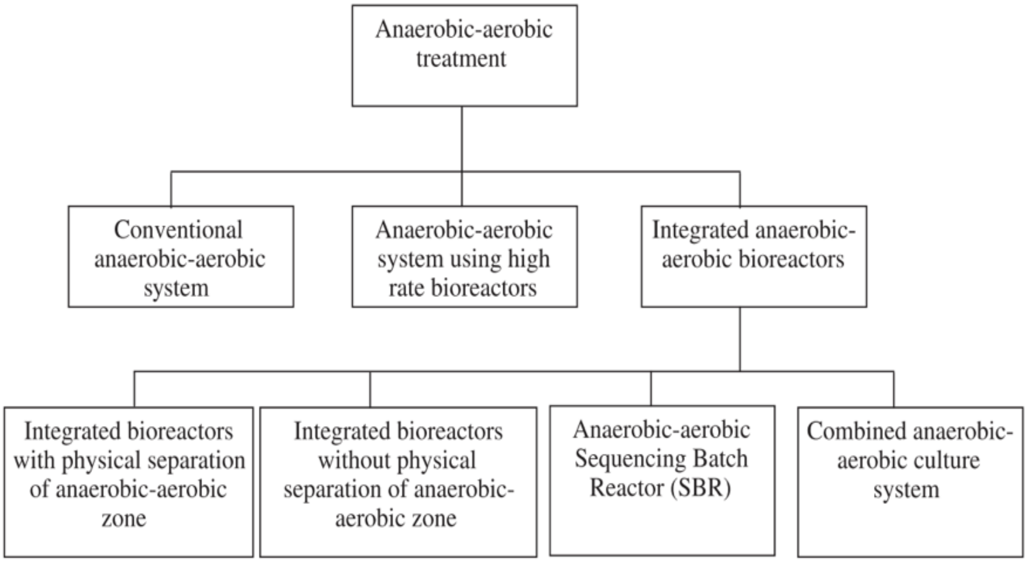

Figure 2- 8. Types of combined anaerobic-aerobic treatment (Chan et al., 2009)

As mentioned before, most of the newly installed breweries are located in urban areas with access to municipal sewers. Therefore, breweries are attracted by the advantages of A/O treatment system. Kothiyal. M et al. (2018) applied a combination of anaerobic hybrid reactor and activated sludge to treat local brewery wastewater in India. This system achieved an overall 75% to 97% COD removal at OLR between 0.9 kg COD /m$^{3\cdot}$ d to 3.6 kg COD /m^3. Joshi and Kumar (2019) used a combination of UASB and aerobic digestor to treat brewery wastewater in Nepal. The wastewater COD range varied widely from 165 to 3950 mg COD/L, with average 52.5% COD removal with the UASB and an average 78.5% COD removal with the aerobic treatment. As well, the study achieved a CH_4 production rate between 0.14-0.16 m^3/kg COD removed. Willie. D et al. (2000) removed 80% COD and 94 % sCOD from an A/O system, which combined an internal circulation reactor (two UASB reactors on top of each other) and an airlift reactor. This full-scale WWTS was designed for OLR at 10.5 COD kg/d and achieved an average biogas production at 0.47 m^3/kg COD removed. A/O treatment systems are not limited to just two reactors and can combine reactors in the series. Swain (2019) in a lab study combined two anaerobic reactors and two aerobic reactors to treat microbrewery wastewater. At 1.03 d HRT and OLR range from 6.5 to 25 kg COD/m$^{3\cdot}$d, the maximum COD removal rate the system achieved is 93%. At the same OLR, prolong HRT to 2 d, the system maximum removal rate is increased to 98%.

A/O systems also can add anoxic treatment to accomplish advanced nutrients removal. This treatment system calls A^2/O (anaerobic/ anoxic/oxic). Also, the A/O system can combine other treatment systems to achieve higher resource recovery. Thien et al. (2019) used A^2/O combined with an MBR system for advanced nitrogen and phosphorous removal. The best result achieved at OLR as 0.75 kg COD /$m^{3\cdot}$ d. Under this OLR removal efficiencies of 94.5% COD, 99.2% NH_4^+-N, 86.7% TN, and 83.6% TP were achieved. Liu (2017) combined MFC with A/O, which operates as the AnMBR/MBR system. The COD removal of the system was maintained at 73.87%, but HRT dropped from 42.4h to 22.6h. As presented above, the studies are showing that A/O systems demonstrate advantages in combining different treatment units for advanced resource recovery and nutrient removal with a small footprint.

2.7 Conclusion

As the small-scale brewery industry expanded, a large number of small-scale breweries were established during the past few years with the trend expected to continue in the future (Couillard, 2019). The brewery industry has a large water demand with a high water to beer ratio (Simate et al., 2011; Werkneh et al., 2019). Most of the water used in the brewery ends up disposed of as wastewater (Fillaudeau et al., 2006; Han et al., 2015). Brewery wastewater is considered medium-to-high strength and easily biodegradable wastewater (BOD to COD ratio is higher than 0.6) and highly variable on both organic and hydraulic loading (Fillaudeau et al., 2006; Parawira et al., 2005; Rao et al., 2007). Additionally, there is no toxic content reported in the brewery wastewater. Therefore, brewery wastewater is very suitable for biological treatment.

Considering the economic and environmental perspective, the attention for treating brewery wastewater that is not limited to removing organic matter should also consider resource recovery from the wastewater. There are various interesting technologies presented above for water and energy recovery purposes, where an appropriate combination of an A/O system can accomplish both objectives at the same time, bringing significant economic and ecological benefits.

This research evaluates and optimizes the performance of an existing aerobic MBBR treatment system for a small-scale brewery and investigates the potential for energy recovery and carbon removal with anaerobic pre-treatment of segregated high-strength brewery waste streams.

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CHAPTER 3

OPTIMIZING OPERATIONAL PARAMETERS FOR ANAEROBIC CO-DIGESTION OF BREWERY WASTEWATER WITH DAIRY MANURE

3.1 Abstract

As the microbrewery industry expands, disposal of brewery wastewater is becoming more of a concern, both for brewery operators and for local municipal wastewater authorities. Anaerobic digestion (AD) processes have obtained wide interest due their ability to achieve energy recovery and organic removal simultaneously. A two-factor Box-Wilson central composite design (CCD) was adopted to find optimum biomethane production conditions for the digestion of brewery wastewater with a dairy manure inoculum. The effects of two major influencing factors of temperature (25-49°C) and brewery wastewater concentration (BWC) (2-9 g VS/L) on Biochemical methane potential (BMP, which is CH_4 yield) and CH_4 maximum production rate R_{max} were evaluated by applying response surface methodology (RSM). All of the trials presented a high organic removal efficiency with volatile solid (VS) 82-91%, soluble chemical oxygen demand (sCOD)77-88%, and total chemical oxygen demand (tCOD) between 47% -76%. The organic removal efficiency results suggested that operating temperature does not play a significant role in degradability. However, the first order rate constant and biogas content (methane percentage in the biogas) was shown to be strongly affected by temperature. The mesophilic regime had the highest average rate constant, while the psychrophilic regime rate constant was significantly lower than the mesophilic and thermophilic regimes. The conditions in the thermophilic range present a high variability for the first order rate constant. At the end of the AD, the psychrophilic, mesophilic, and thermophilic regime had an average methane percentage 47, 65, and 67% respectively. An optimum organic load of 5 gVS/L was observed with both lower and higher concentrations found to inhibit the AD process with an inoculum to substrate (I/S) ratio of 1. Optimum BMP and R_{max} were achieved under conditions of 49 °C and BWC of 5g VS/L. Correspondingly, the BMP and R_{max} were 141.40 mL CH_4/g VS added and 36.5 mL CH_4/ day, respectively. However, the preferable operational condition T=35°C & BWC=5 g/L is

recommended for system stability. At this condition methane yield is 110.07 CH_4/g VS added & maximum methane daily production is 28.06 CH_4/ day, which is similar to the maximum result.

3.2 Introduction

During the past decade the demand for craft beer has dramatically increased with the number of the small-scale breweries in Canada tripling from 2012 to the end of 2019 ("Beer Canada," 2019).

Brewery wastewater is characterized as medium to high strength organic wastewater with high variability in terms of both organic and hydraulic loading; particularly for small-scale breweries. This highly variable wastewater provides challenges to treatment (Parawira et al., 2005; Brewers Assiociation, 2013). AD is an established and proven mature technology widely utilised to reduce organic matter in food and beverage wastewaters while providing energy recovery (Chong, Sen, Kayaalp, & Ang, 2012; Metcalf & Eddy, 2014). However, it's applicability to small breweries is impacted by potentially relatively high capital and operating costs for relatively low energy recoveries.

AD is the microbial (bacteria/anarchea) decomposition of organic matter without oxygen. During decomposition, microorganisms will produce biogas with considerably lower biomass and heat compared to aerobic treatment. Typical percentage by volume for biogas compositions is 55 -75% of CH_4, 25- 40% CO_2, N_2 is around 17%, and a small traces amounts of H_2S (Driessen & Vereijken, 2003). The biogas is produced by microorganisms through four primary reaction stages: hydrolysis, acidogenesis, acetogenesis, and methanogenesis (Yilmazel & Demirer, 2011; C. Zhang et al., 2014). Although, fundamentals for AD are the same, biogas composition varies with the utilized substrate and operating conditions.

AD presents some strong advantages for treating wastewater. Compared to aerobic treatment, AD produces less biomass. Furthermore, to treat the same concentration of organics, AD requires less energy without the need for aeration. Additionally, a significant advantage of AD is that the process can produce biogas for generating heat or electricity. Biogas can be purified to bio-methane (contains 98% CH_4) with a similar composition to fossil-derived natural gas and can replace natural gas. This advantage provides a promising avenue for diminishing dependence on the fossil fuel and reduce emission of Greenhouse gases (C.P. Leslie et al, 1999; Metcalf & Eddy, 2014; Yeoh, 1995).

Biogas produced from AD is a highly complicated process. Microorganisms and reactor parameters determine the quantity and quality of the biogas produced. To achieve the maximum AD potential depends on the operating parameters and interactions between the parameters (Neshat et al., 2017). Therefore, precise control of the parameters can be decisive to obtain an optimized result from the operation. The critical parameters that can influence the performance of AD are considered to be: temperature, hydraulic retention time (HRT), pH, I/S ratio, organic loading rate (OLR), alkalinity, carbon : nitrogen: phosphorous ratio, and ammonia concentration (Neshat et al., 2017).

Temperature and Organic loading rate are two important operating parameters for system design. System pH is also critical for AD, however, the optimum pH range for microorganisms involved in AD is well understood and should be maintained between 6.8 to 7.2 (Hagos et al., 2017; LIU et al., 2008; Rajeshwari et al, 2000). Lower than pH 6.8 can inhibit the growth of microorganisms, and high pH has a negative effect on methane production. To maintain a sustainable operation of the reactor, sufficient alkalinity should be available to maintain system pH close to 7. Most studies choose to add 5 g $CaCO_3$/L buffer solution to protect the system from the acidification. As well, an optimum C:N:P ratio for microbial growth is 100:5:1 (Neshat et al., 2017).

The AD process is highly susceptible to temperature, where a small fluctuation in temperature may lead to large declines in biogas production, as temperature affects the metabolic activity of the microorganisms. Therefore, it is important to operate at a consistent temperature to maintain a stable condition for microorganisms. Additionally, temperature also can affect the gas transfer rate and settling characteristics (Neshat et al., 2017). Temperature ranges for operating AD systems can be classified into psychrophilic (14°C -25°C), mesophilic (35°C -45°C) or thermophilic (45°C -60°C) (Metcalf & Eddy, 2014). AD systems operated at the psychrophilic range are stable and require less energy for maintaining reactor temperature. However, in this temperature range, metabolic activity is low, and will take a longer time to obtain the same results compared to the higher temperature ranges. Generally AD performs best under thermophilic conditions as high-temperatures increase solubility of organics and reaction rates, and lowers the solubility of the gas in the liquid and liquid viscosity (Neshat et al., 2017). Increased reaction rates should result in reduced HRT. However, higher temperatures can also boost the inhibitory effect of the ammonia by increasing the pKa and will require more energy for heating the reactor (Angelidaki & Ahring, 1992, 1993). AD systems are most commonly operated within the mesophilic temperature range as the process is more stable than at thermophilic temperatures, less sensitive to NH_3 inhibition, requires less energy for heating and provides a higher biogas production rate than at psychrophilic temperatures (SANCHEZ, 2005). The potential for small breweries to operate an AD reactor at psychrophilic (room) temperatures could reduce system complexity, capital and operating cost with a trade-off in terms of increased HRT.

The organic loading rate is defined as the mass of organic substrate per volume per time loading to the system (Metcalf & Eddy, 2014). OLR is important because it can affect the microorganism's viability and metabolic activity. A high OLR can lead to preferential development of hydrolyzing and acidogenic bacteria. The highly active hydrolyzing and acidogenic bacteria can cause accumulation of VFAs; consequently, causing system acidification and inhibition of methanogens (Neshat et al., 2017). The optimum OLR is a critical design parameter as it determines HRT and reactor size.

Co-digestion with other substrates can balance the parameters of the system and accomplish a dynamic stability of the system (Alatriste-Mondragón et al., 2006; Misi & Forster, 2001). Brewery wastewater, even if it is non-toxic, contains high strength organic matter with low nitrogen and alkalinity concentration (Driessen & Vereijken, 2003; Olajire, 2012; Simate et al., 2011). On the other hand, dairy manure has a low C/N ratio (Tufaner & Avşar, 2016) and high alkalinity (Cheng & Zhong, 2014). Therefore, co-digesting brewery wastewater and dairy manure can balance the C/N ratio and alkalinity, and can provide more diverse bacteria for the treatment (Ebner et al., 2016; Hashimoto, 1983; Hills & Roberts, 1981). Furthermore, digested dairy manure or other previously digested materials can be added to the AD as inoculum to promote the digestion process. The inoculum can provide a sufficient amount of active and complex microorganisms responsible for a series of biochemical reactions (hydrolysis, acidogenesis, acetogenesis, and methanogenesis) (Boulanger et al., 2012). Therefore, the I/S ratio is critical to the AD process. There are limited research sources for the I/S ratio for AD; there is no related research found for the brewery waste. However, the studies present inhibition on both the low (lower than 0.25) and high (higher than 2) I/S ratio (Raposo et al., 2006). Owen et al.(1979) suggested that the I/S ratio of 1 should be considered as a standard ratio.

This study will evaluate and optimize the anaerobic digestion of a high-strength brewery wastewater. The objectives of this laboratory study are to evaluate the effects of temperature and organic loading rate on the anaerobic digestion of high strength brewery sludge in terms of methane production, maximum methane production rate, and COD removal using a dairy manure digestate inoculum. Study results can help small breweries compare options of implementing an onsite anaerobic pre-treatment solution versus off-site hauling as a feedstock for co-digestion in an existing anaerobic digester.

3.3 Materials and methods

3.3.1 Biochemical methane potential (BMP) test

The BMP test is a bioassay for determining organic matter ability of biogas production at a specific condition (Raposo et al., 2011; Owen et al., 1979). The BMP test can provide maximum biogas potential (Angelidaki et al., 2009a), which is important to design and operate the full-scale AD system. Furthermore, the BMP test also can investigate the parameters used for AD and provide the optimized conditions for further applications. The valid BMP test results should follow the criteria, which were implemented in this research (Angelidaki et al., 2009a; Holliger et al., 2016; Raposo et al., 2011):

1. All the conditions should at least be carried out in triplicate;
2. Need to determine the blank (inoculum) methane potential;
3. The test is terminated only when CH_4 production for three consecutive days is lower than BMP 1% (1 % of the accumulative volume of methane gas);
4. The BMP result should present as dry methane gas under standard condition (273.15 k and 101.33 kPa) after subtracting the methane production of the blank assay.

3.3.2 Statistical methods

3.3.2.1 Response surface methodology: central composite design (CCD)

The Box-Wilson CCD is a widely used second order response surface methodology, which needs a smaller number of experiments than three-level of full factorial design (Box & Wilson, 1951).

In this research, the CCD was applied for determining the surface respond of the operating variables. Furthermore, the CCD can identify optimum variable values from the model. The CCDs should apply to at least two different variables. Table 3-1 shows experimental trial information applied to a two factors Box-Wilson CCD (x_1 and x_2). The central points (0,0) are used to establish the variance of the model. The response function (Equation 3-1) represents a regression analysis from the data obtained from the research.

$$Y = \beta_0 + \beta_1 x_1 + \beta_2 x_2 + \beta_{12} x_1 x_2 + \beta_{11} x^2{}_1 + \beta_{22} x^2{}_2 \qquad (3\text{-}1)$$

Where Y is response, $x_1$and $x_2$is coded design factors, β_0 is the constant coefficient, and β_i is model parameters. The parameters of the response equation were solved using Minitab19.

Table 3- 1

Box- Wilson CCD for two factors

Trial Reference	1	2	3	4	5	6	7	8	9	10	11	12	13
x_1	-1	1	-1	1	-1.41	1.41	0	0	0	0	0	0	0
x_2	-1	-1	1	1	0	0	-1.41	1.41	0	0	0	0	0

3.3.2.2 Statistical significance

The CCD validation was accomplished by analysis of the variance (ANOVA) using Minitab 19. Triplicate test results were also used to calculate single-factor ANOVA using MS Excel to compare results between experimental conditions. Statistical significance for ANOVA test considered as $p<0.05$.

3.3.3 Experimental set-up

3.3.3.1 Waste and inoculums

The high-strength brewery wastewater used in this study was collected from Beau`s All-Natural brewery located in Vankleek Hill, Ontario, Canada. Organic waste and wastewater at the brewery are segregated into three streams: spent grains, low-strength wastewater and high-strength wastewater. Spent grains are sold as feed to a local swine farm. Low strength wastewater, which is pre-treated in an onsite aerobic MBBR system and discharged to the municipal sewer, includes most of the Clean-In-Place (CIP) washwaters from the brewing vessels as well as washwaters from the bottle washing operation. High-strength wastewater is comprised of spent yeast, waste beer and the first rinse from the washing of brewing vessels and is hauled to a local farm for anaerobic co-digestion with dairy manure. The high-strength brewery wastewater was stored at 4±1°C after collection from the Beau`s brewery. The inoculum used for the experiment is a mixture of 75% dairy manure digestate and 25% digestate from previous BMP tests, which can increase the diversity of the microbial community and reduce the start-up time of the system. Dairy manure digestate was collected from a farm-scale anaerobic digester in St. Eugene, Ontario and stored at 4±1°C. Digested sludge used for the inoculum is from a previous BMP test and collected from all three different temperature regimes (psychrophilic, mesophilic and thermophilic). The characteristics of brewery wastewater and the inoculum used for the tests are described in Table 3-2. Each trial was designed with a fixed inoculum content and variable brewery wastewater amount. Therefore, each trial has different I/S ratio as summarized on the Table 3-3.

Table 3- 2

Characteristics of Substrate and inoculum used

Parameters	***Brewery Wastewater (High-Strength)***	***Inoculum***
Total solid (TS) g/L	21.74±0.35	19.15±0.37
Volatile solid (VS) g/L	21.13±0.343	12.37±0.25
Total Chemical oxygen demand (TCOD) g/L	43.42±0.30	18.68±0.31
pH	4.83	7.84
Ammonia (NH_3-N) mg/L	121	476
Orthophosphate (PO_3^{4-}) mg/L	38	34
VS/TS	0.97	0.65

tCOD/VS	2.1	1.5
sCOD/tCOD	0.88	0.42

Table 3- 3

I/S ratio for each trial

Trial Reference	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**
Temperature (°C)	-1	1	-1	1	-1.41	1.41	0	0	0	0	0	0	0
BWC g VS/L	-1	-1	1	1	0	0	-1.41	1.41	0	0	0	0	0
Inoculum to Substrate Ratio	1.65	1.65	0.71	0.71	1.0	1.0	2.47	0.55	1.0	1.0	1.0	1.0	1.0

3.3.3.2 BMP test

Temperature and substrate concentration were chosen as the two factors and applied to a two levels Box-Wilson CCD statistical method for determining of CH_4 yield. The design parameters used in the experiment are shown in Table 3-4. The BMP test was conducted in 600 mL serum bottles, as shown in Figure 3-1 (a). The trials were all carried out in triplicate and include inoculum and water blanks. The serum bottles used in the experiment were each filled with 160 mL of inoculum, 38 to 171 mL of substrate, 45 mL of 90 g/L $NaHCO_3$ and the bottles were topped with deionized water to 400 mL, as shown in Figure 3-1 (b). The serum bottle headspace was flushed with 98% N_2 gas for 3 mins before incubation to eliminate the presence of the oxygen. Five different temperature conditions were tested in temperature control chambers with a shaker. All BMP bottles were continuously mechanically shaken at 100 rpm.

Table 3- 4

CCD parameters: coded factors and experimental design values

Coded design factors	**-1.41**	**-1**	**0**	**1**	**1.41**
Temperature °C	21	25	35	45	49

Brewery wastewater concentration (BWC) g VS/L	2.00	3.00	5.00	7.00	9.00

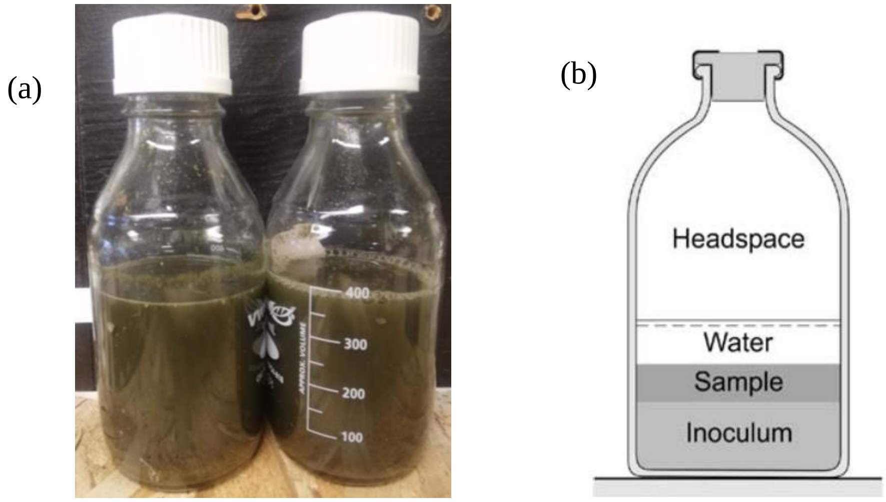

Figure 3- 1. BMP bottle pictures. (a) actual BMP bottles used in the experiment and (b) schematics of BMP bottle contents (Angelidaki et al., 2009)

3.3.4 Analytical methods

Biogas production was measured daily via a vertical U-tube water displacement manometer. The biogas content (CO_2, CH_4 and N_2) identification was measured using gas chromatography (GOW-MAC gas chromatograph Series 400, Gow-Mac Instrument Co., USA). The pH was measured with a Multi-Parameter meter with attached pH probe (VWR, Ontario, Canada). The temperatures were recorded daily and measured with an electric thermometer (ZDT1D-AUX-1, Zacro). The TS, VS, and alkalinity were measured according to the standard method 2540 B, 2540 E, and 2320 B, respectively (APHA, 2012). The soluble content of the sample was measured after filtration through 0.45 µm filter. tCOD and soluble chemical oxygen demand (sCOD) were measured according to HACH “Test N Tube^{TM} Vials” standard method 80000 (HACH, 2013), which is equivalent to Standard Methods 5220 D (APHA,2012). Ammonia and orthophosphate were measured according to HACH “Test N Tube^{TM} Vials” standard method 10031 and 8114 (HACH, 2013).

3.3.5 Numerical calculations

3.3.5.1 Volumetric gas measurements

It is important to correct biogas to the standard temperature (0°C) and pressure (1 atm) (STP) for comparative reporting, particularly when comparing different temperatures (Holliger et al., 2016). The conversion of biogas volume to the STP condition is accomplished by Equation 3-2, while the CH_4 volume is calculated with Equation 3-3.

$$V_{STP}=\frac{V_T\times 273\times(760\text{-}P_w)}{(273+\mathrm{T})\times 760} \tag{3-2}$$

Where V_{STP} is volume of gas measured at STP (L), V_T is volume of gas measure at temperature T (L), T is actual operated temperature °C, and Pw is vapor pressure of the water as a function of temperature (mm Hg).

$$V_{CH_4} = x_{CH_4} \cdot V_{Biogas} \tag{3-3}$$

Where V_{CH_4} is volume of methane production (L), x_{CH_4} is measured methane content in the gas (% by volume), and V_{Biogas} is volume of biogas measured.

3.3.5.2 BMP test value

The BMP value is presented by volume of methane gas produced by per unit of organic matters (VS, COD or biological oxygen demand (BOD)) (Equation 3-4) (Strömberg et al., 2014).

$$BMP = \frac{V_s - V_B \cdot \frac{m_{I,S}}{m_{I,B}}}{m_{S,S}} \tag{3-4}$$

Where BMP is CH_4 gas yield (L CH_4/ g VS removed), Vs is accumulated gas from the substrate (L), V_B is accumulated gas from the inoculum blank (L), $m_{I,S}$ and $m_{I,B}$ are inoculum mass (as VS) in the substrate and inoculum blank respectively (g), and $m_{S,S}$ is substrate mass (g). In this research, $\frac{m_{I,S}}{m_{I,B}}$ value is equal to 1.

3.3.5.3 Kinetic rate

In this study the first order batch kinetic model is applied. This model does not consider the lag phase, maximum biological activity, and system failures, with k values determined only over the exponential growth phase. The First order kinetic equation is presented in Equation 3-5:

$$\ln C_t = lnC_0 - kt \tag{3-5}$$

Where C is biodegradable substrate concentration mg/L, k is first order rate constant 1/d, and t is time d.

3.4 Results and discussion

3.4.1 BMP test

The BMP test operated for 74 days for all the trials with the data presented in Fig 3-2. During the experiment the temperature for each trial was recorded daily and varied within ±1.3°C. pH and alkalinity were monitored throughout the experiment with pH levels maintained above 7. At the end of the experiment, alkalinity concentrations of approximately 5-6 g $CaCO_3$/L remained in each bottle, indicating that the bottles had sufficient buffering capacity. The BMP test for each condition was run in triplicate, except for the central points, which contains 5 replicates for model verification. Some variability was observed in initial VS and COD concentrations due to the heterogenous nature of both the brewery wastewater and inoculum. Table 3-4 summarizes the BMP experimental conditions along with initial and final concentrations of VS and COD. No statistical differences were found between replicates from each trial ($p>0.05$).

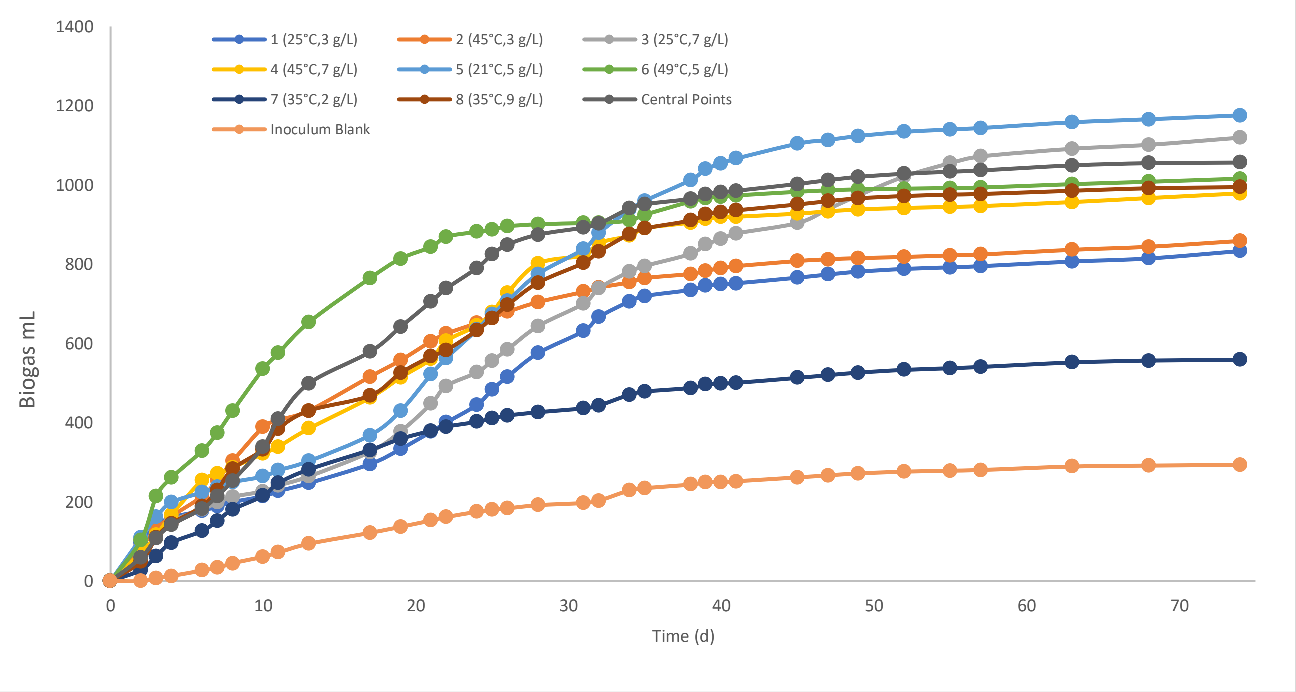

Figure 3- 2. Average cumulative biogas production for each experimental condition (n=3-5)

Table 3- 5

BMP Study Conditions, Initial and Final Concentrations and Removal Efficiencies

Design Parameters			Initial			final			Removal efficiency		
TRIAL NO.	TEMP °C* (x_1)	BWC (x_1)	TOTAL VS (G/L)	TCOD (G/L)	SCOD (G/L)	TOTAL VS (G/L)	TCOD (SD) (G/L)	SCOD (SD) (G/L)	VS %	TCOD REMOVAL %	SCOD REMOVAL %
1 (-1 , -1)	25.4 ±1.34	3.00	8.05 ±0.070	13.9 ± 0.99	8.15 ± 0.21	1.32 ±0.028	6 ± 0.21	1.45 ± 0.21	83.6 ±0.15	57.19 ±3.24	82.21 ±1.51
2 (1, -1)	45 ±1.00	3.00	7.518 ±0.052	13.4 ± 1.19	8.27 ± 0.4	1.22 ± 0.010	5.5 ± 0.61	1.93 ± 0.15	83.8 ±0.21	59.06 ±1.18	76.61 ±1.96
3 (-1,1)	25.4 ±1.34	7.00	11.893 ±0.30	20.9 ± 1.37	16.33 ± 1.02	1.54 ±0.043	6.3 ± 0.85	2.53 ± 0.35	87.1 ±0.68	70.06 ±3.08	84.49 ±3.01
4 (1,1)	45 ±1.00	7.00	11.583 ±0.038	19.7 ± 0.47	15.23 ± 1.21	1.58 ±0.015	6.4 ± 0.26	1.87 ± 0.15	86.4 ±0.11	67.46 ±2.02	87.75 ±0.88
5 (-1.41,0)	21.45 ±0.74	5.00	9.743 ±0.118	18 ± 0.46	12.07 ± 0.7	1.52 ±0.015	5.7 ± 1.07	1.97 ± 0.15	84.4 ±0.33	68.15 ±5.24	83.70 ±1.27
6 (1.41,0)	49 ±1.00	5.00	9.483 ±0.213	18.2 ± 1.15	11.97 ± 0.86	1.15 ±0.043	5.5 ± 0.25	1.53 ± 0.15	87.9 ±0.61	69.65 ±3.36	87.19 ±2.17

7 (0,-1.41)	35.54 ±1.23	2.00	6.98 ±0.40	10.6 ± 0.51	6.7 ± 0.53	1.24 ±0.063	5.6 ± 0.25	1.43 ± 0.75	82.3 ±1.72	46.69 ±2.78	78.61 ±12.71
8 (0,1.41)	35.54 ±1.23	9.00	14.08 ±0.68	26.07 ± 1.68	17.17 ± 0.7	1.25 ±0.040	6.2 ± 0.26	2.1 ± 0.4	91.1 ±0.7	76.21 ± 2.44	87.77 ±2.83
CENTAL POINTS (0,0)	35.54 ±1.23	5.00	9.77±0.28	16.52 ±1.43	12.08 ±0.63	1.17 ±0.03	5.42 ±0.63	2.16 ±0.44	88.00 ±0.57	67.11 ±3.83	82.10 ±3.59
INOCULUM BLANK	35.54 ±1.23	-	4.42 ±0.63	7.7 ± 0.36	3.4 ± 0.26	1.08 ±0.045	4.1 ± 0.86	1.67 ± 0.21	75.4 ± 6.38	43.32 ±9.04	50.98±10.22

Note:

1. Results presented average and standard deviation,1-8 trials the values obtained from the triplicate value (n=3) and for the central point n=5.
2. For temperature 45°C and 49°C the incubator maintained ±1°C.

3.4.2 Treatment of Organic matter (VS, COD, sCOD)

VS and sCOD removal efficiencies were similar and relatively high, ranging from 82-91% and 77-88%, respectively, with remaining sCOD concentrations of 1.4-2.5 g/L, which are within the normal operating range for aerobic treatment systems. tCOD removal rates were lower and varied between 47-76%. There was a significant inverse relationship between organic loading (g VS/L) and removal efficiencies across the three parameters which can be attributed to the lower biodegradability of the inoculum (see VS/TS ratio in Table 3-2); as low organic loading meant a higher inoculum/brewery waste ratio. The effect of temperature on removal efficiency was tested at the central organic loading rate of 5 g/L. A difference in VS removal efficiency was observed between 21°C (84.4±0.3%) and both 35 and 49°C (88.0±0.6 & 87.9±0.6%, respectively); however, no effect of temperature was observed on either COD or sCOD removal efficiencies. In summary, organic content from high strength brewery wastewater has a high degree of treatability (77-91%) with little to no impact of operating temperatures from 21 – 49°C.

3.4.3 Biogas production

The daily cumulative biogas production for all trials is presented in Figure 3-2. Biogas production was influenced by the organic loading, with an optimum observed close to the experimental design centre of 5 g brewery VS/L, suggesting process inhibition at both low and high organic loading (see Figure 3-3).

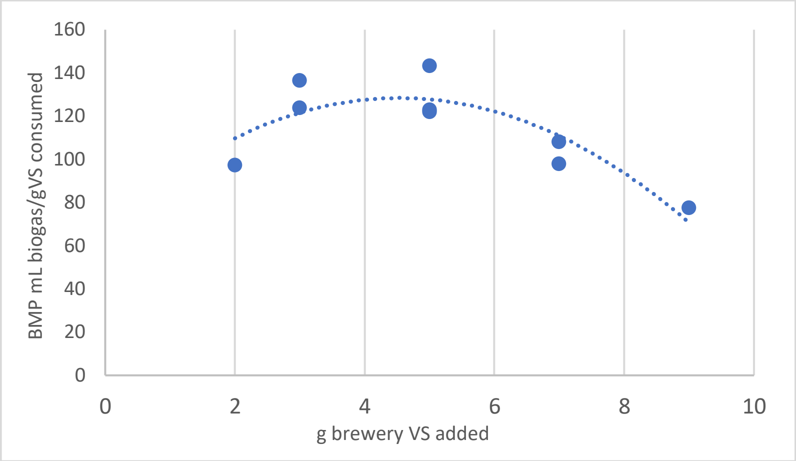

Figure 3- 3. Cumulative Biogas per g consumed vs Brewery VS added

In general, the biogas trend line has three different phases, which includes a lag phase, an exponential growth phase and a stabilization phase. The exponential growth phase was used in each trial to determine a first order batch kinetic rate constant ($R^2 > 99\%$ for each). The average kinetic rate constant per temperature grouping (psychrophilic, mesophilic, thermophilic) is presented in Figure 3-4 and demonstrates that the rate constant for the psychrophilic trials is only half that of the mesophilic trials. This suggests that a similar difference in HRT would be necessary to achieve the same treatment objective, assuming that biogas production directly relates to reduction of COD or VS. Surprisingly, the thermophilic trials do not appear to have a higher 1st order rate constant than the mesophilic trials and the high variability in the thermophilic data suggests a lack a stability in some of the thermophilic trials.

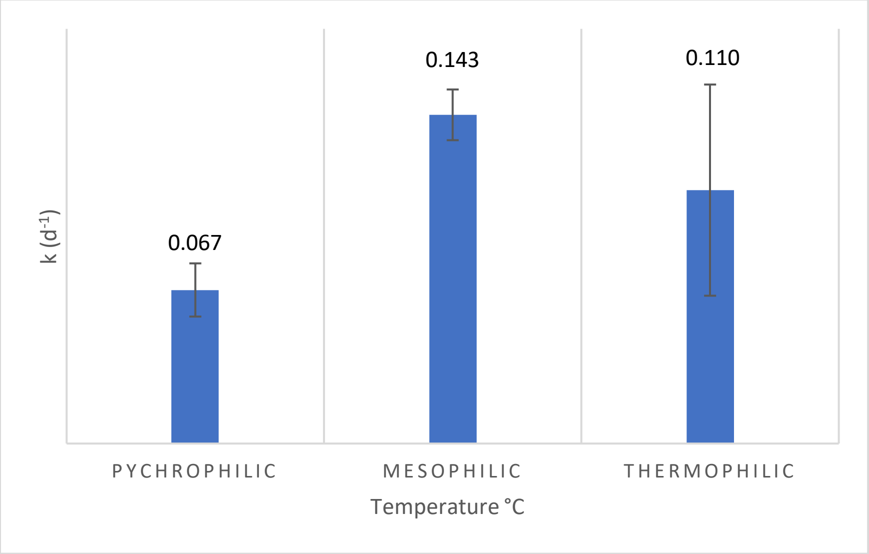

Figure 3- 4. Batch kinetic rates with temperature

3.4.4 I/S ratio

The I/S ratio used in this study ranged from 0.55 to 2.47. There is no related research for the I/S ratio for brewery wastewater. The I/S ratio for other substrate resources decreases the biogas production when the I/S ratio is lower than 0.25 and exceeds 2 (Raposo et al., 2006). As suggested, the I/S ratio for the central point is 1. Figure 3-5 presented different I/S at 35 °C; the figure shows, the high (2.47) and low (0.55) I/S ratio present lower BMP values compared to the standard (1) I/S ratio. However, the result of the I/S ratio is similar to the OLR presented in the previous section. Moreover, the I/S ratios studied in this study are limited. Therefore, it is hard to conclude that the significant difference in the BMP result resulted from the difference between OLR or the I/S ratio. To maintain a favorable performance I/S ratio of 1 is suggested. To reduce the difficulties of controlling the on-site anaerobic digestor, the BWC is analyzed as the main parameter for the rest of the study.

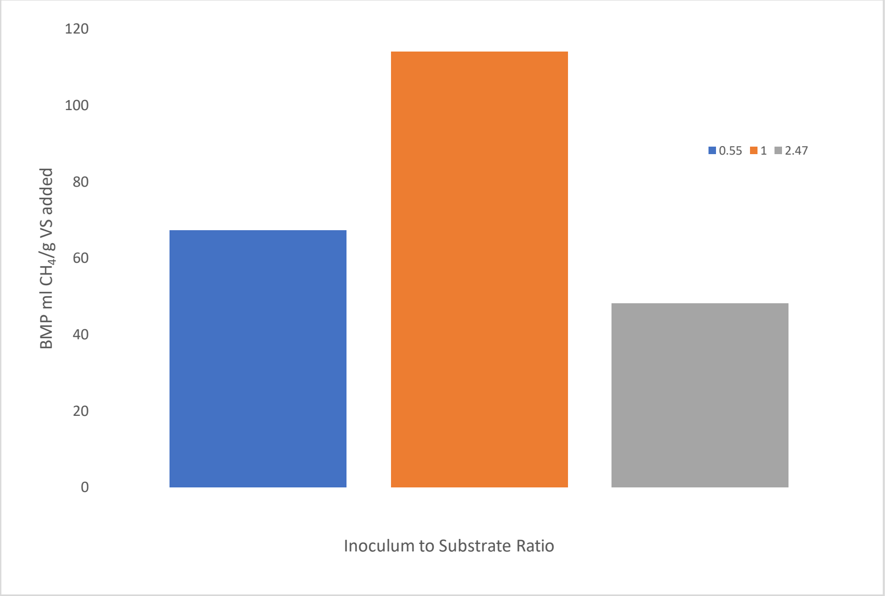

Figure3- 5. Different inoculum to substrate ratio at 35 °C

3.4.5 Biogas content

Biogas content was measured 3 times during the trials with the data described in Table 3-5. Generally, in the lag phase, the methane percentage is lower and increases to reach a steady state as the methanogens fully develop within the bottles. Increase in the methane percentage indicates that methanogenesis phase of the AD process has commenced.

Temperature played an important effect in both the development of the methanogens as well as the final methane percentage (Fig. 3-6). The psychrophilic trials exhibited very low methane content at 11 days compared with both the methanogenic and thermophilic trials. By 25 days, the psychrophilic and thermophilic trials had reached a steady state, at 41 and 65% methane, respectively, while by 74 days, the mesophilic and thermophilic trials had similar methane content of 65 and 67%, respectively, compared with only 47% for the psychrophilic trials.

As mentioned previously, the biomethane percentage produced by AD is usually from 55% to 75%, which was the case for both mesophilic and thermophilic trials, but not for the psychrophilic trials. In a similar research study, Maša et al(2015), applied BMP test for brewery spent sludge and obtained around 56% methane percentage in the biogas at 37°C.

Table 3- 6

CH_4 biogas content at different time periods

TRIAL NO.\TIME PERIOD	DAY 11	DAY 25	DAY 74
1 (25°C, 3MG/L)	5.55±7.00	45.08±2.76	37.75±5.21
2 (45°C, 3MG/L)	40.83±0.78	64.33±0.67	61.23±3.7
3 (25°C, 7MG/L))	7.66±0.8	47.27±0.71	59.8±2.28
4 (45°C, 7MG/L)	48.42±0.9	60.87±0.32	67.52±1.58
5 (21°C, 5MG/L)	1.8±1.56	39.99±0.56	41.62±3.32
6 (49°C, 5MG/L)	59.22±1.81	71.44±1.81	72.39±1.27
7 (35°C, 2MG/L)	52.33±1.86	53.09±1.86	63.61±1.8
8 (35°C, 9MG/L)	52.54±4.83	54.04±4.83	64.27±0.78
CENTRAL POINTS (35°C, 5MG/L)	36.29±2.98	54.83±2.08	65.57±1.71
INOCULUM BLANK	0±0	55.18±2.79	69.14±1.14

Note:

1. Value presented as % CH4 presented in the biogas vol %.
2. Results presented average and standard deviation,1-8 trials the values obtained from the triplicate value (n=3) and for the central point n=5.

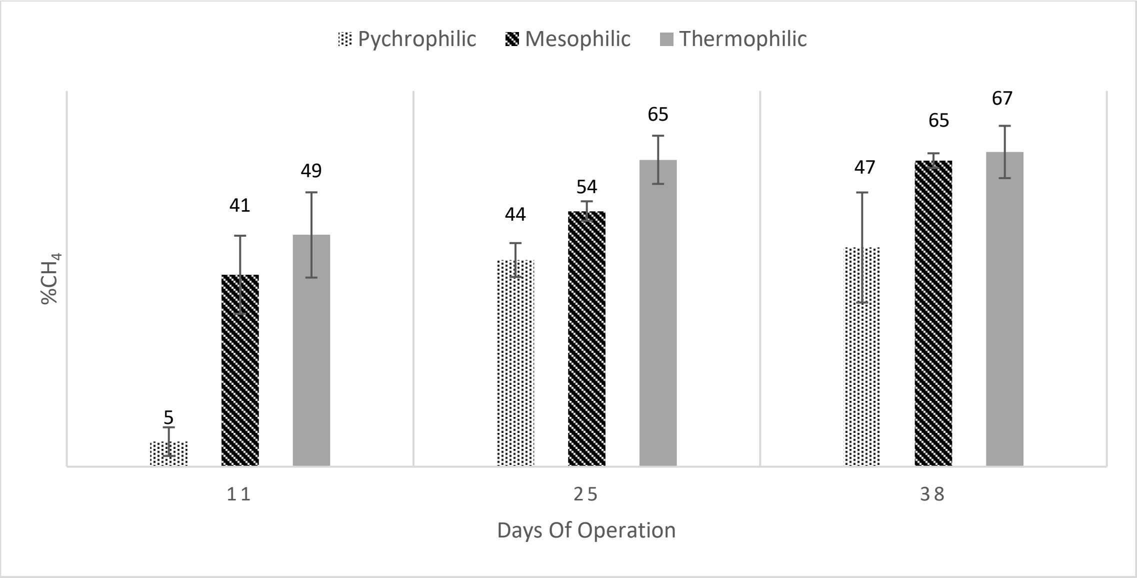

Figure3- 6. Methane content for each temperature regimes at different time period

3.4.6 CCD result

3.4.6.1 Effect of temperature and BWC on BMP

The experimental data for all thirteen trials are presented in the Table 3-6. The Regression response of the CCD in this research is described by Equation 3-6 for evaluating the influence of the temperature and BWC on BMP:

$$Y = -105.58 + 2.24x_1 + 54.70x_2 - 5.45{x_2}^2 \tag{3-6}$$

Analysis of the effect of the independent variables on the BMP is examined by the analysis of variance (ANOVA) and is presented in Table 3-7. The indicators to evaluate the adequacy of the fitted model used this research are model significance (F-value), standard error (SD), and coefficient of determination (R^2). The model significance (F-value) indicates the level of confidence that the selected model cannot be due to experimental error (Khuri & Cornell, 1987). The R^2 is the proportion of variation in the response attributed to the model rather than to random error with an R^2 value above 80% suggesting a good fit for a model (Oglaker & May, 1987).

The result of the statistical significance of the second-order model (Equation 3-6) showed that the model is highly significant as the probability is higher than F (P-value is less than 0.05). Equation 3-5 can be considered as a good fit model with an $R^2 = 0.92$. Table 3-7 can conclude that the main effect of x_1 (temperature), x_2 (BWC) and second order of the effect of x_2^2 are the significant model terms ($P<0.05$). Other model terms were not statistically significant ($P>0.05$), which means they had little impact on the BMP (CH_4 yield). Therefore, the nonsignificant terms were removed from the model.

Table 3- 7

Total BMP result, Substrate BMP and R_{max}

Trial NO.	Designed temperature °C (x_1)	Designed BWC (G vs added/L) (x_2)	Total BMP (ML CH_4/ G VS added)	Substrate BMP (ML CH_4/ G VS added) Y	R_{max} (mL CH_4/d)
1 (-1, -1)	25.0	3.00	116.64 ±0.69	66.38±0.24	20.13
2 (1, -1)	45.0	3.00	183.72 ±0.93	129.89±1.30	30.48
3 (-1,1)	25.0	7.00	111.23±25.17	77.20±26.01	12.34
4 (1,1)	45.0	7.00	128.57±1.97	93.64±1.86	23.97
5 (-1.41,0)	21.0	5.00	120.66±2.65	79.12±2.88	12.26
6 (1.41,0)	49.0	5.00	191.31±3.36	148.64±3.99	30.61
7 (0, -1.41)	35.0	2.00	106.11±10.23	48.17±13.4	10.03
8 (0,1.41)	35.0	8.00	95.92±2.04	67.30±3.37	13.93
9 (0,0)	35.0	5.00	146.06	103.69	35.13
10 (0,0)	35.0	5.00	146.06	104.98	32.19
11 (0,0)	35.0	5.00	146.17	105.7	29.13
12 (0,0)	35.0	5.00	141.01	97.96	22.55
13 (0,0)	35.0	5.00	161.66	121.96	32.98

Inoculum blank	35.00	0.00	91.62±18.08	n/a	n/a

Note:

1. Total BMP and substrate BMP presented as average value ± standard deviation (except central points).
2. R_{max} is value of highest slope for average CH_4 production for each trail.
3. n/a is referred as not applicable.

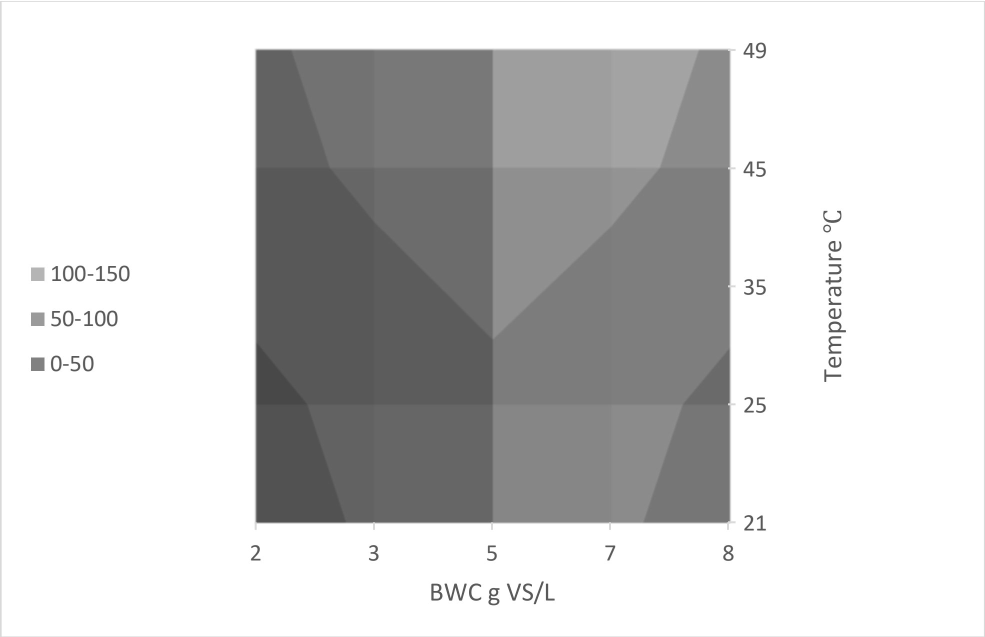

Figure3- 7. Two-dimensional contour graph of Temperature and concentration influence on BMP

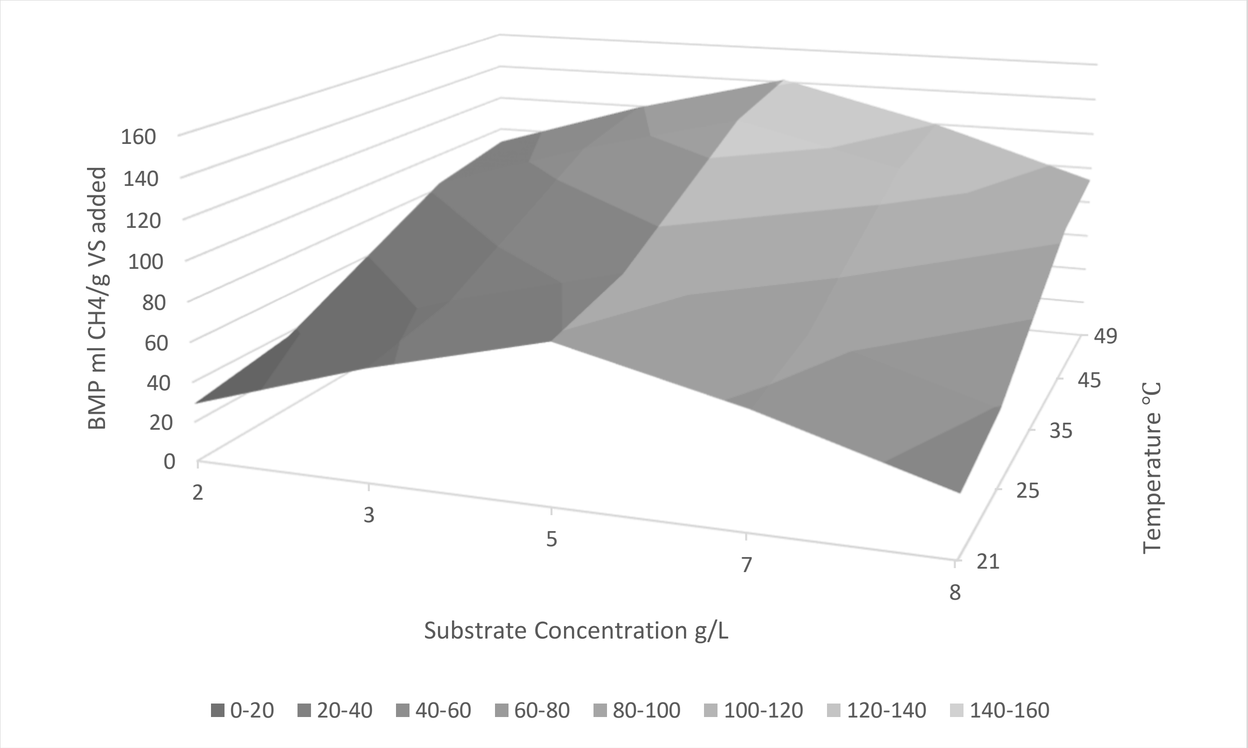

Figure3- 8. Three-dimensional graph of Temperature and concentration influence on CH_4 yield

The model results are presented as both a two-dimensional contour graph and three- dimensional surface response graph presented in Figure 3-7 and Figure 3-8. The figures present a clear peak, indicating that the optimum conditions are inside of the design boundary. According to Equation 3-6, at the temperature of 49℃ and BWC around 5 g/L the maximum BMP result is obtained, which is around 141.4 mL CH_4/g VS added.

Biomethane production from brewery wastewater has been reported in several studies. However, there are no studies that we found evaluating the co-digestion of brewery wastewater with dairy manure. Youngsam et al. (2018), co-digested brewery wastewater with food waste in a BMP trial comparing mixing ratios and reported 176-263 mL CH_4/g VS added, with the lower range similar to the optimum value found in this study. These results demonstrate that at optimum VS loading of 5 g VS/L, both mesophilic and thermophilic conditions produce between 100-150 mL CH_4/g VS with methane production declining at lower temperatures as well as at both lower and higher loading rates. The temperature effect can be explained by the higher lag time for the psychrophilic bottles, which exhibited low methane percent for the first part of the study. The BMP limitation observed at low substrate concentrations could be due to limited substrate while high substrate concentrations could result in substrate and product inhibitions (Ginkel, Sung, & Lay, 2001).

Table 3- 8

ANOVA for depended variable: BMP

Source	Sum of squares (Adjusted)	df	Mean of squares (Adjusted)	F-Value	P-Value.
Corrected model	8045.3	3	2681.77	17.26	0.000
$\boldsymbol{x_1}$	3968.1	1	3968.06	25.54	0.001
$\boldsymbol{x_2}$	3942.6	1	3942.58	25.37	0.001
$\boldsymbol{x_2}^2$	4076.3	1	4076.27	26.23	0.001
Error	1398.5	9	155.39		
Total	9443.80	12			
SD	12.47	R^2	0.8519	$adj - R^2$	0.8026

3.4.6.2 Effect of temperature and concentration on R_{max}

Similarly, the R_{max} data were analyzed to obtain regression equation 3-7 with ANOVA results presented in Table 3-8. The main effect of x_1 (temperature), x_2 (BWC) and second order of the effect of x_2^2 are significant terms (P) for the model. Other model terms which were not statistically significant (P>0.05) were removed from the regression equation. However, the R_{max} regression model had a lower R^2 (0.7259) than the BMP regression model and was below the 80% threshold to be considered a good fit.

$$Y = -33.64 - 0.60x_1 + 16.74x_2 - 1.72{x_2}^2 \quad (3\text{-}7)$$

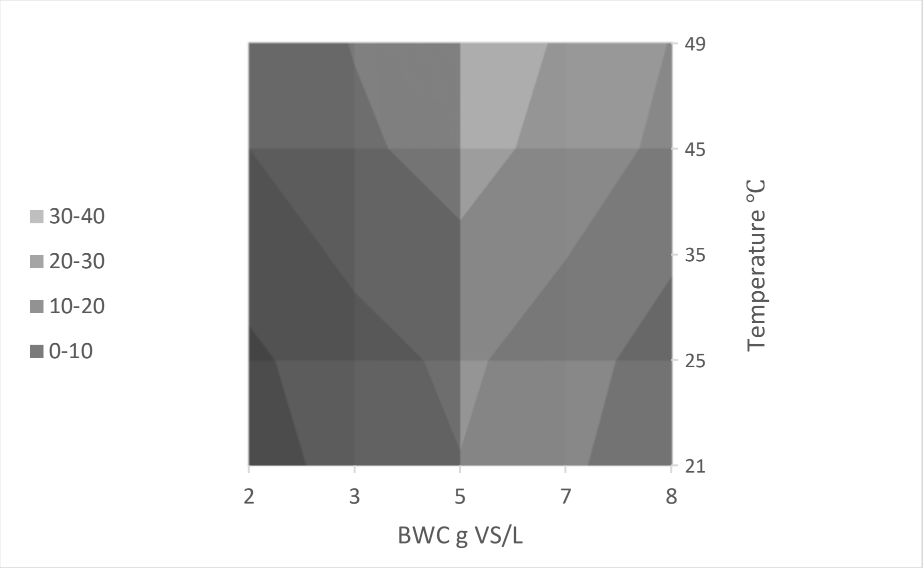

Figure3- 9. Two-dimensional contour graph of Temperature and concentration influence on R_{max}

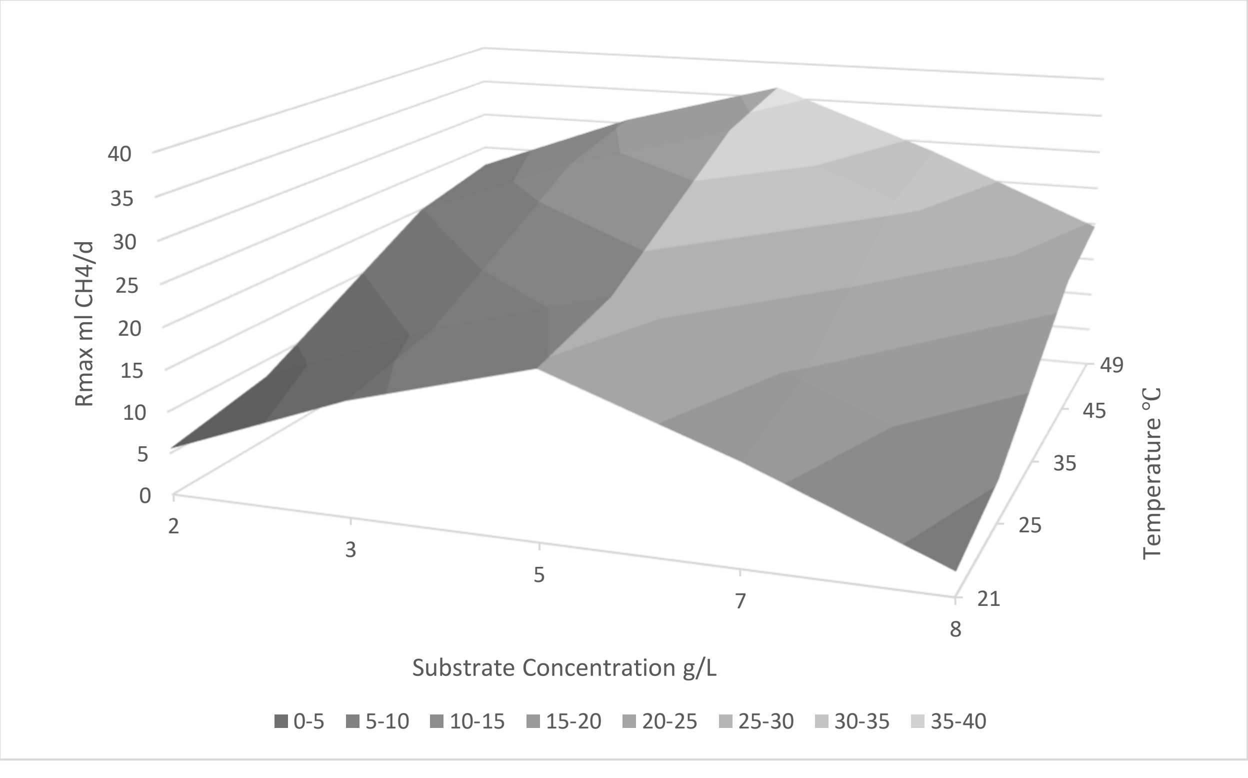

Figure3- 10. Three-dimensional graph of Temperature and concentration influence on R_{max}

The model results are presented as both a two-dimensional contour graph and three- dimensional surface response graph presented in Figure 3-9 and Figure 3-10. Both figures presented a clear peak, indicating that the optimum conditions are inside of the design boundary. According to Equation 3-7, at the temperature of 49℃ and BWC around 4.9 g/L the maximum R_{max} was 36.5 mL CH_4/ day. The R_{max} regression returned the same optimum conditions as the BMP regression, with the highest values corresponding to conditions of T=49°C and BWC =5 g/L. R_{max} was also clearly affected by both temperature and OLR. Similar to the BMP results, R_{max} was shown to be significantly related to temperature, roughly doubling between psychrophilic and thermophilic conditions. The effect of OLR on R_{max} was likely the same as for the BMP results, with low OLR constraining microbial growth through a lack of substrate and high OLR creating conditions of substrate or product inhibition.

Table 3- 9

ANOVA for depended variable: R_{max}

Source	**Sum of squares (Adjusted)**	**df**	**Mean of square (Adjusted)**	**F-Value**	**P-Value.**
Corrected model	702.57	3	234.19	7.95	0.007
$\boldsymbol{x_1}$	286.86	1	286.86	9.73	0.012
$\boldsymbol{x_2}$	369.23	1	369.23	12.53	0.006
$\boldsymbol{x_2}^2$	407.29	1	407.29	13.82	0.005
Error	265.28	9	29.48		
Total	967.85	12			
SD	5.43	R^2	0.7259	$adj-R^2$	0.6345

3.5 Conclusion

In batch experiments, brewery wastewater was digested with a dairy manure digestate innoculum for biomethane production by anaerobic digestion considering the effect of temperature and organic loading. Findings from this study are:

1. High strength brewery wastewater was shown to have a high degree of treatability at all temperature conditions with 82-91% removal of VS and 77-88% removal of sCOD observed.
2. The psychrophilic rate constant was significantly lower than both the mesophilic and thermophilic rate constants by roughly a factor of two, while the thermophilic rate constant was highly variable, suggesting process instability.
3. An optimum organic load of 5 gVS/L was observed with both lower and higher concentrations found to inhibit the AD process corresponding to an I/S ratio of 1.
4. Significantly lower stabilized methane content of 47% was observed at psychrophilic conditions compared with 65 and 67% at mesophilic and thermophilic conditions, respectively.
5. Central composite design models were developed to describe CH_4 yield (BMP) and R_{max} as functions of temperature and organic loading. The operational condition of biomethane production was also optimized with RSM and a desirability function. Optimized operational parameters were achieved from the regression model at 49 °C and BWC 4.9 g/L. Correspondingly, the BMP and R_{max} were 141.4 mL CH_4/g VS added and 36.9 mL CH_4/ day, respectively. However, in terms of process stability the preferable operational condition T=35℃ & BWC=5 g/L is recommended. At this condition methane yield is 110.1 CH_4/g VS added & maximum methane daily production is 28.1 CH_4/ day, which is similar to the maximum result.

3.6 Reference

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Chapter 4

Optimization of Operational Conditions for a Small-Scale Brewery Moving Bed Biofilm Reactor (MBBR) Wastewater Treatment System

4.1 Abstract

An on-site brewery wastewater treatment system equipped with two Moving bed biofilm reactor (MBBR) reactors was evaluated from October 12th, 2018 to February 10th, 2020 in Beau`s All-Natural Brewing Company, Vankleek Hill, Ontario, Canada. The aim of the study was to characterize the wastewater production (flow and organic loading rate), evaluate operating conditions and performance of the MBBR system and recommend improvements. Discharge from the brewery is highly variable for both organic and hydraulic loading with flow balancing recommended. The MBBR full-scale reactors operated at relatively stable conditions at Surface area loading rate (SALR) of less than 25 $g/m^2 \cdot d$ and dissolved oxygen (DO) greater than 2 mg/L. Kinetic rate constants for suspended growth and attached growth biomass in the reactors were found to be similar at 0.0764-0.0908 h^{-1}, however, much larger attached growth mass in the reactors suggests that only a fraction of the attached growth biofilm is active. Effluent recycle was shown to be effective at controlling filamentous bacteria (type-0041) sludge bulking, reducing Mixed liquor suspended solids (MLSS) and effluent soluble chemical oxygen demand (sCOD) concentration.

4.2 Introduction

The Brewery industry is facing a world-wide transformation with rapid growth of small-scale operations. Due to their growing popularity, 93% of breweries in Canada at the end of 2018 were classified as a small-scale breweries including: craft brews, microbreweries, nanobreweries and brewpubs (Beer Canada, 2019). The brewery industry has a significant demand for water and energy (Olajire, 2012) and produces a large amount of wastewater (Van der Merwe & Friend, 2002), which is highly variable in both organic and hydraulic loading (Table 4-1). Especially, for small-scale breweries, wastewater production can vary widely based on production and cleaning practices and can range from 6-10 L wastewater per L beer produced (Brewers Assiociation, 2013; Brewers Association, 2011; Van der Merwe & Friend, 2002).

Table 4-1 presents typical characteristics of brewery wastewater compared with an Ontario microbrewery and Beau`s All Natural Brewing company (this study). As shown, there is a large variation in the concentration of organic matter, which is especially true for small-scale breweries. Since brewery wastewater is a high strength wastewater with high variability in both organic loading and flow, if discharged without pre-treatment, it can destabilize the operation of municipal wastewater treatment plants in small communities (Brewers Assiociation, 2013). Additionally, municipalities apply Over-Strength Discharge Fees (ODFs) to industries discharging high strength wastewater to the municipal sewer, which can represent a considerable cost to breweries. For both these reasons, the onsite pre-treatment of brewery wastewater is increasingly becoming a requirement for small brewery operations.

Table 4- 1

Characteristics of typical brewery with two small-scale breweries

Parameters	**Brewery Wastewater Composition**		
	Typical (Brito et al., 2007; Driessen & Vereijken, 2003)	Wellington Brewery (Swain, 2019)	This Study Beau`s Brewery (Avg±SD)
COD (mg/L)	2,000-6,000	3,700-30,000	3,647±2,031
BOD (mg/L)	1,200-3,600	n/a	2,598±1,377[a]
TSS (mg/L)	200-1,000	10-28,367	897±954
T (°C)	18-20	n/a	15.9-30.9[b]
pH	4.5-12	4.99-7.62	n/a[c]
TN (mg/L)	25-80	78.3-80.9	13.6±13.6[d]
TP (mg/L)	10-50	30-2,335	12.2±10.3[e]

Note: n/a is referred as not available

[a] sCOD

[b] Range in MBBR1

[c] pH balancing maintained approx. pH = 7.0

[d] NH_4^+-N

[e] SRP

The most common treatment method for brewery wastewater has been anaerobic digestion due to the high organic content of the wastewater and the large scale of traditional breweries (Metcalf & Eddy, 2014). Anaerobic treatment accomplishes high organic removal with less biomass production (Metcalf & Eddy, 2014). However, start-up time and required operational time (hydraulic retention time HRT) are longer, requiring a larger footprint for operation (Simate et al., 2011). Additionally, the anaerobic process is highly sensitive to operating conditions (i.e. pH, temperature, organic loading rate); and therefore, requires significant operational oversight. With the trend to smaller breweries, which are typically located in urban areas with limited space (Van der Merwe & Friend, 2002), the advantages of anaerobic treatment are to a certain degree offset by the advantages of aerobic treatment including a smaller footprint (lower HRT) and operational simplicity. The requirements for the on-site treatment of wastewater from small breweries include high removal efficiency, high stability with variable loading, simple operation with a relatively small footprint. Traditional activated sludge reactors treat relatively low organics (<2000 mg/L sCOD) compared to anaerobic treatment (>2000 mg/L sCOD) and have the disadvantages of high sludge production and high energy costs (Metcalf & Eddy, 2014). The application of advanced aerobic treatment technologies can mitigate these disadvantages by increasing treatment efficiencies and reducing sludge production.

The Moving Bed Biofilm Reactor (MBBR) is a wastewater treatment technology utilising attached growth submersed carriers, which can significantly increase the surface area for the microorganisms to interact with the organic matter in the reactor (Metcalf & Eddy, 2014; Ødegaard, 1999). Carriers are usually designed with a density of 1 g/m^3 (Ødegaard et al., 2000), allowing the carriers to move freely in the reactor (Metcalf & Eddy, 2014; Ødegaard, 1999; Ødegaard et al., 2000). This results in the actual volume being equal to the active volume, which is an advantage compared to other biofilm technologies. The MBBR technology has reported high removal efficiencies for organics and ammonia for both municipal and industrial wastewater (Metcalf & Eddy, 2014; Ødegaard, 1999; Ødegaard et al., 2000).

The MBBR system can achieve favourable removal efficiency for the small flow applications and can work as a stand-alone technology or can be operated as a post-treatment for the anaerobic system (Boyle et al., 2019). Biofilm in the reactor is always attached on the carriers and can reduce the suspended solids in the reactor with MLSS typically lower than 800 mg/L; which can reduce sludge management costs (Bassin et al., 2016; Johnson et al., 2000; Metcalf & Eddy, 2014; Ødegaard, 1999; Rusten et al., 1992). Compared to the traditional activated sludge systems, the MBBR system can provide a larger surface area for reaction, lower HRT, continuous operation, no sludge recycle, and low sludge production (Aygun et al., 2008; Ibrahim et al., 2016; Ibrahim et al., 2012; Metcalf & Eddy, 2014; Ødegaard, 1999; Ødegaard et al., 2000). Compared to other biofilm technologies, the MBBR system does not need to be backwashed (McQuarrie & Boltz, 2011). The MBBR system can be operated in a small footprint, making it favourable for applications with space constraints (Ødegaard, 1999; Ødegaard et al., 2000).

The MBBR system is broadly applied to municipal and industrial wastewaters; however, there is limited information on MBBR systems treating brewery effluent. Moreover, most of the research has been at the lab-scale. A previous laboratory study was conducted by Boyle (2019) to optimize the MBBR treatment of the Beau's wastewaters. In this study, no difference was observed in removal efficiency between suspended growth (SG) and two different types of carriers at the SALR range from 10-55 g sCOD/m$^{2}\cdot$d with 12h HRT. When HRT was reduced to 3h, there were significant drops in the SG removal efficiency; however, there were no significant changes in removal performance for both carriers.

Sludge bulking is one of a common problem for suspended growth systems. Usually, sludge bulking has a high frequency for occurring when wastewater contains high carbohydrate, such as food and beverage processing wastewater (Schwartz et al., 1980). Sludge bulking is a sludge that settles slowly, compacts poorly and has a high-volume sludge index and those properties can result in a thick sludge blanket in the secondary clarifier. According to the type of microorganism sludge bulking can be divided into filamentous and nonfilaments bulking. Among these two types, filamentous bulking usually has a higher likelihood to occur (PASVEER A, 1969; Schwartz et al., 1980; Sezgin et al., 1978; Wanner, 1994). Sludge bulking is unlikely in pure MBBR systems, however, can be a problem in IFAS systems which have both attached and suspended growth.

Usually, Filamentous bulking results from extreme conditions, such as low dissolved oxygen (DO), lack of nutrients, high organic loading rate (OLR) and extreme pH levels. To prevent filamentous bulking DO should be higher than 2 mg/L (Sezgin et al., 1978), pH range between 6 to 8 and a balanced carbon to nutrients (mostly nitrogen and phosphorus) ratio (Sezgin et al., 1978). The following methods can be used to address filamentous bulking (Fan et al., 2018; PASVEER A, 1969; Schwartz et al., 1980; Sezgin et al., 1978; Wanner, 1994):

a. Sludge recirculation: sludge from secondary clarifier returns to the reactor
b. Kraus process: extra aeration before sludge recirculate back to the reactor
c. Chemical addition: add chemicals to reduce filamentous bacteria in the reactor
d. Chemical coagulation: add coagulants in the secondary clarifier for assist sludge to settle down

As presented above, various methods can assist system recover after sludge bulking. Sludge recirculation is the most common method, which is easy to apply and economically feasible. The Kraus process is an upgrade process for sludge recirculation but needs extra energy for the aeration. Chemical addition usually uses chlorine and hydrogen peroxide to reduce the filamentous bacteria in the system. However, this method poses significant risk to destroy other types of bacteria. Chemical coagulation can settle the sludge efficiently but does not have a significant effect to eliminate filamentous bacteria (Fan et al., 2018; PASVEER A, 1969; Schwartz et al., 1980; Sezgin et al., 1978; Wanner, 1994). Delatolla (2019) suggested that an increase in internal flowrates within each reactor will result in increased stress on the carrier biofilm; thus promoting biofilm attachment while reducing suspended growth concentrations and addressing problems with bulking sludge. Stable operating conditions are also important to preventing sludge bulking problems, which can be achieved through flow equalization.

The objectives of this study are to:

- Characterise the wastewater production at Beau`s brewery in terms of flow and organic loading and recommend system design improvements in terms of flow balancing.
- Characterise and evaluate the full-scale treatment of brewery wastewater with an MBBR system in terms of organic loading, DO and temperature (T).

- Evaluate the effect of effluent recycling on filamentous bulking and treatment efficiency.

4.3 Materials and methods

4.3.1 Full-scale brewery wastewater treatment system (Beau`s All Natural Brewery Company)

Beau`s brewery implemented a full-scale wastewater treatment system in October 2016. As shown in Figure 4-1, the brewery segregates two waste streams for off-site valorization: the spent grains are sold to a local farm for animal feed and the high-strength liquid wastes (waste yeast, waste beer first rinse from the fermenters) are hauled to a local farm anaerobic digestor for biogas production. The remaining wastewaters from the Clean-in-Place (CIP) processes and the bottle washing operation are treated in the on-site wastewater treatment system prior to discharge to the municipal sewer. The treatment system comprises an equalization tank, a pH dosing tank and two MBBR reactors in series each followed by a settling tank. The designed flow for the on-site wastewater treatment system is 40 m^3/d with each MBBR reactor having a 12h design HRT. MBBR reactor 1 (MBBR1) was filled with Kontakt carriers (surface area 500 m^2/m^3) and MBBR reactor 2 (MBBR2) was filled with Kontakt and K5 (surface area 800 m^2/m^3) in 13 to 1 ratio. Both reactors have a filling percentage of 70%. The sludge produced from the reactor is stored in the sludge tank which is periodically hauled off-site.

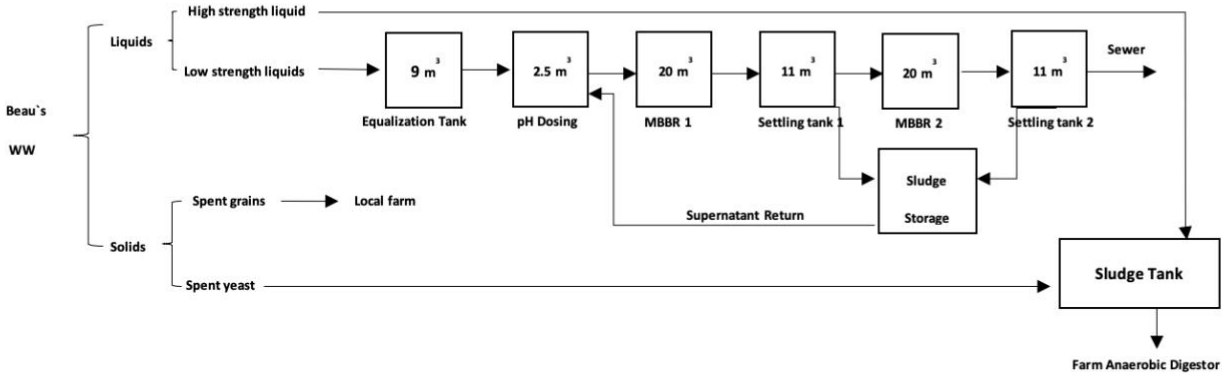

Figure 4- 1. Beau's All Natural Brewing Company wastewater treatment train schematic

4.3.2 Experimental set-up

4.3.2.1 Sampling and Recycle Plans for different time periods

Wastewater samples were collected from the dosing tank (inlet), MBBR1, and MBBR2 using three ISCO-6712 auto samplers. Each daily sample (1L) consisted of a composite of four samples (250 ml) collected at 6 hours intervals with sample bottles containing H_2SO_4 for sample preservation. The samples were collected every 14 days from November 5th 2018 until February 12th 2020 and taken back to the University of Ottawa's Environmental Engineering laboratory for analysis. Once at the lab, samples were stored at 4±1°C prior to analysis with analyses conducted on average within 2 weeks.

Two effluent recirculation strategies were applied to the system to address sludge bulking issues and increase the organic removal efficiency via promote growth of AG. The effluent recirculation strategies are summarized in the Table 4-2.

Table 4- 2

Recycle plan summary for different period

Operation parameters	*No Recycle*	*Recycle Strategy 1 (S1)*	*Recycle Strategy 2 (S2)*
start date	Oct 12th,2018	May 17th, 2019	December 13th,2019
Recycle pathway	n/a	From settling tank 2 to MBBR2	From settling tank 2 to MBBR1
Recycle rate m^3/h	n/a	7.38	7.67
Recycle duration time	n/a	continuous	15min per hour
Designed /Recycle Flow m3/d	40	177	46
Recycle ratio[a]	0	1:4	1:1

n/a is referred as not applicable

[a] recycle ratio = recycle rate compared to design flow

4.3.2.2 Kinetic test

A laboratory bench-test was conducted to simulate the actual condition of Beau`s brewery on-site full-scale wastewater treatment system in order to compare the kinetic rates of MBBR1 and MBBR2 attached growth (AG) carrier biofilm to the suspended growth (SG) bacterial biomass. For attached growth conditions (MBBR1 Kontakt (carrier surface area 500 m^2/m^3), MBBR2 Kontakt, and MBBR2 K5 (carrier surface area 800 m^2/m^3)), SALR=20 g sCOD /$m^{2\cdot}$d for each trial was maintained and the influent brewery wastewater collected from the dosing tank was used as substrate. For suspended growth, samples from the reactors were collected and the SG biomass was settled in an Imhoff cone with the supernatant decanted. Each Trial condition was triplicated, and a control wastewater sample was run without carriers or SG biomass added. As shown in Figure 4-2, 1 L plastic bottles were used for the jar tests with a working volume of 450 ml. To maintain similar SALR, the trials with Kontakt carriers had 30% carrier fill while the trials with K5 carriers had 20% carrier fill. The experiment was conducted over a 72 hours period with samples collected periodically and analysed for sCOD. Biomass and MLSS concentrations were measured at the beginning of the experiment. DO levels were maintained at greater than 2 mg/L using an aquarium air compressor in each bottle.

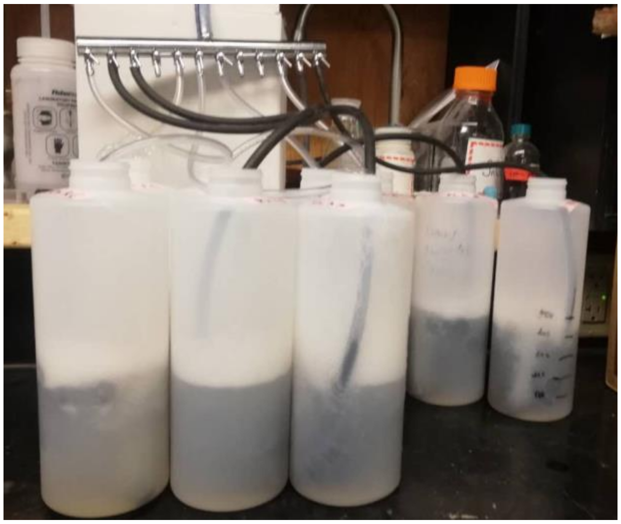

Figure 4- 2. Kinetic bench-test setup

4.3.3 Analytical methods

4.3.3.1 Biofilm thickness and sludge morphology

Five replicate carriers were collected from both MBBR1 and MBBR2 reactors at each sampling event. The carriers were imaged using a Zeiss Stereomicroscope Stemi 305 (Zeiss, US, VA) within 20 min of removal to minimize the potential effects of biofilm dehydration. During this process, five images across each carrier were captured corresponding 25 total images per reactor per time (Young et al., 2016). The acquired images were analyzed for thickness on Digimizer software resulting in a total of over than 2500 thickness measurements per sample. Biofilm Bulking sludge morphology were observed through the use of a Vega II-XMU variable pressure scanning electron micro- scope (VPSEM) (Tescan USA Inc., USA, Pennsylvania) with 100 magnification.

4.3.3.2 Biofilm mass

Five replicate carriers were collected from both MBBR1 and MBBR2 reactors at every sampling event. Within 20 minutes the carriers were placed in a drying oven at 105°C for 24 hours. After 24 hours, the carriers were placed in a desiccator for half an hour for cooling to room temperature. Room temperature carries weighted (W1) and cleaned with warm water and a stiff-bristled brush and placed back in the drying oven at 105°C for another 24 hours. The cleaned and dried carriers were placed in a desiccator for half an hour to cool down to the room temperature and then weighed (W2). The difference between W1 and W2 is the mass of the biomass (Schopf et al., 2018; Young et al., 2016).

4.3.3.3 Flow rate measurement

Inlet wastewater flow to the brewery wastewater treatment system was measured using a transit time flow meter (Greyline TTFM 6.1). The flow meter records every 15 seconds and is reported as an hourly average. For increasing the accuracy, all the flow data applied negative flow filtration, which reported negative values as 0 m^3/s. Daily flow rate and peak flow rate obtained from the hourly averages.

4.3.3.4 DO, Temperature and pH measurement

The DO, T and pH were measured by brewery staff with a Multi-Parameter meter with attached DO, pH probe (VWR, Ontario, Canada) and recorded daily for both MBBR reactors 1 and 2 times on weekdays.

4.3.3.5 General wastewater sample analysis

The soluble content of the sample was measured after filtration through 0.45 µm filter. Total chemical oxygen demand (tCOD) and soluble chemical oxygen demand (sCOD) were measured according to HACH "Test N Tube^{TM} Vials" standard method 80000 (HACH, 2013), which is equivalent to Standard Methods 5220 D (APHA,2012). Total suspended solids (TSS = MLSS) were measured according to Standard Methods (APHA,2012). Ammonia (NH_4-N) and orthophosphate (SRP) were measured according to HACH "Test N Tube^{TM} Vials" standard method 10031 and 8114 (HACH, 2013).

4.3.4 Statistical Methods

Statistical significance tested via single-factor ANOVA on the Excel with p values data, including less than 0.05 considered significant.

4.3.5 Numerical calculations

Surface area loading rate:

$$SALR = \frac{[\,sCOD\ added]\left(\frac{g}{L}\right) \times flow\ rate\left(\frac{L}{d}\right)}{Total\ surface\ area\ m^2}\left(\frac{g}{m^2}\cdot d\right) \qquad (4-1)$$

Equation 4-1 is a general equation for calculating SALR, while during the kinetic test instead of the flow rate ratio of volume to HRT is applied to the equation.

Surface area removal rate:

$$SARR = \frac{[\,sCOD\ removed]\left(\frac{g}{L}\right) \times flow\ rate\left(\frac{L}{d}\right)}{Total\ surface\ area\ m^2}\left(\frac{g}{m^2}\cdot d\right) \qquad (4-2)$$

Total surface Area:

$$Total\ surface\ area = Carrier\ surface\ area \times reactor\ size \times filling\ percentage \quad (4-3)$$

4.4 Results and Discussion

4.4.1 General Performance

The wastewater quality was monitored from October 12th, 2018 to February 12th, 2020, while flow was monitored from Dec 3rd, 2018 to Feb 12th, 2020. Water quality parameters included: tCOD, sCOD, TSS, NH_4^+-N, and SRP. Soluble nutrient levels were quite low in the wastewater (NH_4-N = 13.6 ± 13.6 mg/L; SRP = 12.2 ± 10.3 mg/L) but showed only a 25% reduction throughout the treatment train, suggesting nutrient cycling within the reactors. The COD results are presented in Figure 4-3 with the sCOD presented in Figure 4-4, and Table 4-3 summarized general operating parameters for each time period. As mentioned previously, increase in internal flowrates can promote biofilm to attach on the carrier (Delatolla, 2019). Therefore, total flow was increased in May 2019, by diverting low-strength bottle washing waters to the treatment system (from direct discharge to the sewer), which resulted in higher flows and reduced COD concentrations in the dosing tank. As a significant parameter- SALR, which also presents the organic loading rate normalized to carrier surface area, from Table 4-3 can observe that overall SALR for MBBR1 was ranged from 5.8 to 11.03 g sCOD/ $m^{2}\cdot$d and for MBBR 2 was ranged from 1.9 to 6.99 g sCOD/ $m^{2}\cdot$d.

Reductions in sCOD concentration from Dosing to MBBR1 were very similar over the first three operating periods with no recycle, ranging from 1637 to 1724 mg/L sCOD while average SALR dropped from 11.0 to 6.6 g sCOD/ $m^{2}\cdot$d from the fall to both winter and spring periods, and suggests oxygen limitation in MBBR1, where DO levels averaged between 1.9-2.0 mg/L (Table 4-3). The reductions in sCOD from MBBR1 to MBBR2 over the first three operating periods ranged from 613 to 1100 mg/L sCOD and increased with increasing concentration, suggesting a carbon limitation, while SALR varied from 1.9 to 7.0. MBBR2 outlet sCOD concentrations ranged from 295 to 625 mg/L, and generally exceeded the sewer use bylaw of 300 mg/L BOD_5 during the non-recycle periods. During the recycle periods SALR remained within comparable ranges to the non-recycle periods but with higher flows and lower influent (dosing) concentrations. As with the previous periods, limited DO could have been the limiting factor in MBBR1 removal, while in MBBR2, sCOD concentrations were likely reaching a limiting state at 110 mg/L. Importantly, the effluent limit was met under both recycle scenarios.

Figure 4- 3. Average COD Concentration with Time

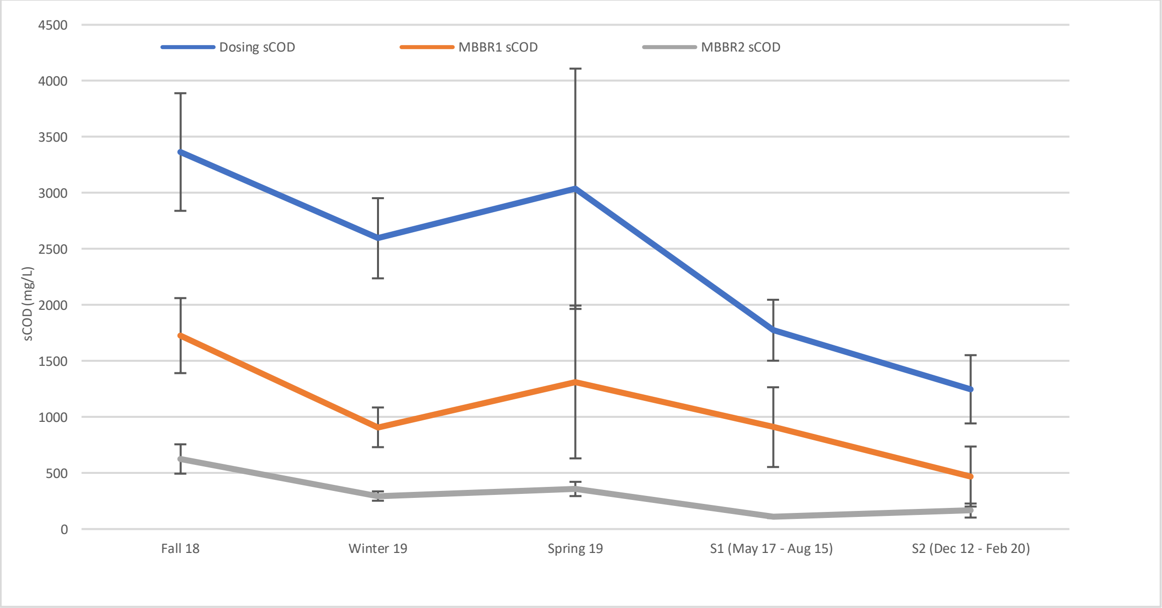

Figure 4- 4. Average sCOD concentration with time

Table 4- 3

Summary of other general parameters for the different time period

Time	MBBR1					MBBR2				
	n	SALR g sCOD/ m^2·d	DO mg/L	pH	T °C	n	SALR g sCOD/ m^2·d	DO mg/L	pH	T °C
Fall 18	24	11.03±6.55	N/A	N/A	N/A	23	6.99±3.58	N/A	N/A	N/A
Winter 19	56	6.4±5.14	1.89±1.32	7.8±0.5	21.9±2.7	66	1.9±2.08	3.7±2.0	7.9±0.36	23.5±2.1
Spring 19	14	6.6±7.53	2.00±1.63	7.6±0.7	22.4±2.1	34	5.0±5.32	3.6±2.1	7.8±0.54	24.8±0.6
S1	14	5.8±4.01	1.22±1.16	7.0±1.2	27.2±1.9	50	5.8±3.21	1.9±1.7	7.5±0.8	29.8±1.8
S2	11	10.2±5.84	0.01±0.01	7.3±2.1	22.5±0.4	8	4.8±3.07	0.0±0.0	7.1±2.0	23.8±0.9

Note: At fall 2018 monitoring general parameters (DO, pH, and T) is not started yet and for S2 DO recording is not available.

4.4.1.1 Flow equalization

As shown in the schematic of the brewery wastewater treatment system, an equalization tank was operated in a 9 m^3 volume; however, actually has an operating volume of only 4.5 m^3. An analysis of average daily flows as well as variation within each 24-h period was conducted to size a balancing tank for current flow conditions. Hourly flow data was analysed from September 2019 to February 2020 to evaluate temporal flow variation (Figure 4-5). Figure 4-5 present variation by the week and day respectively. Each daily and hourly value presents the average value from six-months of flow data (from September 2019 to February 2020). As shown in Figure 4-5 (A), there was a significant difference between weekdays and weekends. Weekday flow rate was slightly higher than the design flow (40 m^3), while weekend flows were much lower. This operational flow data supports the implementation of weekly flow balancing which would reduce hydraulic and organic loading during weekdays and increase loading over the weekends, when there could be carbon limitations in the system. Fluctuating organic loading rates are harmful to maintain a healthy microorganism community and results in unexpected problems in the system, such as sludge bulking. As Figure 4-5 (B) presents, there was no significant average variation during the day, therefore hourly flow balancing was not considered in the new equalization tank design.

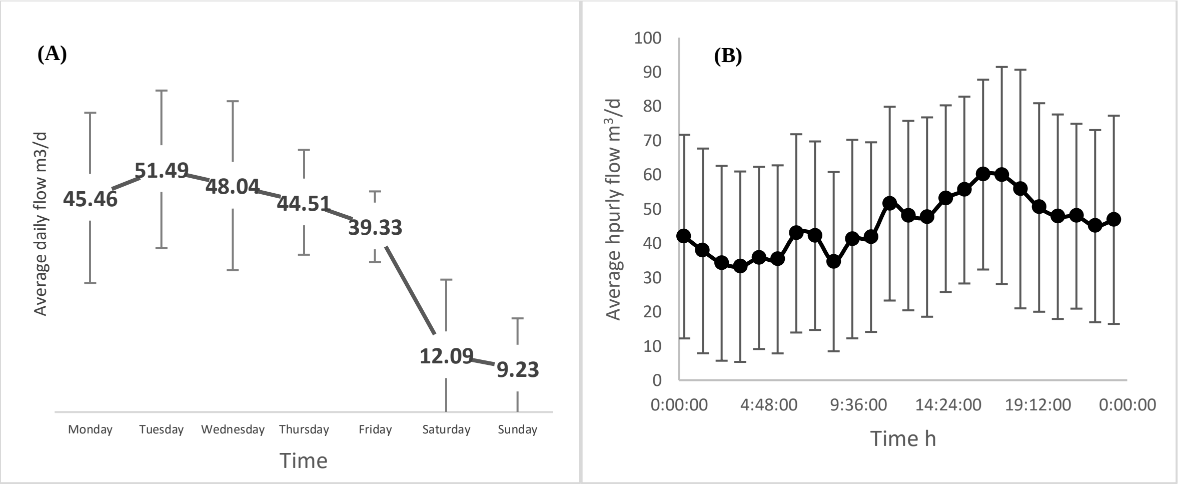

Figure 4- 5. Average daily and hourly flow for days of the week from September 2019 to February 2020: (A) is daily average and (B) is weekday hourly average

Figure 4-6 shows cumulative volume from the actual flow rate and cumulative volume from the average daily flow during the week. The average daily flow data (Figure 4-5(A)) applied for the equalization tank design with the equalization tank volume equal to the sum of the lowest cumulative volume and highest cumulative volume, which from Figure 4-6 is 51 m^3. A 15% safety factor was applied to the tank design and the new equalization tank required volume is 59m^3. For practical propose, the actual tank volume can round to 60m^3.

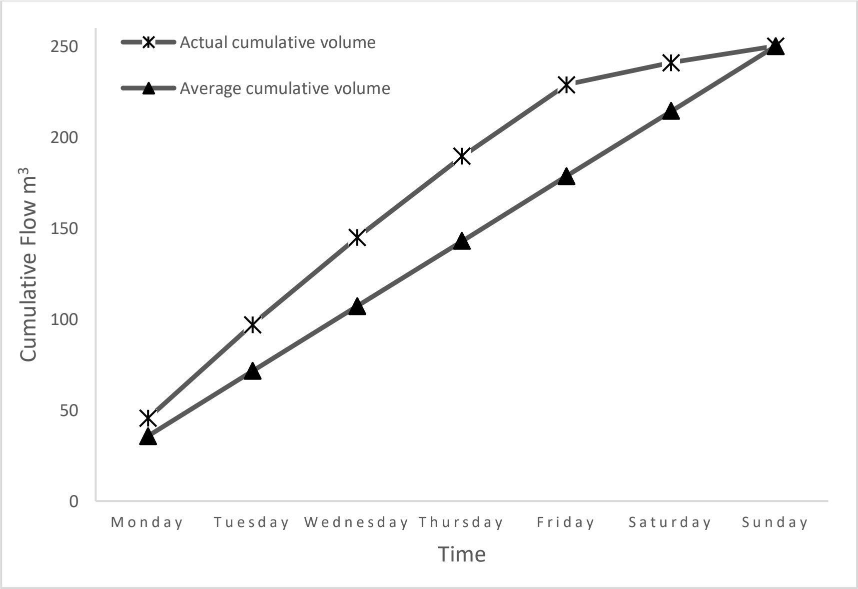

Figure 4- 6. Actual cumulative weekly volume compares to the cumulative volume for average weekly flow

4.4.2 Operating Parameters

4.4.2.1 SALR vs SARR

Surface area loading rate versus removal rate for both MBBR 1 & 2 are presented in Figure 4-7. The SALR can be divided into three ranges: 0-9.9, 10-24, and 25 and greater g sCOD/ m^{2}·d considered as low, medium and high SALR ranges, respectively. Typical carbon removal reactors operate between 15 to 20 g sCOD/ m^{2}·d SALR (Metcalf & Eddy, 2014). However, the actual Beau`s brewery wastewater treatment system operated at lower SALR conditions. Table 4-4 compares removal efficiency with SALR range across both reactors. MBBR1 exhibited similar removal efficiencies of 71 and 69% at low and medium SALR and significantly lower removal efficiencies of 54% at high SALR, where removal kinetics likely transited from first order kinetics with carbon as the limiting substrate to zero order kinetics with oxygen limitation. MBBR2 exhibited significantly higher removal efficiencies at medium SALR (79%) than at low SALR (65%), likely due to carbon limitations at low loading rates. MBBR2 also exhibited higher removal efficiencies at Medium SALR compared with MBBR1 at Medium SALR, again likely due to oxygen limitation in MBBR1.

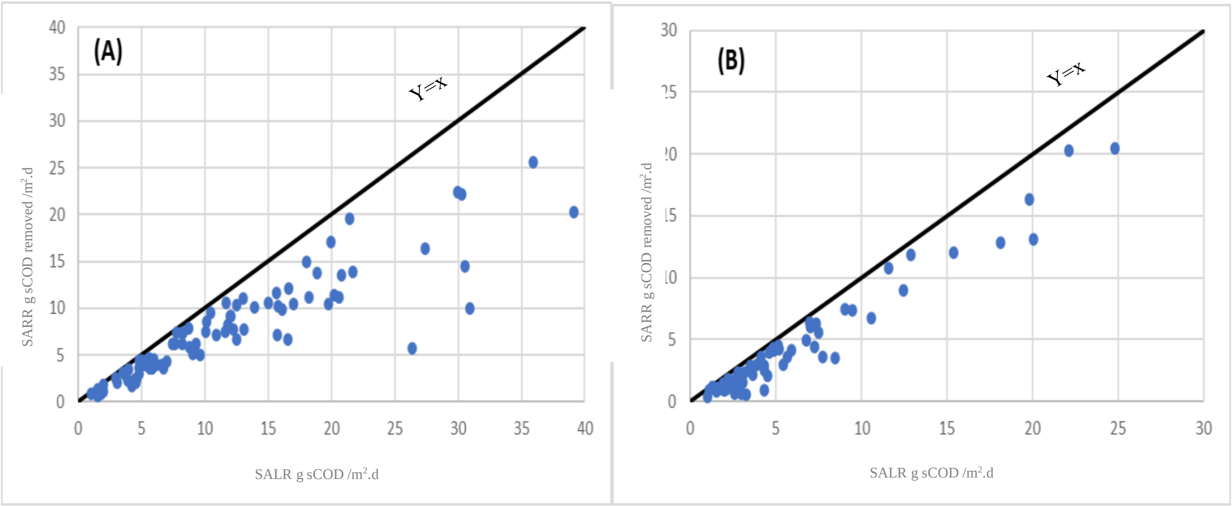

Figure 4- 7. SALR vs SARR, (A) MBBR 1 and (B) MBBR 2

Table 4- 4

sCOD removal efficiency with SALR

	1.0-9.9 g sCOD/ m^{2}·d	10.0-24.9 g sCOD/ m^{2}·d	25.0-40.0 g sCOD/ m^{2}·d
MBBR1	71±15%	69±13% [c]	54±20% [a]
MBBR2	65±20%	79±11% [b,c]	-

[a] Significantly lower than 1.0-9.9 and 10.0-24.9 MBBR1 loading rates

[b] Significantly higher than 1.0-9.9 MBBR2 loading rates

[c] MBBR 2 removal rate significantly higher than MBBR1 at SARL in the range of 10.0-24.9 g sCOD/ m^{2}·d

4.4.2.2 DO and Temperature

Maintaining sufficient dissolved oxygen levels is critical for aerobic treatment, while excess aeration can needlessly increase energy costs. Temperature plays two competing roles in reactor operation: higher temperatures are beneficial to heterotrophic bacteria; however, dissolved oxygen saturation concentrations decline with increasing temperature. SALR was divided into 0-5 and 5-10 g sCOD/ m2·d for both MBBR reactors for analysis as there was limited data at higher SALRs. Figure 4-8 presents the effect of DO on removal efficiency at different SALR ranges. At SALR range from 0-5: for MBBR1 there was significant difference ($p<0.05$) observed between DO range from 0-2 mg/L and over 2 mg/L, which for MBBR1 DO higher than 2 (average removal efficiency 80.3%) had a higher removal efficiency than DO lower than 2 (average removal efficiency is 52.0%); for MBBR2 there were no significant influence observed for different DO ranges. At SALR range from 5-10 g sCOD/ m2·d, for both MBBR1 and MBBR2 there were significant difference ($p<0.05$) observed between DO range from 0-1 and 1-2 mg/L. For both of the reactors the higher the DO level higher the organic removal efficiency. In general DO levels should be maintained above 2 mg/L, however, differences were observed between 1 and 2 mg/L at SALR between 5-10 g sCOD/ m^{2}·d.

In this study, the relation of temperature and organic removal rate at different SALR range (0-40) and seasonal difference were analyzed. Both of the reactor mostly operating at 20-30 °C. However, there were no significant difference of the temperature on organic removal rate at the temperature range higher and lower than 25 °C. Therefore, can conclude that when operating T at 20-30 °C, T cannot affect the organic removal efficiency.

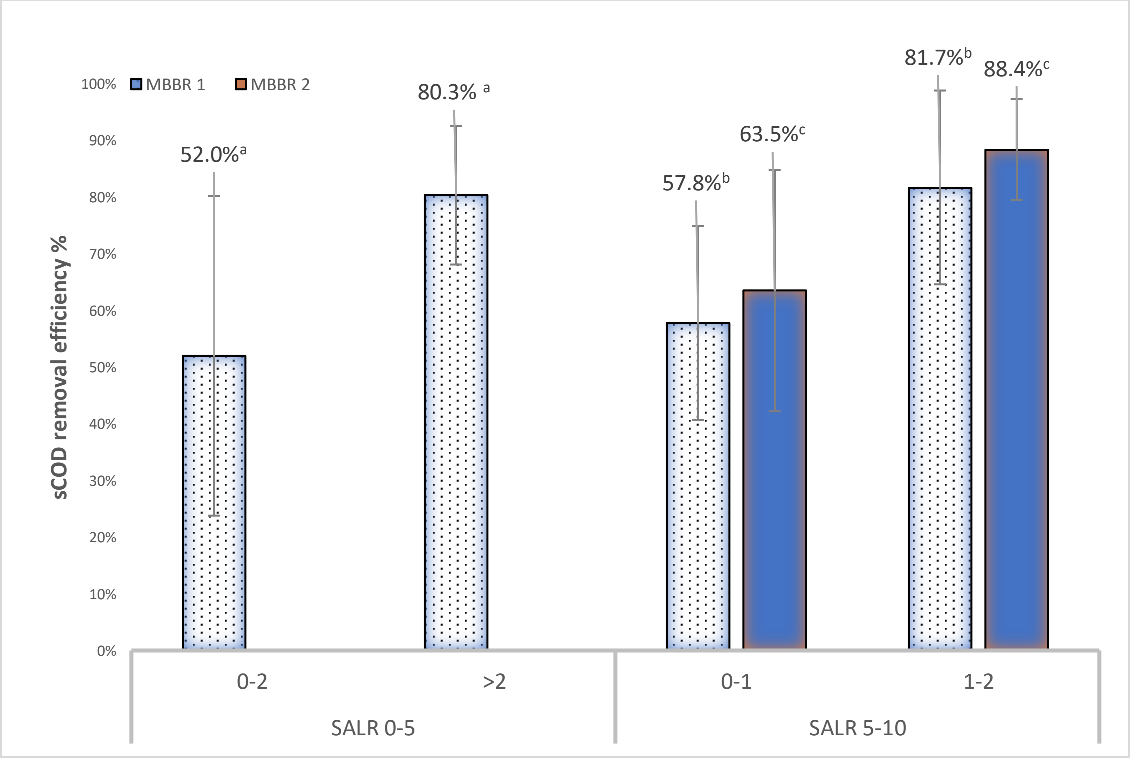

Figure 4- 8. DO effect on removal efficiency at different SALR range for MBBR reactors

[a]. for DO level 0-2 mg/L: n=8, SALR= 2.4 ± 1.6 g sCOD/ m^{2}·d and for DO>2 mg/L: n=14, SALR= 2.8 ± 1.5 g sCOD/ m^{2}·d. Two DO range for this data set presented as statistically significant ($p<0.05$).

[b]. for DO level 0-1 mg/L: n=6, SALR= 7.72 ± 1.32 g sCOD/ m^{2}·d and for DO level 1-2 mg/L: n=14, SALR= 2.8 ± 1.5 g sCOD/ m^{2}·d. Two DO range for this data set presented as statistically significant ($p<0.05$).

[c]. for DO level 0-1 mg/L: n=8, SALR= 7.0 ± 1.7 g sCOD/ m^{2}·d and for DO level 1-2 mg/L: n=5, SALR= 7.8 ± 0.9 g sCOD/ m^{2}·d. Two DO range for this data set presented as statistically significant ($p<0.05$).

4.4.2.3 Kinetic test- comparison of attached growth to suspended growth

A jar test was conducted to study the relative contribution to carbon removal of the biofilm on the carriers and the suspended growth bacteria present in the reactors. The batch-test results are presented in Figure 4-9, with summary kinetics described in Figure 4-10 and Table 4-5. The batch test for all the condition showed first order reaction kinetics, which follow Equation 4-4:

$$C = C_0 e^{-kt} \tag{4-4}$$

where C is final concentration (mg/L), C_0 is the original concentration (mg/L) (In this study is 2250 mg/L), k is rate constant (hr^{-1}) and t is time (hr).

The Kinetic rate constants were quite similar and ranged from 0.0821 to 0.0861 hr^{-1} for SG and from 0.0764 to 0.0908 hr^{-1} for the carriers. Typical k value for multi-component industrial wastewater biodegradability tests in lab-scale aerated batch reactors range over a 30-hr period from 0.029 to 0.147 hr^{-1} (Argaman et al., 2000), which is similar to the ranges observed in this trial. The important difference observed is that the mass of AG biofilm was 2.9 to 3.8 times higher than the mass of SG bacteria. This suggests that not all of the attached growth bacterial mass is active and that a relatively small fraction of suspended growth bacteria in the reactors could be contributing much more significantly to the treatment than their relative mass.

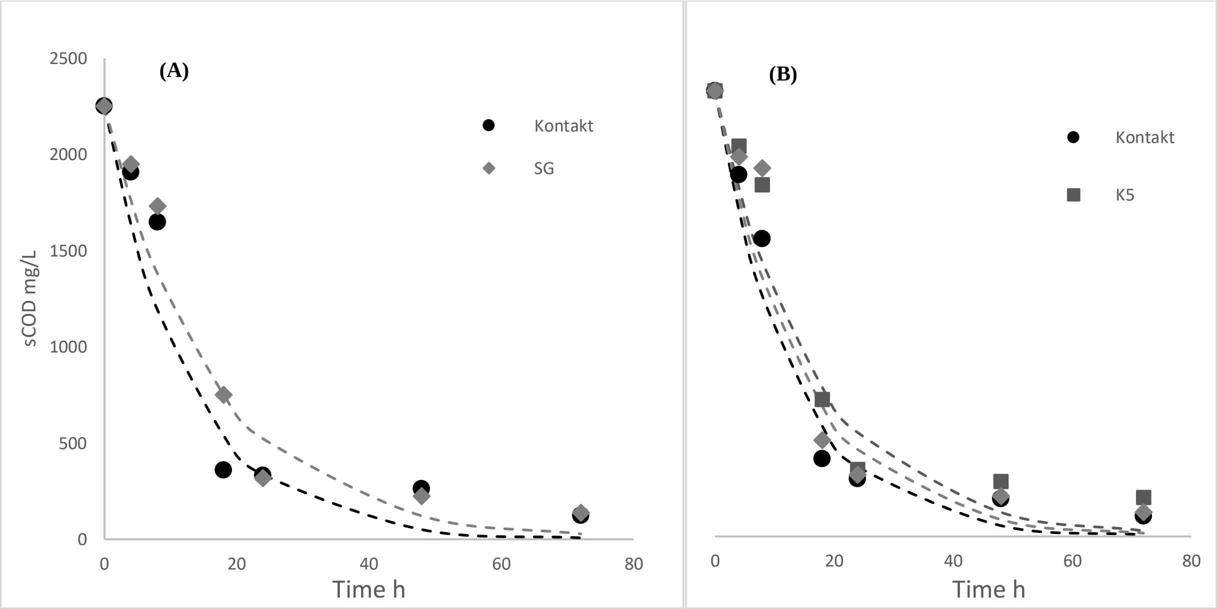

Figure 4- 9. Time series for biodegradability test for MBBR Reactors AG compares to SG: (A) MBBR 1 and (B) MBBR 2

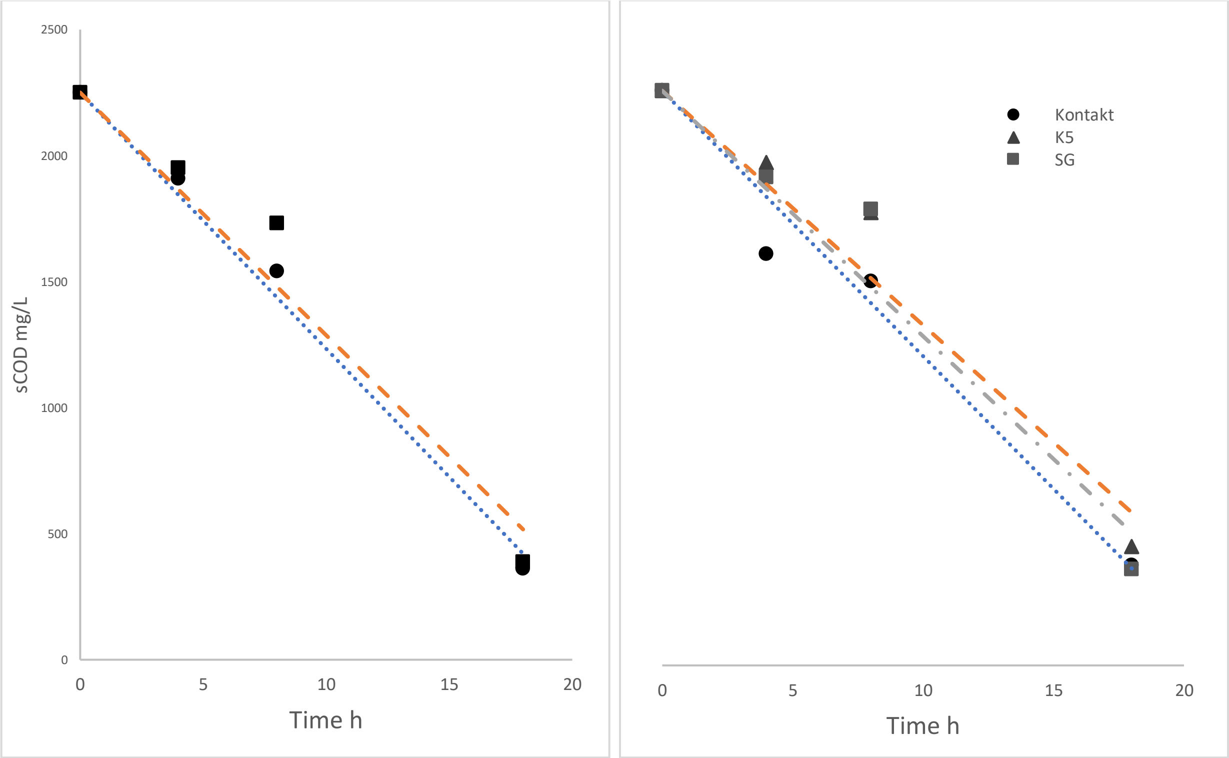

Figure 4- 10. Kinetic analysis for MBBR Reactors 1&2 AG compares to SG: (A) MBBR 1 and (B) MBBR 2

Table 4- 5

Kinetic result summary

OBJECT	EQUATION	R^2	K H^{-1}	BIOMASS IN THE JAR G
R1 KONTAKT	$C = 2250e^{-0.0908t}$	0.9067	0.0908	0.838
R1 SG	$C = 2250e^{-0.0821t}$	0.8686	0.0821	0.292
R2 KONTAKT	$C = 2250e^{-0.0893t}$	0.9348	0.0893	1.136
R2 K5	$C = 2250e^{-0.0764t}$	0.8707	0.0764	1.137
R2 SG	$C = 2250e^{-0.0861t}$	0.8572	0.0861	0.300

4.4.3 Effluent Recycle

4.4.3.1 Sludge Bulking

During the operation, Beau`s wastewater treatment system suffered a bulking sludge problem, particularly in the sedimentation tank 2. The sludge from the sedimentation tank 2 was analyzed under the microscope for determination of the cause of bulking sludge. The result shown in Figure 4-11, exhibits filamentous microorganisms, indicating that the sludge bulking problem is due to filamentous bacteria. The filamentous bacteria observed in the Figure appears to be type -0041 (square to cylindrical and enclosed within a rigid sheath), which one most frequent bacterium result for filamentous sludge bulking, especially in industrial wastewaters (Guo et al., 2014).

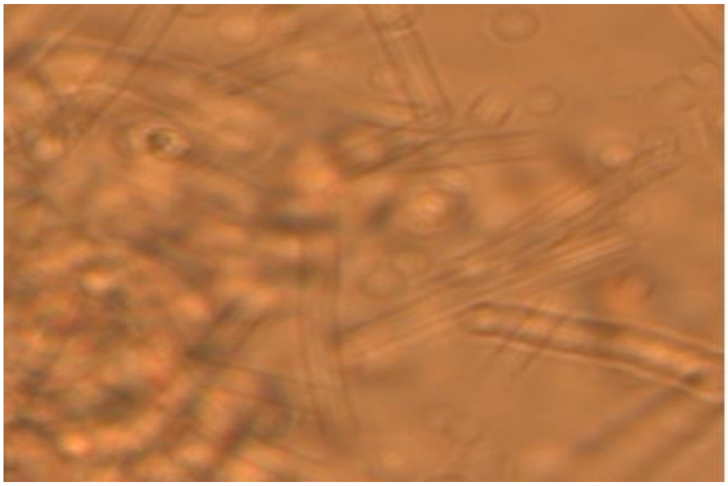

Figure 4- 11. Sludge under the VPSEM (X100 magnification)

To eliminate the bulking sludge, effluent recycling scenarios were employed to increase internal flow velocities, wash out the bulking sludge and at the same time promote the development of the carrier biofilms. The bulking sludge was effectively eliminated with the first recycle scenario (400% recycle of MBBR 2 settling tank to MBBR2 inlet) and there were no further bulking sludge problems observed during the second recycle scenario (100% effluent recycle from MBBR 2 settling tank to MBBR 1 inlet).

4.4.3.2 Organic removal efficiency

As previously shown in the Figure 4-4, there was a significant drop in the effluent sCOD concentration from MBBR2, which is one of main goals of this study. Comparing three different operating conditions at similar average SALR, No Recycle (Spring 19), S1, and S2 MBBR2 sCOD concentration, there were significant differences ($p<0.05$) observed between the No recycle period and both of the recycle conditions. During the No recycle period, the effluent concentration of sCOD was 358± 264 mg/L compared with 110 ± 59 mg/L at S1, and 165 ± 90 mg/L at S2. These results indicate that implementing effluent recycle increased the performance of the MBBR system, possibly through increased internal flow velocities and reduced HRTs shifting microbial degradation from SG to AG mechanisms. No differences in sCOD between the two recycle conditions were observed.

4.4.3.3 MLSS

There were two different recycle strategies applied to the system (Table 4-2) with MLSS concentrations presented in Figure 4-12. Significant differences ($p<0.05$) in MLSS concentration were observed between the No recycle condition and both S1 and S2 for MBBR2 for SALR range from 0-5 g sCOD/m^{2}·d. Significant difference ($p<0.05$) was also observed between No recycle and S2 for MBBR1 at a range of 5-10 sCOD/m^{2}·d, while no significant difference was observed between No Recycle and S1 as expected, as there was no recycle around MBBR 1 under the S1 condition. These results indicate that the recycle strategy was effective at reducing MLSS and thus sludge production in the system.

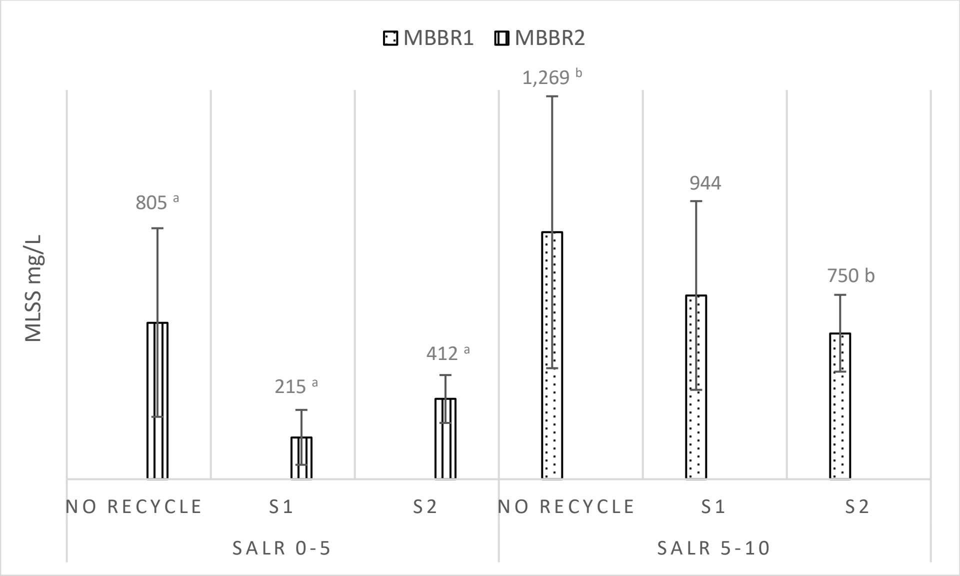

Figure 4- 12. MLSS changes during the different recycle strategy

[a]. for SALR 0-5 g sCOD/ m^{2}·d, MBBR2, no recycle: n=95, SALR= 1.6 ± 1.3 g sCOD/ m^{2}·d, S1: n=24, SALR= 3.1 ± 1.0 g sCOD/ m^{2}·d and S2: n=5, SALR= 3.01 ± 1.0 g sCOD/ m^{2}·d. For both of operation condition DO level higher than 1 mg/L. Three operating stagey presented as statistically significant ($p<0.05$). There were no enough data for MBBR1 at this range;

[b]. for SALR 5-10 g sCOD/ m^{2}·d, MBBR1, no recycle: n=18, SALR= 7.00 ± 1.3 g sCOD/ m^{2}·d and S2: n=8, SALR= 7.23 ± 1.77 g sCOD/ m^{2}·d. For both of operation condition DO level higher than 1 mg/L. These two operating stagey presented as statistically significant ($p<0.05$). There were no enough data for MBBR2 at this range;

4.4.3.4 Carrier biofilm characteristics

Ideally, most of the microorganisms in the MBBR reactor are attached on the carriers. Application of recirculation can potentially promote carrier biofilm attachment. Carrier biofilm mass and thickness were measured from the end of the No Recycle period (Day 0) through S1 and S2 conditions with data presented in Figure 4-11 with corresponding SALR. During S1, biofilm mass in MBBR1 remained quite stable until around Day 140, where a process upset spiked the pH of the influent to almost 14, killed most of the biofilm on the carriers. Interestingly, the biofilm recovered very quickly with an unexpectedly higher biomass grown compared to the earlier period at similar SALR. Biofilm mass on carriers in MBBR2 were more variable than MBBR1, showed no trend over time, and were less influenced by the pH spike than MBBR1. In a biweekly period, the SALR range varied with no observed relationship between biomass and SALR in either reactor. Furthermore, the SALR is not consistent, therefore, it was no possible to compare operating conditions on the AG. However, according to the Figure 4-13 &14, while no change in mass was observed during the S1, a significant decrease in carrier thickness over time was observed indicating that the biofilm on the carriers became denser with time. In summary, no changes in biofilm mass were observed due to recycle, while a decrease in biofilm thickness during S1 indicates the development of a denser biofilm.

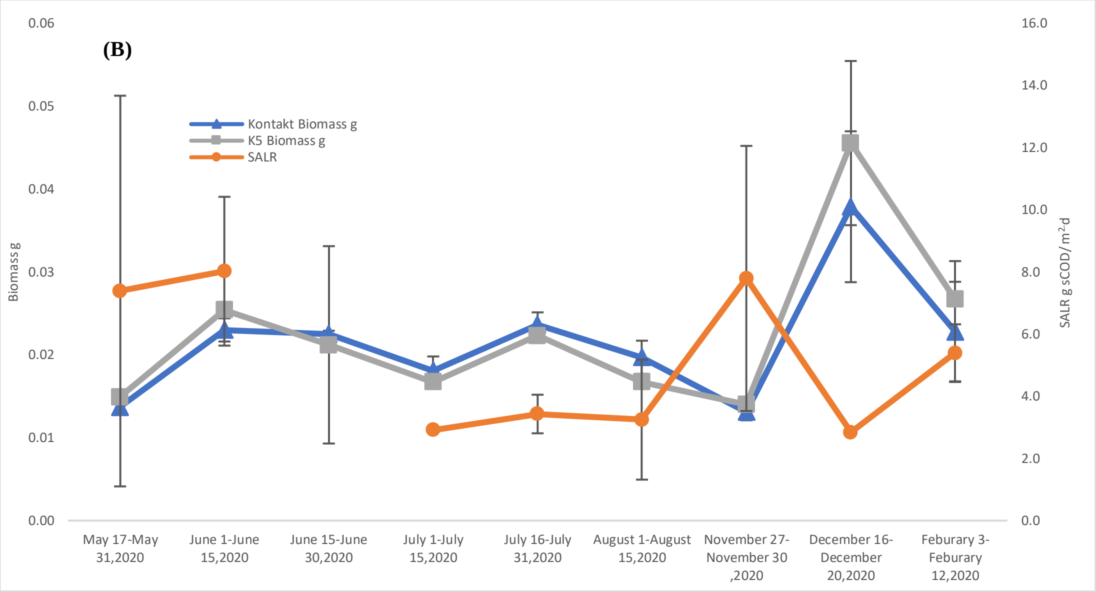

Figure 4- 13. Carrier biomass and SALR changes during the recycle: (A)MBBR1, and (B) MBBR2

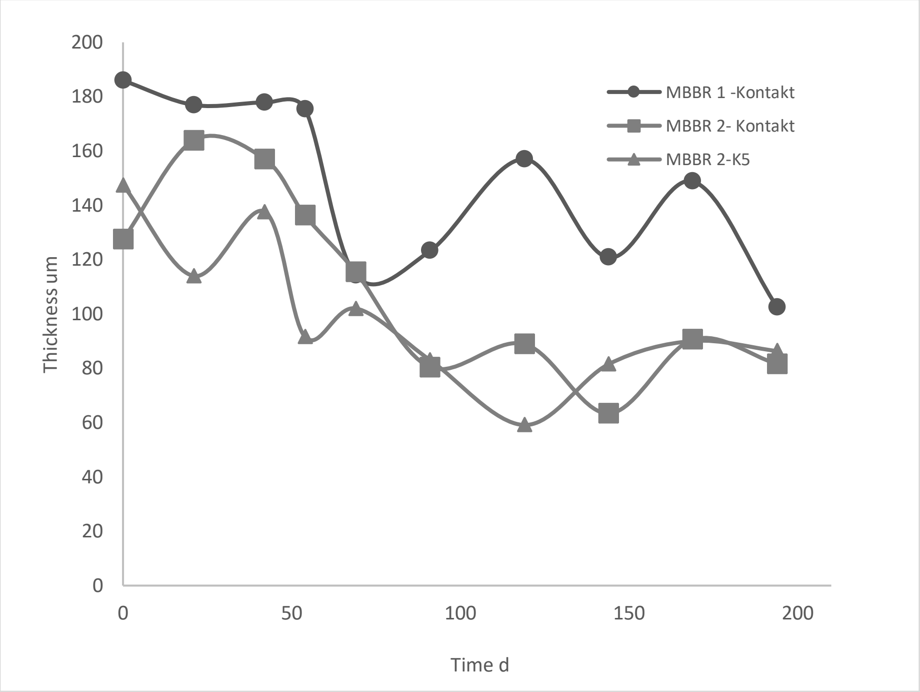

Figure 4- 14. Carrier thickness changes during the recycle

4.5 Conclusion

In this research, an on-site MBBR treatment system at craft brewery was monitored for evaluation and optimization purposes. The wastewater quality was monitored from October 12th, 2018 to February 10th, 2020, and flow rate from December 3rd, 2018 to February 10th, 2020. The observations found during the period are presented below:

1. The monitoring result present that the wastewater produced from the brewery is highly variable in terms of both organic and hydraulic loading, resulting in unstable treatment. During the period, after consideration, the actual flow equalization tank is not enough for reducing the shock to the system, especially after changing the bottle washing line from directly dispose to the sewer system to discharge to the on-site treatment system.
2. Hydraulic loading was shown to be significantly higher on weekdays compared to weekends, while no average differences in hourly loading during weekdays was observed. A 60 m^3 balancing tank is recommended to balance daily flows.
3. The operating parameters SALR and DO were shown that have a significant effect on organic matter removal efficiency. Consistent removal efficiencies of 71 and 65% were observed at low SALR (1-9.9 g sCOD/ $m^{2}\cdot d$) for both MBBRs, while removal efficiency increased from 69 to 79% from MBBR1 to MBBR2 at medium SALR (10-24.9 g sCOD/ $m^{2}\cdot d$), likely due to oxygen limitations in MBBR1. A reduction in treatment efficiency was observed at high SALR (>25 g sCOD/ $m^{2}\cdot d$) in MBBR1. For both MBBR reactors at SALR range from 5 to 10 g sCOD/ $m^{2}\cdot d$, DO concentrations should be maintain higher than 1 mg/L for achieve a higher organic removal rate. For SALR range from 0-5 sCOD/ $m^{2}\cdot d$, MBBR1 DO level suggested to be higher than 2 mg/L for achieving favorable organic removal efficiency. At the temperature range between 20-30 °C, temperature was not a decisive parameter for the organic removal rate at all SALR range studied (0-40 g sCOD/ $m^{2}\cdot d$).
4. Laboratory batch experiments conducted with both suspended growth and attached growth biomass from both reactors confirmed first order carbon degradation with similar kinetic rates varying from 0.0764 to 0.0908 h^{-1}; however, much higher attached growth biomass compared to suspended growth suggests that not all the biofilm were active for organic removal.

5. Sludge bulking occurred during the study due to the development of filamentous bacteria type-0041. Effluent recycling was tested to address the sludge bulking issue and to potentially improve treatment processes. Both recycle scenario, the MBBR2 effluent sCOD concentration was reduced to below the discharge limit and MLSS concentrations in both MBBR1 and MBBR2 were significantly lower than under no recycle conditions. This indicates that increasing internal flowrates through effluent recycling reduces MLSS and potentially promotes the development of biofilm on the carriers. Biofilm density was shown to increase with effluent recycling.

4.6 Reference

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Chapter 5

Conclusions and recommendations

5.1 Conclusions

In this research, a batch and field study conducted for improving brewery wastewater treatment efficiency. In batch experiments, high strength brewery wastewater was co-digested with dairy manure for biomethane production by anaerobic digestion to obtain optimum conditions. In-field research, an on-site MBBR treatment system at a craft brewery, was monitored for evaluation and optimization purposes. The wastewater quality was monitored from October 12th, 2018 to February 10th, 2020, and flow rate from December 3rd, 2018 to February 10th, 2020. The main conclusions were drawn from this work and are listed as follows:

- High strength brewery wastewater was shown to have a high degree of treatability under the anaerobic condition at all temperature conditions with 82-91% removal of VS, and 77-88% removal of sCOD observed.
- The psychrophilic digestion rate constant was significantly lower than both the mesophilic and thermophilic rate constants by roughly a factor of two, while the thermophilic regime was highly variable, suggesting process instability.
- An optimum organic load of 5 g VS/L and inoculum and substrate ratio of 1 were observed with both lower and higher concentrations found to inhibit the AD process.
- Significantly lower stabilized methane content was observed from psychrophilic conditions at 47% compared with both mesophilic and thermophilic conditions at 65 and 67%, respectively.
- Central composite design models were developed to describe CH_4 yield (BMP) and Rmax as functions of temperature and organic loading. The operational condition of biomethane production was also optimized with RSM and a desirability function. Optimized operational parameters were achieved at 49 °C and BWC 4.9 g/L. Correspondingly, the BMP and Rmax were 141.40 mL CH_4/g VS added and 36.90 mL CH_4/ day, respectively. However, by pursuing a stability the preferable operational condition T=35°C and BWC=5

g/L is recommended, at this condition methane yield is 110.07 CH_4/g VS added and maximum methane daily production is 28.06 CH_4/ day, which is similar to the maximum result.

- The monitoring result present that the wastewater produced from the brewery is highly variable in terms of both organic and hydraulic loading, resulting in unstable treatment.
- Hydraulic loading was shown to be significantly higher on weekdays compared to weekends, while no average differences in hourly loading during weekdays was observed. A 60 m^3 balancing tank is recommended to balance daily flows.
- Consistent removal efficiencies of 71 and 65% were observed at low SALR (1-9.9 g sCOD/ $m^{2\cdot}$d) for both MBBRs, while removal efficiency increased from 69 to 79% from MBBR1 to MBBR2 at medium SALR (10-24.9 g sCOD/ m2.d), likely due to oxygen limitations in MBBR1. A reduction in treatment efficiency was observed at high SALR (>25 g sCOD/ m2.d) in MBBR1 due to oxygen limitation.
- For both MBBR reactors at SALR range from 5 to 10 sCOD/ $m^{2\cdot}$d, DO concentrations should be maintain higher than 1 mg/L for achieve a higher organic removal rate. For SALR range from 0-5 g sCOD/ $m^{2\cdot}$d, MBBR1 DO level suggested to be higher than 2 mg/L for achieving favorable organic removal efficiency.
- At the temperature range between 20-30 °C, temperature was not a decisive parameter for the organic removal rate at all SALR range (0-40 g sCOD/ $m^{2\cdot}$d).
- A design SALR of < 25 g sCOD/ $m^{2\cdot}$d is appropriate for small brewery operations with highly variable loading conditions with DO concentrations maintained above 2 mg/L.
- Laboratory batch experiments conducted with both suspended growth and attached growth biomass from both reactors confirmed first order carbon degradation with similar kinetic rates varying from 0.0764 to 0.0908 h^{-1}; however, much higher attached growth biomass compared to suspended growth suggests that not all the biofilm was active for organic removal.
- Sludge bulking occurred during the study due to the development of filamentous bacteria type-0041. Effluent recycling was tested to address the sludge bulking issue and to potentially improve treatment processes. Both recycle scenario, the MBBR2 effluent sCOD concentration was reduced to below the designated level and MLSS concentrations in both MBBR1 and MBBR2 were significantly lower than under no recycle conditions. This

indicates that increasing internal flowrates through effluent recycling reduces MLSS and potentially promotes the development of biofilm on the carriers. Biofilm density was shown to increase with effluent recycling.

5.2 Recommendations

The following recommendations are intended to assist research working on further use anaerobic digestion to treat the high strength brewery wastewater and actual operation of the on-site treatment plant of the Beau`s brewery.

- The other operating conditions for anaerobic digestion can be investigate for optimizing BMP, such as pH and nutrients concentration.
- Conduct a pilot-scale experiment for obtaining values for build a full-scale anaerobic treatment system.
- Optimize effluent recycle and study the effects on both suspended growth and attached growth biomass.