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Anaerobic Hydrogen and Methane Production from Dairy Processing Waste: Experiment and Modeling

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ANAEROBIC HYDROGEN AND METHANE PRODUCTION FROM DAIRY PROCESSING WASTE: EXPERIMENT AND MODELING

by

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ABSTRACT

Anaerobic Hydrogen and Methane Production from Dairy Processing Waste: Experiment and Modeling

by

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Utah State University, 2016

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Dairy processing waste (DPW) can cause many environmental problems if not treated well. Various wastewater treatment technologies have been applied to reduce the organics and inorganics in DPW. The overall objective of this research was to develop cost effective anaerobic digestion technology for hydrogen and methane production from DPW. This search included three phases of studies.

In phase 1, we investigated continuous fermentations of algae, lawn grass clippings and DPW, commingled and digested in duplicate 60 L and 3,800 L Induced Bed Reactor (IBR) anaerobic digesters at mesophilic conditions in trials that went for about two years. The goal was to commingle municipal waste in such a way that no pH control chemicals would be required. The research also yielded information about solids loading rate (SLR), efficiency of chemical oxygen demand (COD) and solids removal and biogas

production. Under the conditions of the study, commingling algae or grass with DPW made it possible to avoid the addition of pH control chemicals.

In phase 2, we investigated the effects of pH, temperature, and hydraulic retention time (HRT) and organic loading rate (OLR) on hydrogen production from DPW in semi-continuous 60 L pilot IBR. Results show pH played a key role on hydrogen production and the optimal pH range was 4.8-5.5. Digestion under thermophilic temperatures (60 °C) had advantages of gaining higher hydrogen yield and suppressing the growth of methanogens. The optimal OLR was 32.9 g-COD/l-d at HRT of 3 days. Under optimal conditions, highest hydrogen yield was 160.7 ml/g-$COD_{removed}$ with 44.6% COD removal.

In phase 3, a mathematic model was built and implemented in R based on Anaerobic Digestion Model No. 1 (ADM1) for predicting and describing the anaerobic hydrogen production process. The modified ADM1 was then validated by comparing the predictions with observations of anaerobic hydrogen production from dairy processing waste. The model successfully predicted hydrogen production, hydrogen content, methane content, VFA concentration, and digestion system stability. This study provides a useful mathematical model to investigate anaerobic hydrogen production process and stability.

(158 pages)

PUBLIC ABSTRACT

Anaerobic Hydrogen Production from Dairy Processing Wastes: Experiment and Modeling

Jianming Zhong

Dairy processing waste (DPW) is the waste produced from manufacturing dairy products: cheese, yogurt, ice cream, milk, butter, etc. DPW is high in chemical oxygen demand (COD) due to its lactose, fat and protein content, and therefore needs to be appropriately treated. An investigation was conducted to produce energy (hydrogen and methane) from DPW by anaerobic digestion. This project developed an effective engineering method for stable methane production from DPW without adding pH control chemicals. This study also explored the optimal operational condition for hydrogen production from DPW. We further built a mathematical model to help us monitor and predict anaerobic hydrogen production process. The achievements in this study will help dairy or other food industries to not only manage their waste but also make sufficient energy to supply their production plants.

(158 pages)

DEDICATION

I dedicate my dissertation to my parents-Jinfeng Wang and Wenxue Zhong. It was not easy for them to raise me up and support me in school for over 20 years. You showed and taught me how to be a good person. I also want to dedicate this to my wife Ying Lu and my son Luke Zhong; you are my motivation of finishing this dissertation.

Jianming Zhong

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Jianming Zhong

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LIST OF ABBREVIATIONS

DPW = Dairy processing waste

IBR = Induced bed reactor

COD = Chemical oxygen demand

HRT = Hydraulic retention time

SRT = Solids retention time

AD = Anaerobic digestion

UASB = Upflow anaerobic sludge blanket

ADM1 = Anaerobic digestion model No.1

VFA = Volatile fatty acids

ORL = Organic loading rate

HOAc = Acetic acid

TOC = Total organic carbon

TN = Total nitrogen

TS = Total solids

VS = Volatile solids

VSS = Volatile suspended solids

SATP = Standard ambient temperature and pressure

CHAPTER 1

INTRODUCTION

The Energy Information Administration reported in 2011 that about 80.2% of the primary energy consumption in the world was from fossil fuels, which consisted of 35.3% petroleum, 19.7% coal and 24.8% natural gas (EIA, 2011). Within the past decade, researchers have paid more and more attention to the development of renewable and clean energy sources. Reasons for the great interest in this area are: (1) increasing prices of fossil fuels, and (2) climatic changes or environmental issues (Panwar et al., 2011).

Hydrogen is considered an alternative fuel of great potential. Hydrogen is environmentally friendly because only water is produced when it is combusted. It was identified as a clean energy carrier for the future at the first World Hydrogen Conference (Lattin and Utgikar, 2007). Hydrogen has the highest energy content per unit mass among all commonly used fuels. It is 2.6 times higher than methane and 3.3 times higher than gasoline (Table 1.1). Hydrogen has great potential to reduce the use of fossil fuels. However, the majority of hydrogen is produced from fossil-fuel sources natural gas, oil, and coal. Figure 1.1 illustrates the worldwide distribution of hydrogen production sources. More renewable and economical production methods are required before a sustainable hydrogen economy can be established.

Dairy processing waste (DPW) is the waste produced from manufacturing dairy products: cheese, yogurt, ice cream, milk, butter, etc. DPW is high in chemical oxygen demand (COD) due to its lactose and protein content, therefore, needs to be appropriately treated. The discharge of DPW, such as cheese whey, onto land can damage

the chemical and physical structure of soil, pollute groundwater (Ben-Hassan and Ghaly, 1994) and may also affect air quality (Bullock et al., 1995). Now there are more and more whey protein concentrate and isolate products, which has reduced DPW (Whetstine et al., 2005). However, finding a cost-effective disposal or utilization technology for waste has been an important issue for the dairy industry because of:

1. High lactose content in DPW;
2. High investment cost in whey protein processing equipment;
3. Increased volume of dairy processing byproducts;
4. Increasingly strict legislative requirements.

Anaerobic digestion (AD) is a process of converting organic materials into oxidized end products, mostly carbon dioxide, methane, and new bacterial mass under anoxic condition. AD is also a potential technology for both hydrogen production and food waste management. Anaerobic digesters can produce hydrogen from inexpensive and renewable energy sources such as food processing waste. Recent research proved that certain strains of bacteria (e.g., bacteria from the genus *Clostridium*) are particularly effective at producing hydrogen as a by-product during anaerobic digestion of organic waste material (Zhang et al., 2006).

Although various studies have been done on hydrogen anaerobic digestion, there are still several obstacles that must be overcome before applying this technology economically at an industrial level. These problems may include: feedback inhibition such as volatile fatty acids (VFA) and partial hydrogen pressure, digester's low buffering

capacity resulting in expensive chemical usage for pH control, high energy input, etc. Furthermore, a mathematical model is needed to examine the inhibition factors and improve the hydrogen production process.

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Table 1.1-Energy Density Values of Common Fuels (Lattin and Utgikar, 2007).

Fuel sources	Phase*	Energy Density(MJ/kg)*	Density(Kg/m3)	Energy Content(GJ/m3)*
Hydrogen	gas	143	0.0898	0.0128
Methane	gas	54	0.7167	0.0387
Ethanol	liquid	29.6	794	23.5
Gasoline	liquid	44	740	32.6
No. 2 Diesel	liquid	46	850	39.1
Coal	liquid	35	800	28

*: Values were measured at 25°C and 105 kPa

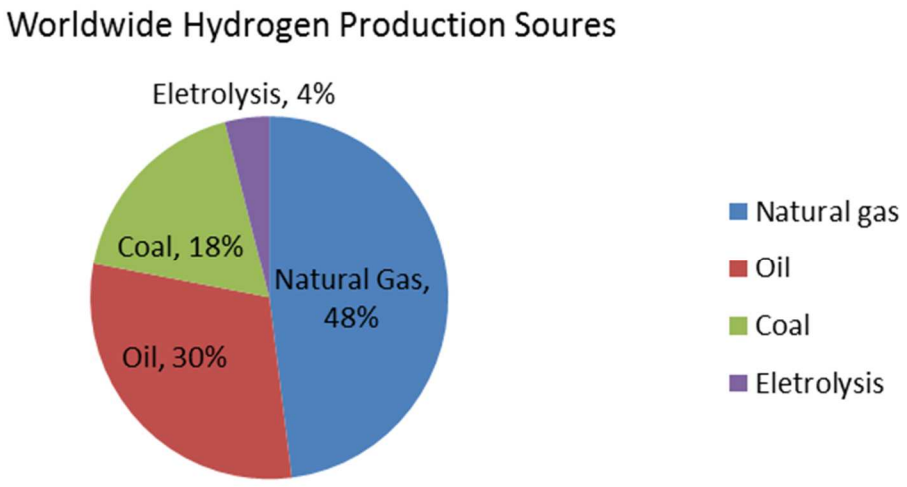

Figure 1.1-Worldwide hydrogen production sources (Bockris, 2003)

CHAPTER 2

OBJECTIVES

The overall objective was to develop cost effective anaerobic digestion technology for hydrogen and methane production from DPW. Specific objectives and sub-objectives are listed below:

Objective 1: Experiment

Build a hydrogen anaerobic digestion system and optimize at pilot scale first, then apply it to large-scale digesters.

a. Determine characteristics of DPW. Gather chemical and physical characteristics data of DPW. The measured characteristics include pH, COD, total solids (TS), alkalinity, volatile suspended solids (VSS), total organic carbonate (TOC), total nitrogen (TN), and total fatty acid (VFA).
b. Install and run two 60 L pilot Induced Bed Reactors. Design the digesters and send them to a manufacturer for construction. Install pumps, pH controllers, temperature controls, mass flow rate storage system, etc. Run these two pilot digesters for biogas production to test their performance.
c. Inoculum. Pretreat the sludge from an anaerobic digester with heat and low pH to enrich the hydrogen-producing bacteria and inhibit the hydrogen- consuming bacteria.
d. Run the digestions to find optimal parameters for hydrogen production. The

tested variables are pH, temperature, and HRT.

e. Measure the effluent characteristics. The measured characteristics include pH, COD, TS, alkalinity, VSS, TOC, TN, and total VFA.

f. Test the performance of two-stage digestion. The aim was to produce energy (methane) from hydrogen digestion effluents that still have a high level of COD. The second stage may also provide the effluent buffer that may potentially be recycled in the first stage of hydrogen digestion.

g. Perform hydrogen and methane anaerobic digestion in 3,800 L IBRs using the results found in the pilot-scale study

Objective 2: Model Development

Develop a mathematical model to describe and predict hydrogen anaerobic production from DWP.

a. Implement the anaerobic digestion model No. 1 (ADM1) in R software. Write the R codes to describe all of the processes and mathematical dynamic equations that are described in the ADM1 model.

b. Test the performance of ADM1 in R using the pilot anaerobic digestion data to check the sensitivity and accuracy of running ADM1 in R.

c. Build dynamic equations of specific inhibition factors that play important roles in hydrogen anaerobic digestion. Those factors may include volatile fatty acid, pH, hydrogen partial pressure, etc.

d. Modify the processes in ADM1 to make it specific for hydrogen production

rather than methane production.

e. Test the model by comparing model prediction values to the experiment data from both pilot and full-scale data obtained in objectives 1.

CHAPTER 3

LITERATURE REVIEW

Hydrogen

Characteristics of Hydrogen

Hydrogen gas is an odorless, colorless and non-poisonous gas with extremely low density. Among all the gases, hydrogen gas is the lightest. Even liquid hydrogen has only 76.3 Kg/m^3 density at its melting point. Molecular hydrogen has a melting point of -259.14 °C and a boiling point of -252.87 °C. The low boiling point means a lot of energy is required to obtain liquid or solid hydrogen. Hydrogen gas can burn in the range from 4% to 74% by volume in air and thus is highly flammable (Carcassi and Fineschi, 2005). The enthalpy value of hydrogen combustion is −286 kJ/mole (energy density -143.0 MJ/kg).

$$2\,H_2\,(g)\,+\,O_2\,(g)\,\rightarrow\,2\,H_2O\,(l)\,+\,572\,kJ\,(286\,kJ/mole) \quad (1)$$

As shown in Table 1, hydrogen has the highest energy density (per mass unit) among all commonly used fuels, which is 2.6 times higher than methane and 3.3 times higher than gasoline. However, due to the extremely low density, the energy content of hydrogen per unit volume is significantly less than that of traditional fuel sources, 3 times lower than methane and 2,547 times lower than gasoline, although the energy density per mass unit is higher. Therefore, efficient compacting and storing techniques are required for the wide application of hydrogen as an energy carrier.

Hydrogen as a Fuel

The world's reserves of major fossil energy sources such as petroleum, coal, and natural gas are limited and non-renewable. The World Energy Outlook (WEO) 2015 claims that fossil fuels continue to meet more than 80% of total primary energy demand (WEO, 2015). Moreover, the uneven distribution of these fossil fuel sources throughout the world leads to higher fuel costs because of overseas transportation (Huber, 2009).

The combustion of these fossil fuels can cause environmental problems. Over 90% of energy-related emissions are CO_2 from fossil fuels combustion. It is considered to be the largest contributing factor to the release of greenhouse gases into the atmosphere (WEO, 2015). In addition to carbon dioxide, fossil fuel combustion also releases nitrogen oxides, carbon monoxide and sulfur oxides. These gases are not only harmful to human health, but they also contribute to form small particles which cause serious air pollution problems (Hill et al., 2009). Today many alternative clean and renewable energy sources are proposed and applied, such as hydrogen, which produces no greenhouse gases and releases a large amount of energy when burned. It is considered one of the most promising alternative clean energy sources in the future (Gupta, 2008).

Hydrogen Applications

Today the majority of hydrogen is used as a feedstock in industry (Edwards et al., 2008). In the fertilizer industry hydrogen is used as a feedstock to produce ammonia. In the petrochemical industry hydrogen plays a role in the cracking and hydrogenation of hydrocarbons and the removal of sulfur, nitrogen, oxygen, and metals for gasoline, diesel,

and other petroleum products' production. In the food industry hydrogen is added as a hydrogenating agent in the process of solidification of oil and fat. In the chemical industry hydrogen is added in the production of many chemicals (e.g. methanol, acetic acid, butanediol, and benzene). In the metallurgical industry hydrogen is used as an oxygen scavenger, and in the mechanical industry hydrogen is used as a shielding gas in welding. Also, a small amount of hydrogen is used as an energy carrier, mainly in the space exploration industry as a rocket fuel. Additionally, hydrogen has potential application in the future as a feedstock used in fuel cell technology in vehicles, in electricity production and in other areas when new technologies are being developed (Edwards et al., 2008).

Hydrogen Production Methods

In industry, the majority of hydrogen is produced from fossil-fuel sources natural gas, oil, and coal. Figure 1.1 illustrates the worldwide distribution of hydrogen production sources. 48% of global hydrogen is produced from steam reformation of natural gas (mainly methane), 30% is from coal, 18% from oil and 4% from water electrolysis (Although various studies have been done on hydrogen anaerobic digestion, there are still several obstacles that must be overcome before applying this technology economically at an industrial level. These problems may include: feedback inhibition such as volatile fatty acids (VFA) and partial hydrogen pressure, digester's low buffering capacity resulting in expensive chemical usage for pH control, high energy input, etc. Furthermore, a mathematical model is needed to examine the inhibition factors and

improve the hydrogen production process..1) Bockris, 2003). The following equation represents the process of steam reformation of methane;

$$CH_4 + H_2O\,(steam) \rightarrow CO + 3\,H_2 \qquad (2)$$

At high temperature (700–1100 °C) and high pressure (2.0 MPa), methane reacts with steam to produce carbon monoxide and H_2. Fossil fuels sources are unsustainable. Water electrolysis method is clean and renewable but needs high electrical energy input. Today water electrolysis is considered a promising method only when high purity hydrogen is needed and low cost electricity is available (Zeng et al., 2010).

Besides the methods shown in Figure 1.1, there are other potential alternative methods to generate hydrogen. Hydrogen production through a biological process is considered an attractive field because it can generate hydrogen without fossil fuels. Biological hydrogen processes usually require the growth of microorganisms, the addition of substrates, and the presence of oxygen or sometimes light. Based on light dependence, two different processes are defined: dark fermentation, which is also called anaerobic process, and photosynthetic process.

Food Waste Management

Food waste is the second largest component of Municipal Solid Waste (MSW) generation in the United States. This food waste may come from kitchen wastes, left-over food, plate waste and restaurant order returns, and industrial sources such as dairy companies. In 2012, about 14.5% of total MSW by weight was food waste, more than

any other material except paper. However, less than 3% of food waste was recovered or recycled (

) (EPA, 2012). Most was thrown away and finally treated by/in landfills and incinerator.

Throwing away food waste not only wastes lots of money that people invested during food production, it also causes big environmental problems. Food waste, which consists of a high percentage of organic materials like carbohydrates, proteins, and lipids, is easily and quickly digested in the landfill. During the digestion large amounts of methane gas are produced (EPA, 2012).

Methane is a potent greenhouse gas which has 21 times more global warming potential than carbon dioxide. According to an EPA report, more than 20 percent of all human-related methane emissions are from landfill gas.

Cheese whey is the lactose rich by-product from the cheese manufacturing process. In 2006, the United States produced about 9.5 billion pounds of cheese which resulted in an estimated 84.5 billion pounds of cheese whey (FAO 2010). Cheese whey has a very high COD value (up to 70 g COD/L) because of its composition (Table 3.1). Typically, dumping large amounts of untreated cheese whey to the sewage system will lead to COD overloading for the local waste water treatment plant and damage its system. Thus, cheese whey disposal has become a major concern for cheese producers in recent years due to the larger amounts of whey generated and the more stringent legal requirements for effluent quality.

Many treatment and utilization methods for whey have been developed: utilization as animal feed directly; processing as whey protein powder for human supplement or energy foods; land application as field fertilizer; treatment by wastewater treatment systems and fermentation of whey to ethanol.

Anaerobic digestion of whey is another good approach for not only lowering the COD values but also for energy conservation. The methane produced can provide part of the energy needs of dairy plants (Malaspina et al., 1996). Although whey has sufficient organic components (mostly lactose) that are easily biodegradable and a high biogas potential level, it is rarely treated by anaerobic digestion directly due to its low pH and instability during digestion.

Microbiology and Biochemistry of Anaerobic Digestion

The primary objectives of organic wastes anaerobic digestion are COD and pathogen reduction, with concurrent biogas production. This is accomplished through biological degradation of organic substrates to carbon dioxide and methane in the absence of oxygen with the involvement of several groups of bacteria. The digestion process consists of several interdependent, complex sequential and parallel biological reactions. During these reactions, the products from one group of microorganisms serve as the substrates for the next. The overall conversion process is often described as a three-stage process which occurs simultaneously within the anaerobic digester (Young and McCarty, 1969; Lettinga et al., 1980; Switzenbaum and Jewell, 1980). The first is hydrolysis of insoluble biodegradable organic matter, the second is the production of acid from smaller

soluble organic molecules, and the third is methane generation. The three-stage scheme involving various microbial species can be described as follows: (1) hydrolysis and liquefaction; (2) acidogenesis, and (3) methane fermentation (Figure 3.2).

Hydrolysis and Liquefaction

Hydrolysis and liquefaction is a process in which complex and/or insoluble organics are converted to a simpler and soluble form that can pass through bacterial cell walls and be metabolized for use as energy or nutrient sources. Most of the constituents of the organic wastes in anaerobic digestion are insoluble and cannot be assimilated by bacteria directly. Hence, hydrolysis and liquefaction is a necessary and sometimes limiting process during digestion (Young and McCarty, 1969; Lettinga et al., 1980; Switzenbaum and Jewell, 1980; Parawira et al., 2004). This process is accomplished by multiple enzymes, such as extracellular or hydrolytic enzymes, excreted by the specific group of bacteria. In order to effect hydrolysis without limiting the overall digestion rate, the above enzymes must be produced by the bacteria in sufficient quantity and make intimate contact with organics. Thus, large amounts of active microorganisms, thorough mixing, and good bacteria-growing conditions are important during digestion. However, not all the organics break down into small molecules that can be utilized by bacteria.

Acidogenesis

Acidogenesis is a complicated process comprising acid-forming fermentation, and hydrogen and acetate formation. Acid forming fermentation: once complex organics are hydrolyzed, they are fermented to long-chain, organic acids, sugars, amino acids, and

eventually to the intermediary products (smaller organic acids) such as propionate, butyrate and ethanol (Young and McCarty, 1969; Lettinga et al., 1980; Switzenbaum and Jewell, 1980; Parawira et al., 2004). Acetic acid, carbon dioxide, and hydrogen are also produced during this process. Hydrogen inhibits the growth of many acid-forming bacteria and hence must be kept in low concentration in order to keep digestion going continuously (Das and Veziroglu, 2008). Fortunately, hydrogen is an energy source in a later methane-producing step and can be rapidly removed.

Hydrogen and acetic acid formation: in addition to the fermentative microorganism (Group 1 in Figure 3.2), hydrogen and acetate can also be produced by hydrogen-producing and acetogenic microorganisms (Group 2 in Figure 3.2). Studies show that during acidogenesis hydrogen concentration is very important in regulating organic acid production and consumption (Das and Veziroglu, 2008). Once hydrogen partial pressure is high ($>10^{-4}$ atm), methane production will be inhibited and the organic acid concentration will continuously increase. Thus, hydrogen partial pressure must be controlled closely in efficient methane production as well as hydrogen production. As stated above, this hydrogen can be rapidly removed in the later step.

Methanogenesis

Methanogenesis is the last step of anaerobic digestion, which essentially is the conversion of acetic acid, hydrogen, and carbon dioxide to methane (Young and McCarty, 1969; Lettinga et al., 1980; Switzenbaum and Jewell, 1980; Parawira et al.,

2004). The produced methane separates from the sludge to the top gas which leaves the system. At the same time, carbon dioxide is produced.

The microorganisms involved in methanogenesis are called methanogens. They are Archaea and belong to the genera *Methanosarcina, Methanococcus Methanobacterium and Methanospirillum* (Henze, 2002). Methanogens are unique archaea because they can only use certain types of nutrients as energy sources. It is reported that the only substrates they can use are acetic acid, methanol, hydrogen, and formic acid (Balch et al., 1979). Acetic acid is the main substrate for methane production during dark fermentation. Approximate two-thirds of the methane formed in the anaerobic digestion of many substrates is from the acetate conversion by acetoclastic archaea. The reaction can be simplified as:

Acetate: $CH_3COOH \rightarrow CH_4 + CO_2$ (3)

The rest of the methane is from hydrogen conversion by hydrogenophilic methanogens with the reaction:

Hydrogen: $4\,H_2 + CO_2 \rightarrow CH_4 + 2H_2O$ (4)

Anaerobic Hydrogen Production

Dark fermentation is a promising method for hydrogen production. As shown in Figure 3.2, the end products of dark fermentation are methane and carbon dioxide. Hydrogen, which can be produced in several sub-pathways in Figure 3.2, is an intermediate product and is quickly consumed during methane production. In order to transfer the whole pathway from methane production to hydrogen production, process

controls are required to block the hydrogen-consuming sub-pathways (red crosses in Figure 3.2). The key parameters that play important roles in the control of hydrogen production include organic sources, organic loading rate (OLR), inoculum, pH, temperature, hydrogen partial pressure, and hydraulic retention time (HRT) (Wang and Wan, 2009).

Substrates and Organic Loading Rate

Hydrogen dark fermentation can be fed with various inexpensive organic sources such as food waste, municipal solid waste, sewage sludge, and paper mill waste. However, some research shows that carbohydrates are the preferred substrate because of the higher hydrogen yields (mole H_2 per mass unit of COD) compared to proteins and lipids (Nath and Das, 2004). Recently, more research studies suggest that the co-digestion of mixed feedstock can increase the digestion efficiency and hydrogen yields (Fernandes et al., 2010). For example co-digestion with the mixture of food wastes and sewage wastewater shows a better performance and also increases the digester's buffering capacity (Azbar et al., 2009).

Besides the feed organic sources, the OLR is also an important parameter during process control. Usually higher organic loading is required in order to achieve high hydrogen yields. But excessive organic loading can inhibit the digestion process. Several studies demonstrated that higher hydrogen yields were obtained when feeding with a low substrate concentration (Chong, et al., 2009; Sreethawong et al. 2010 and Intanoo, et al., 2014). OLR can be controlled by the substrate's dilution/enrichment/combination and the

HRT. Optimal OLR and HRT should be determined to achieve the highest hydrogen yield, as well as maintain unfavorable conditions for hydrogen-consuming bacteria (Lin and Jo, 2003). Most studies show that maximum hydrogen yield is achieved under the following conditions for completely mixed reactors: 40 g COD/L organic substrate and 2-72 hours HRT (Lay et al., 1999; Vijayaraghavan et al., 2006; Wu and Lin, 2004).

Inoculum

Starter culture is a mixed symbiotic culture that the operator wants to modify for removing hydrogen-consuming microorganisms groups so that the remaining culture is producing primarily hydrogen as its end product. Hydrogen-producing bacteria can be selected through heat or acid/alkali treatment (Kawagoshi et al., 2005; Li and Fang, 2007). During the treatment, hydrogen-consuming microorganisms such as methanogens are inactivated or killed. In the thermal treatment process, most microorganisms are killed at 60-90° C, but some heat-resistant microorganisms can survive because of their spore-forming ability. Several hydrogen-producing bacteria are heat resistant, and therefore, can be selected under high temperature. However, it was recently reported that some methane was still produced after heat treatment (Luo et al., 2011), which may indicate the existence of heat-resistant methanogens (Venkata Mohan et al., 2008). Using the acid method, the methanogens can be removed almost completely. However, the main disadvantage of this treatment is the low efficiency of hydrogen production (Vijayaraghavan et al., 2006). Recently, more researchers are using a method of combining heat and acid treatment, which has better performance in both methanogen

removal and hydrogen production (Wang and Wan, 2008).

Both mixed culture and pure culture can be used as seed inocula. The pure culture such as *Clostridum* shows better performance in hydrogen production when using specific feedstocks like glucose and other carbohydrates. To obtain higher hydrogen yield and overcome limitations during the process, genetic modifications have been made on several microorganisms, such as *Costridum acetobuylicum* and *Escherichia coli*. These modifications may include the overexpression of the hydrogenase gene and/or the inhibition of other pathways to push the metabolism towards the hydrogen production pathway (Lay et al., 2010; Vijayaraghavan et al., 2006). However, these genetic modifications on microorganisms may have very limited application in hydrogen production (Oh et al., 2003; Venkata Mohan et al., 2008). Because such modified culture usually needs sterilized substrate/feedstock to survive, in real application complex substrates such as wastewater sludge are often used to produce hydrogen. Sterilization of the feedstock will significantly increase the cost of the hydrogen fermentation. Therefore, the mixed culture which is more commonly used has several advantages such as higher efficiency and easy control.

pH

The pH of the digestion environment has a crucial effect on both hydrogen yield and the hydrogen production rate. Different microorganisms have various specific pH ranges for growth. Under a certain pH of digestion, the H^+ in the extracellular environment selects the bacteria that can survive at this pH range, and at the same time

suppresses or kills other organisms that cannot grow (Fang and Liu, 2002; Temudo et al., 2007). The reported optimal pH for the hydrogen production is around 5.5. It varies from 4.7 to 6.0 under different substrates, microbial groups, and operational conditions. At low pH (<6), the activity of methanogen with an optimal pH range of 7.0-7.5 is greatly inhibited. However, inhibition of the methanogens is not enough to eliminate the hydrogen-consuming organisms (Horiuchi et al., 2002). Some homoacetogenic bacteria can grow or survive in a broad pH range of 4-8. Therefore, the method of simply adjusting pH is sometimes not enough to stop the hydrogen-consuming process.

With hydrogen production, volatile fatty acids (VFA) such as acetate and butyrate are produced continuously. The VFA will lower the pH of the digester, especially when the digester has low alkalinity. Thus, when the digester has high ORL and low HRT, increasing the pH buffering is important, especially in continuously hydrogen-producing tanks (Chong et al., 2009; Zoetemeyer et al., 1982). Thus, pH buffer addition should be considered, for making hydrogen in continuous hydrogen production tanks, especially when the digester has high ORL and low HRT. The addition of chemical reagents is one option to increase the buffer capacity and control the pH. Lin et al. (Lin and Jo, 2003) found that in a batch reactor, adding phosphate can increase the buffer capacity and hydrogen yield. Another possible way is to use the co-digestion of high alkalinity feedstock such as sewage sludge. However, more research is needed to find inexpensive buffers that can be used in hydrogen dark fermentation.

Temperature

Most hydrogen dark fermentation takes place under mesophilic (25 - 40 °C) or thermophilic (40 - 65 °C) conditions; few studies have been done under extreme thermophilic (65 - 80 °C) conditions (Wang and wan, 2009). Increasing the temperature typically can enhance the activity of the enzymes until the optimal temperature is reached. On one hand, hydrogen production above 60°C has several advantages, such as high hydrogen yield, increased the solubility of some polymeric substrates, and inhibition of the growth of methanogens (de Vrije et al., 2009; Egorova and Antranikian, 2005). High-temperature fermentation is used widely for some biomass containing substances that are difficult to hydrolyze, e.g., lignocelluloses. On the other hand, fermentation at high temperature means more energy input (Ivanova et al., 2009).

Hydrogen Partial Pressure

Hydrogen partial pressure inside the digester has a negative effect on fermentation through feedback inhibition on the microbial hydrogen production process by maintaining high hydrogen concentrations in the liquid phase. Moreover, high hydrogen partial pressure not only affects hydrogen production but also triggers a shift of metabolic pathways towards the accumulation of acetate, ethanol, acetone and butanol (Adams, 1990; Angenent et al., 2004; Chou et al., 2008).

Recently, some studies have attempted to decrease the hydrogen partial pressure inside the digester. Increasing the agitation speed is an effective method. Research Chou et al. (2008) showed that the hydrogen yield increased three times when the stirring speed increased from 20 rpm to 100 rpm. Another method to improve gas extraction is gas

sparging, with nitrogen or argon as the sparging gas (Logan et al., 2002; Rodríguez et al., 2006). However, these two methods increase the production costs for agitation or purification of the biogas. Further research is needed to develop an efficient and inexpensive gas extraction system for industrial application (Batstone et al., 2006; Mizuno et al., 2000; Veeken and Hamelers, 1999).

Anaerobic Digestion Model

Anaerobic Hydrogen Model

The technology of anaerobic digestion for biogas production was established a long time ago and is now widely applied. But the process is not fully understood due to the complexity of microbial metabolism. An example is hydrogen anaerobic digestion, which is a promising method to produce hydrogen economically. But it faces several problems due to limited understanding of its microbial metabolism. A good mathematical model is needed to analyze and further understand the microbial metabolism process, especially hydrogen anaerobic digestion which is very attractive for future hydrogen production but faces several limitations at present. A few models have been developed to describe the hydrogen production process, but all are limited in scope. The Monod model was used to describe the relationship between the organic substrate degradation rate and the growth rate of hydrogen-producing bacteria (Kumar et al., 2000). The Andrew model is usually used to show the impacts of pH on the specific hydrogen production rate, although it is sometimes used to describe the effects of temperature on the hydrogen production process (Majizat et al., 1997; Mu et al., 2006; Nath et al., 2008; Zheng et al.,

2008). A modified Gompertz model was specifically developed to examine the batch hydrogen fermentation process (Lay et al., 1999; Wu and Lin, 2004). The Luedeking–Piret model and its modified version were developed to describe the correlation between hydrogen production rate and the growth rate of hydrogen-producing bacteria (Lo et al., 2008; Mantis et al., 2005). However, none of the above models describes the whole process of hydrogen production and the effects of inhibition factors, such as hydrogen partial pressure and fatty acid concentration.

ADM1 Model

Since the International Water Association (IWA) in 2002 developed the anaerobic digestion model No. 1(ADM1), this model has attracted wide attention in the field of research and practical application of anaerobic digestion (Batstone, et al., 2002). ADM1 is a mathematical model that is often used as a framework model that investigators can modify and choose coefficients according to their specific substrates and digester. The reactions occurring in anaerobic digestion are very complex. They have many sequential and parallel steps. ADM1 divides those reactions into two main types during model development: biochemical reactions and physicochemical reactions.

Biochemical reactions. Microorganisms play the key role in this process. ADM1 starts the biochemical reactions at disintegration; that is, the conversion of organic materials to carbohydrates, proteins and lipids, and the hydrolysis of these particles to sugars, amino acids, and long-chain fatty acids. This process is treated as first order kinetics and is the rate-limiting step in the model development. Acidogenesis and

methanogenesis are also included in the model. Implemented as a differential equation system, the model describes 19 processes and 24 components (Figure 3.3).

Physicochemical reactions. The model also describes gas-liquid transfer and ion association and dissociation. An additional reaction, not included in the ADM1 is precipitation. During ADM1 development, the concentration of free ammonia, hydrogen, inorganic nitrogen, as well as pH, are considered as inhibitors in some processes.

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Table 3.1-Typical composition of dry sweet and acid whey (Posati and Orr, 1976)

Whey type	Fat %	Protein %	Lactose %
Sweet cheese whey	1.1	12.9	74.4
Acid cheese whey	0.5	11.7	70.0

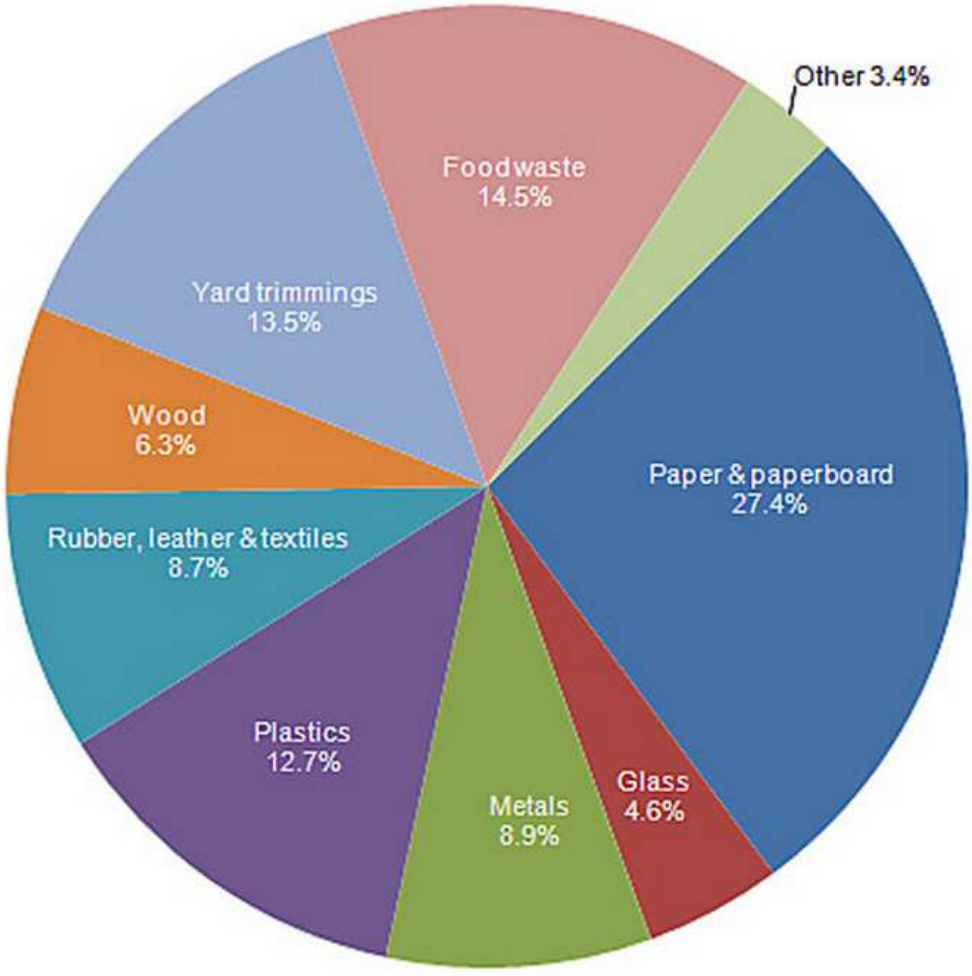

Figure 3.1-2012 U.S. total MSW Generation by Material (EPA, 2012)

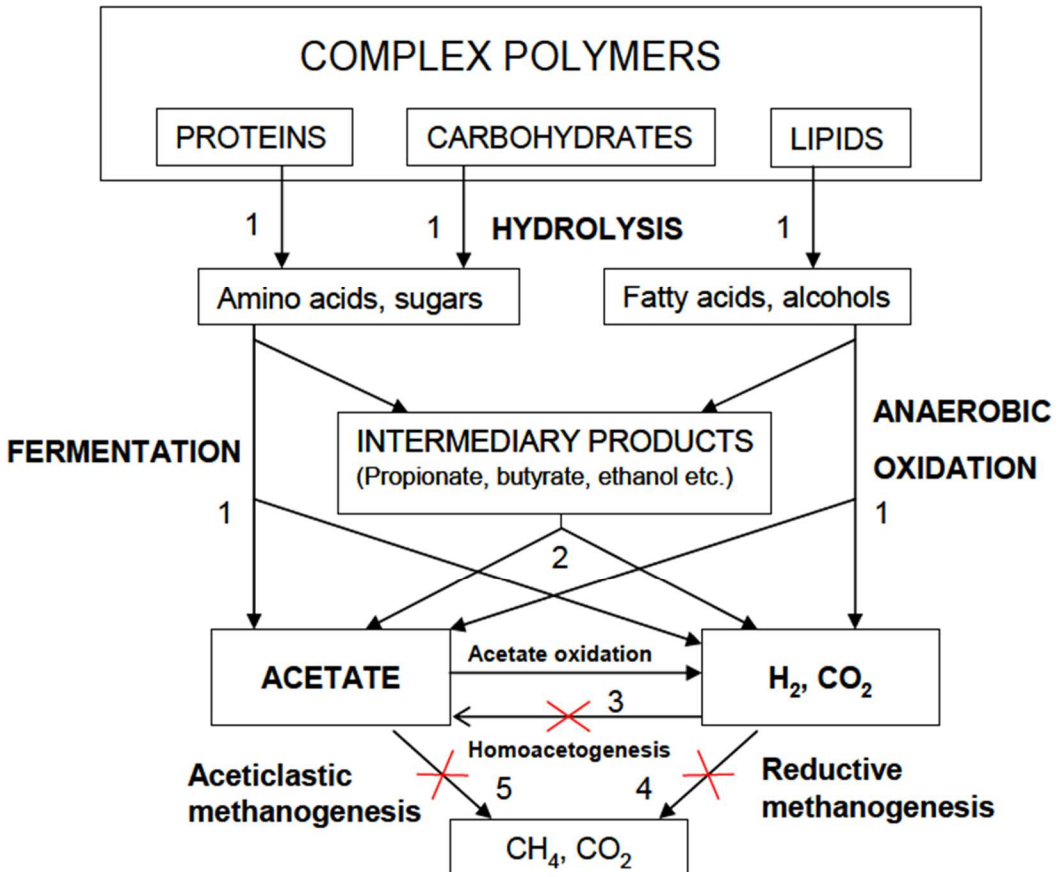

Figure 2.2-Process of anaerobic digestion of organic compounds. (Modified from Pavlosthathis and Giraldo-Gomes, 1991). 1, fermentative microorganism; 2, hydrogen producing acetogenic microorganism; 3, hydrogen-consuming acetogenic microorganism; 4. CO_2-reducing methanogens; 5, aceticlasctic methanogens

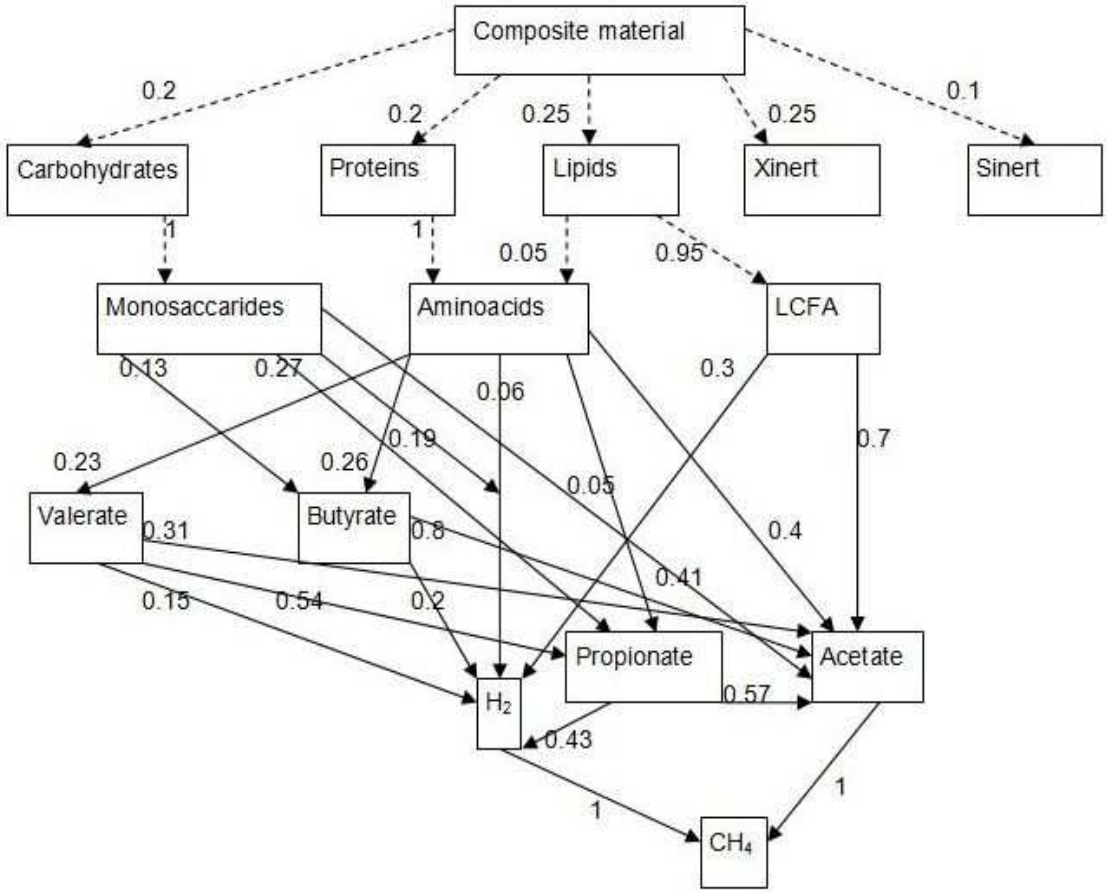

Figure 3.3-Diagram of components and processes in ADM 1 model (Batstone et al., 2002). (The numbers in this diagram are fractions in sewage sludge anaerobic digestion)

CHAPTER 4

ANAEROBIC DIGESTION OF DAIRY PROCESSING WASTE, ALGAE & GRASS IN PILOT AND FULL SCALE[1]

Abstract

This paper presents results of continuous fermentations of algae, lawn grass clippings and dairy processing waste (DPW), commingled and digested in duplicate 60 L and 3,800 L Induced Bed Reactor (IBR) anaerobic digesters at mesophilic conditions in trials that went for about two years. It was hypothesized that commingling DPW, algae and grass would be better than trying to digest them individually primarily because of problems with low pH but also to help balance nutrient content. The goal was to commingle municipal waste in such a way that no pH control chemicals would be required. The research also yielded information about solids loading rate (SLR), efficiency of chemical oxygen demand (COD) and solids removal and biogas production. Under the conditions of the study, commingling algae or grass with DPW made it possible to avoid addition of pH control chemicals. When treated alone, COD removal from algae was about 45% with a hydraulic retention time (HRT) of 24 day and specific SLR of 0.9 g total solids (TS) $L^{-1}d^{-1}$. Adding up to about 92% (solids basis) DPW that included hard and soft cheese whey and milk processing and yogurt waste (COD = 107 g

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[1] The authors are Conly Hansen, Jianming Zhong, and Jerald Hansen.

L^{-1}) to the algae improved COD removal to as high as 87% with SLR = 2.3 g L^{-1} d^{-1}. Under these conditions, biogas yield was 0.37 L (SATP (T = 25°C P = 100 kPa)) g^{-1} of COD loaded. The pH of commingled influent was 3.5 – 5.4. When algae were no longer available, fresh grass clippings were slurried and commingled with DPW. Adding 1.61% grass to DPW (solids basis) resulted in COD removal of 94% with SLR = 1.21 g^{-1} $L^{-1}d^{-1}$. Biogas yield was 0.37 L (SATP) g^{-1} of COD loaded.

Keywords.

Algae, anaerobic digestion, dairy processing waste, pH control, grass

Introduction

Purpose

The purpose of this project was to research and demonstrate anaerobic digestion (AD) of the municipal waste of Logan, Utah USA. These included commingled algae, grass clippings and diary processing waste (DPW). Substrates were digested in an induced bed reactor (IBR) anaerobic digester at mesophilic conditions. In this project, AD destroyed organic matter that otherwise would have been treated by municipal liquid or solid waste treatment systems.

Food wastes (food processing waste, food scrapes) is the largest percentage (up to 21%) among the classes of municipal solid waste listed by the USEPA (2013) going into sanitary landfills and incinerators. Sometimes food waste is fed to livestock; however for various reasons, including poor control of nutrition, odor, vectors and threat of diseases, it

is difficult to find farmers who will take it. Landfilling is not a good alternative as it can be expensive and it is environmentally prudent not to dispose of this material in landfills where it produces greenhouse gases (European Council, 1999). Hence, efficient ways to utilize this material must be discovered. Biological treatment methods include composting and AD. Composting represents an energy consuming process (30 - 35 kWh consumed per ton of waste input) and it releases a relatively large amount of CO_2 as well as pungent odors into the air. AD produces much more energy than is required to run the process (100 - 150 kWh net energy per ton of input waste) and odors are usually not released (Braber, 1995). Energy is produced during AD because methane gas is produced as part of the anaerobic digestion process. Methane is the primary component of natural gas.

Logan city provided the algae substrate for these studies. It came from the facultative lagoons used to treat up to 14 million gallons per day (MGD) of municipal wastewater. During the first year of this study, Logan was experimenting with removal of algae from these lagoons as a way to reduce phosphorus concentrations in the effluent. A company in Logan that produced cream cheese, processed cheese and yogurt provided DPW for both years of the study. Algae were commingled with DPW the first year and digested in duplicate 60 L Induced Bed Reactors (IBRs). Duplicate 3,800 L IBR's were used in the second year of the study. All the IBR's were located near the Logan lagoons Algae was not available the second year of the study because Logan had completed its trials with algae removal. Grass clippings were chosen to replace the algae as a

commingled substrate because this material was of interest to the municipal waste industry for creating energy (Buckle 2010). There was ample storage of grass clippings located near the Logan lagoons. The near neutral pH, carbon, nitrogen, phosphorus and potassium content of grass clippings are generally favorable for commingling with food waste (Yu et al., 2002; Mata-Alvarez et al., 2000, Starbuck, 2003). Preliminary trials (data not shown) adding grass clippings to the 3,800 L IBR's were conducted early in the second year that showed that adding grass clippings to DPW did not harm the AD process.

It was hypothesized that commingling moderate and low pH substrates would control pH without additional buffer chemicals. The goal was to commingle municipal waste in such a way that no pH control chemicals would be required. There may have been an additional benefit of mixing substrates with relatively high and low carbon to nitrogen (C:N) ratios. Optimal C:N ratio in anaerobic digestion is thought to be about 20:1 to 30:1 (Yen & Brune, 2007). Algae and grass clippings were low at 6:3 and 17:1 respectively (Michel et al., 1993, Wahal, 2010). C:N ratio of DPW can be very high (>70:1) depending on the degree of deproteinization (De Haasta1 et al., 1985). The effect of C:N ratio in these experiments, however, was not a goal of this study.

The experimental approach was to first conduct trials in the 60 L IBR anaerobic digester to gather information about commingling substrates in a small scale and then to scale up the same experiments in a larger IBR. However, since algae were not available for the scale-up experiments, grass clippings were used. The DPW was from the same

source and similar for both the smaller and larger scale experiments.

Induced Bed Reactor

The IBR effectively decouples hydraulic retention time (HRT) from solids retention time (SRT) making it possible to significantly reduce HRT for many organic wastes that may contain significant amounts of undissolved solids. The IBR is like an upflow anaerobic sludge blanket (UASB) digester in that solids are captured within the digester tank due to a solid/liquid/gas separator located ⅔ – ¾ of the distance from the bottom of the tank. Influent enters the bottom of the tank. The IBR is self-mixed by the rising gas bubbles and solid particles surrounded by gas bubbles. Slow growing anaerobic microorganisms need to be captured and maintained within a digester in order to have a concentrated area of sludge that is made up mostly of anaerobic microorganisms that will relatively quickly consume organics. The liquid can pass through whilst the solids are captured at the solid/liquid/gas separator and sink back into the sludge bed. The IBR differs from the USAB in that the upward flow of liquid is lower and the solid/liquid/gas separator is such that relatively large sized solids will not plug the outlet (Dustin et al., 2012). Advantages of the process include a high rate digestion, which brings down capital costs for tanks and handling equipment, a relatively small space requirement, ease of management and the fact that the IBR can handle a relatively abundant amount of large, solid particles in the influent.

Landfills are the most common disposal method for most solid municipal organic materials, which results in the release of large amounts of methane to the environment,

even when provisions are made to capture it. Methane is considered to be 21 times worse than carbon dioxide as a greenhouse gas (USEPA, 2009). AD aids in the treatment of municipal organic wastes, as well as provides renewable energy in the form of biogas. AD effectively reduces the volume and mass of organic waste products. Anaerobic microorganisms convert their organic substrates mostly into biogas. The biogas is a mixture of primary methane with carbon dioxide. Because of the slow growth of anaerobic bacteria, there is a relatively little solid byproduct from the organics destroyed in the process.

Biogas produced in an anaerobic digester must be cleansed of certain contaminants to facilitate its use for beneficial purposes such as combined heat and power (electrical generation) or producing compressed natural gas fuel. Zeolite regeneration was accomplished with a temperature swing at temperatures below 250°C without the consumption of reagents.

Materials and Methods

Two 60 L and two 3,800 L IBR's were installed at the Logan wastewater treatment facility. Influent substrate characteristics are shown in Table 4.1. These digesters were operated continuously during the time data was taken with loading rates over time as given in Table 4.2 (60 L) and Figure 4.1 (3,800 L). The temperature was monitored and controlled with Cole-Parmer 16B-33 controllers (Vernon Hills, IL) and heating cable (Mor Electric Heating Assoc., Comstock Park, MI) for the 60 L tanks and water jackets on the 3,800 L tanks. Electric water heaters (Sentra 220V, Advantage

Engineering, Greenwood, IN) heated and circulated the jacket water. The pH could be controlled with Cole-Parmer 350 controllers, (Vernon Hills, IL) and associated peristaltic pumps (Cole-Parmer 7553-80) to keep pH above 6.8 in the digesters by adding sodium hydroxide. After startup it was not used in any of the digesters and no acid addition was needed. There was no attempt to control pH in the 3,800 L digesters, the reason being that part of the experiment was to control pH without the addition of chemicals. Biogas production was monitored with Alicate mass flow meters (Tucson, AZ). Feed rate for the 60 L IBR's was automated with timers (Cole Parmer Model # R-94400-62, Vernon Hills, IL) and electrically controlled valves (Ingersoll Rand Model # P251SS-120-A, Dublin, Ireland) that controlled air supply to a diaphragm pump(ARO 1", Ingersoll Rand, Dublin Ireland). The larger IBRs were also automated for control of feed rate using an Omicron H3CR timer (Kyoto, Japan) with associated valves as were for the 60 L digesters except that the 3,800 L digesters were fed with a Sandpiper 2" diaphragm pump (SA2, Staffordshire, UK).

The 60 L IBR's were fed every four hours and the 3,800 L were initially fed every six hours then fed manually. The amount of substrates added per day was verified by noting the change in substrate depth in the semitransparent storage containers which were marked with graduations.

The 60 L digesters were operated in duplicate for six months with stable biogas production and then data were collected and reported for six months of operation. The temperature in the 60 L IBR's was 39 – 40 °C during these trials. DPW for the 60 L

digesters was provided by the Utah State University dairy processing lab that makes dairy products for the USU campus. DPW from USU consisted mostly of hard cheese whey and out of specification dairy products including ice cream mix, yogurt and milk. Experiments started with 100% algae. Following that, 20% DPW was added to the algae, then 50%, then 80% on a wet basis which equaled the solids numbers shown in Table 4.2.

The two 3,800 L IBR's were operated for two years, at first running simultaneously with the 60 L IBR's except in the coldest months. They were shut down from December to April because of freezing weather and snow which made it difficult to deliver and store substrate at the site. Data given in this paper are representative of observations over the two-year operation and covers a time period from early June through September of the second year. The temperature in these IBR's was consistent at 40°C during the trials. On startup, sludge was pumped from the bottom of the Logan lagoons wastewater treatment facility into the 3,800 L IBR's to about 40% volume as inoculum. DPW for the 3,800 L digesters was provided by Schreiber Foods, Logan, UT plant. The USU dairy could not supply sufficient DPW for the 3,800 L digesters. DPW from Schreiber consisted mostly of cream cheese whey and processed cheese and yogurt wastes. Algae were not available for the 3,800 L IBR's for the study because there was no way to separate it from lagoon water. Grass clippings were crudely chopped (≤13 mm) and mixed with water to a little more than 1% solids (Table 4.1) to make a grass slurry (GS) before being pumped into the 3,800 L digesters along with DPW. The digesters could handle higher grass solids, but GS with higher solids content was difficult to pump

even using the diaphragm pump with 50 mm inlet and outlet. Each batch of substrate was sampled and chemical oxygen demand (COD) and total solids (TS) analyses were performed as the characteristics changed slightly between batches. Normally batches were picked up once per week and stored on site at ambient temperature until used. Total P and N were measured for algae only. All analyses were performed according to standard methods (APHA-AWWA-WEF, 1992). Biogas methane (CH_4) percentage was analyzed with an Agilent 6890 GC using an RT-Msieve 5A Plot capillary column (Restek) (Agilent, Santa Clara, CA).

A zeolite based, regenerable biogas conditioner supplied by AD Tec, (Springville, UT) was used to effectively clean the biogas produced. Zeolite is a hydrated silicate of aluminum with alkali metals. H_2S was removed to below 10 ppm and H_2O to <1%.

Results and Discussion

Results of the experiments for the 60 L IBR trials are summarized in Table 4.2. It can be seen that the IBR effectively digested algae resulting in a COD removal of 45% with a 24 day HRT. Dissolved air flotation (DAF) with aluminum sulfate addition was used to separate the algae. The addition of aluminum sulfate ($Al_2(SO_4)_3$) did not appear to affect the AD process. The DAF operators reported aluminum sulfate residual in the algae was never above 100 ppm. Addition of up to nearly 92% DPW solids to the algae solids (commingled substrate COD = 84 g L^{-1}) improved COD removal to as high as 87% with SLR = 2.3 g L^{-1} d^{-1}. Therefore DPW addition appeared to improve the process as was expected. The results were encouraging indicating success in digesting algae alone and

that addition of other wastes improved digestion of both substrates. One thing that was obviously beneficial about adding algae to DPW was its aid in controlling pH. Under the conditions of this study, commingling algae with DPW made it possible to avoid addition of pH control chemicals to the 60 L digesters. The COD removal efficiency compared favorably with results reported for algae alone (Salerno et al., 2008; Golueke et al.) but the HRT for digesting other organic materials (dairy manure) in IBRs has been 3.8 to 7.5 days (Dustin, Hansen, & Dustin, 2012), which is much shorter than the HRTs for these algal digestion trials. The relatively high biogas yield with an HRT of 24 d and 100% algae in Table 4.2 was likely not accurate. It probably reflected the fact that the 24 d HRT experimental trials immediately followed a 10 d HRT trial in the same IBR (data not reported in this paper). There was likely a buildup of substrate from the 10 d HRT trials that was slowly broken down. However, the 24 d HRT was probably too long for algae as the IBR was able to handle excess substrate throughout the remaining trials without addition of pH control chemicals. More experimentation will have to be done to find the best HRT. The COD removal efficiency, particularly when DPW was added, was impressive. The removal efficiency was best when only 20% algae were added. The average specific biogas yield with 20% algal addition in the 60 L digester was 0.37 L (SATP (25°C, 100 kPa)) g^{-1} TS loaded.

Results of experiments in the 3,800 L IBR's are shown in Table 4.3 and Figure 4.1. COD removal was continuously above 90%. As shown in Figure 4.1, the feeding rate was increased about 8x after GS was regularly added. Interestingly, it was discovered that

the IBR would tolerate a single daily bulk feeding equivalent to the amount of DPW fed four times/day. This may have been because the digester needed time to recover between feeding of even low volume acidic substrate. The pH in the digester always dropped immediately after feeding even for the low feed rate of four per day feedings and low volume added at the start of the trial. Typically, it would drop by 0.5±0.5 pH points. It always recovered under the loading rates given in Table 4.3 and Figure 4.1 after grass clippings were added. The influent pH was 3.5 – 4.6 whenever commingled influent contained ≥ 50% DPW. Not shown in the Tables was the fact that pH of DPW was 3 – 4 for both the 60 L and 3,800 L trials.

GS helped to control pH. As shown in Figure 4.1, the pH at the start of the experimental trials in the larger IBR's dipped below 6.6. No biogas was produced before 7/2/2012 (Table 4.3) indicating failure of the digester. The first grass clipping were added on 7/3/2012. In the first time period, with very little DPW added; percent GS added was nearly 180% that of DPW on a solids-solids basis. Under these conditions the pH rose and the failed digester recovered without addition of starter or pH control chemical. After that, GS were added to help stabilize the digester. As little as 1.61% GS (solids to solids basis) added to DPW maintained pH with a relatively low SLR (1.21 g L^{-1} d^{-1}) for an IBR (Dustin et al, 2011). Further research will have to be done to understand what the maximum loading rate can be. The pH of commingled influent was acidic (3.5 – 5.4) regardless of the percentage of DPW commingled in either digester (Tables 4.2 and 4.3). However, since organic acids are the cause of low pH in food wastes, the methane

forming bacteria in the digesters were able to consume these acids and maintain a stable pH in the digesters. COD removal for the commingled waste containing 1.61% grass solids was 94% (Table 4.3). The specific biogas yield was 0.37 L (SATP) g^{-1} TS loaded. Biogas contained 70% methane.

Based on SLR, algae were a better substrate for commingling with DPW than GS (Tables 4.2 and 4.3). When GS solids were added up to nearly 6.5%, the specific SLR was not nearly as high as for 8.16% algae. More DPW could not be added because the digester would not keep a stable pH (Figure 4.3) though it appeared to acclimate over time as DPW was increased and a steady amount of grass added solids kept pH near neutral. The optimal SLR and percent GS or algae were not determined. It will take much more experimentation to determine those values. GS was much more difficult to pump at equivalent solids concentration as for algae and thus the solids content of GS commingled with DPW was not as high as for algae. With the equipment available, the solids addition of GS was about half that of algae when equal volume ratios of GS (solids ~1.1%) and algae (solids ~2.2%) were commingled with DPW. More experimentation will have to be done in full scale with better grinding and pumping equipment to optimize the amount of GS commingled with a low pH and high COD substrate like DPW. It is much more difficult to conduct AD experiments in full scale compared to lab scale, but full-scale data is needed to help potential investors decide how best to utilize certain food wastes. It can only be concluded that GS did help to control pH and GS is usually available in most municipalities in the USA whereas algae is not.

A zeolite based biogas conditioner removed sulfur compounds including H_2S, H_2O and other contaminants from biogas produced in the 3,800 L IBR's to non-detectable limits (< 0.050 ppmv) (Table 4.4). Hydrogen sulphide or H_2S is a corrosive, highly poisonous gas and it is best to remove it from the biogas. This was done with proprietary zeolite. Zeolite is a molecular sieve with molecule-size pores that can temporarily lock H_2S and thereby trap this and other objectionable gases in biogas. The zeolite used did not remove CO_2. After passing through the zeolite bed which was 6.1 m (20 ft.) long and 76 mm (3 in) in diameter, the treated biogas was nearly pure methane and carbon dioxide. It had no detectable odor. Table 4.4 shows the results of total reduced sulfur biogas analysis (ASTM D-5504). According to the manufacturer, the zeolite can be reconditioned indefinitely with moderate temperature (250º C) swing to about 90% of its uptake capability when new (ADT, 2012). The zeolite conditioner used would remove H_2S and water vapor with a 10 L/min biogas flow rate for about two weeks without reconditioning. By condensing much of the water vapor in the gas at 4º C before it reached the zeolite bed, the biogas conditioner would not need to be regenerated for about 10 weeks at the 10 L/min biogas flow rate.

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Table 4.1-Substrate characteristics

Substrate	COD[1] (g/L)	Total solids (g/L)	Total phosphorus (mg/L)	Total nitrogen (mg/L)
Algae	24.7±2.3	21.7±4.1	1.75±1.40	2.06±2,81
Dairy processing waste 1st year	98.7±4.0	61.1±4.8	ID[2]	ID
Dairy processing waste 2nd year	107.0±13,4	65.4±13.3	ID	ID
Grass Slurry	8.23±6.07	11.1±8.2	ID	ID

[1]Chemical oxygen demand, [2]Insufficient data

Table 4.2-Data taken in 60 L IBR fed dairy processing waste and algae with pH control

HRT [1] (d)	% algae[2]	Influent pH	Specific SLR	COD (g/L)		COD Removal (%)	Total Solids (g/L)		TS Removal (%)	Biogas yield L/g of COD loaded[3]	Methane content (%)
			(g/L-d)	out	in		out	in			
24	100/0	7.4±0.4	0.9±0.17	13.6±0.7	24.7±2.8	44.94±0.02	13.5±3.3	21.7±4.1	37.8±7.2	0.32±0.04	59.5±0.6
24	58.7	5.4±0.3	1.3±0.20	8.3±0.6	33.5±2.7	75.22±0.03	11±3.0	31.2±5.0	64.7±7.6	0.29±0.02	62.3±0.7
24	34.6	4.9±0.2	1.7±0.20	24.2±0.8	62.7±3.2	61.40±0.01	14.3±3.4	41.3±5.0	65.4±4.5	0.24±0.01	63.5±0.4
24	19.1	4.8±0.5	2.0±0.22	10.7±0.5	70.5±3.0	84.82±0.01	13.6±4.0	47.0±5.2	71.1±4.4	0.33±0.01	62.7±0.5
24	8.2	4.6±0.3	2.3±0.22	10.8±0.6	83.6±3.8	87.08±0.02	14.3±2.9	55.0±7.0	74.0±6.3	0.37±0.01	64.2±1.0

[1]Hydraulic retention time, [2]Percent algal solids mixed with dairy processing waste solids, [3]At SATP (25 °C and absolute pressure of 100 kPa)

Table 4.3-Data taken in 3,800 L IBR fed dairy processing waste and grass.

Time period	% GS[1]	Specific SLR[2] (g/L-d)	Influent pH	pH in IBR	COD (g/L)		COD Removal (%)	Total Solids (g/L)		TS Removal (%)	s yield (L/g of COD	Methane content (%)
					out	in		out	in			
6/4 to 7/2	179.6	0.11	ID[4]	6.05±0.6	ID	ID	ID	ID	ID	ID	0	ID
7/3 to 8/27	6.42	0.26±0.0	3.5±0.5	6.84±0.1	2.4±0.96	57.9±1.32	95.4±0.03	3.6±1.2	36.5±0.2	90.2±0.3	0.37±0.42	70.3±17
8/27 to 10/3	1.61	1.21±0.0	3.5±0.5	7.00±0.1	5.3±1.8	88.5±1.43	94±0.03	6.6±1.7	58±4.2	88.7±0.7	0.37±0.21	ID

[1]Percent grass solids mixed with dairy processing waste solids, [2]Solids loading rate, [3]At SATP, [4]Insufficient data

Table 4.4-Total reduced sulfur compounds analysis results

Analyte	Post filtration Result	Pre-filtration result
Hydrogen Sulfide	< 0.050 ppmv	198 ppmv
Carbonyl Sulfide	<0.050 ppmv	< 0.100 ppmv
Sulfur Dioxide	< 0.050 ppmv	< 0.100 ppmv
Methyl Mercaptan	< 0.050 ppmv	1.93 ppmv
Ethyl Mercaptan	< 0.050 ppmv	< 0.100 ppmv
Dimethyl Sulfide	< 0.050 ppmv	0.746 ppmv
Carbon Disulfide	< 0.050 ppmv	<O.IOO ppmv
Isopropyl Mercaptan	< 0.050 ppmv	<O.IOO ppmv
tert-Butyl Mercaptan	< 0.050 ppmv	< O.IOO ppmv
n-Propyl Mercaptan	< 0.050 ppmv	2.17 ppmv
Methylethylsulfide	< 0.050 ppmv	< 0.100 ppmv
sec-Butyl Mercaptan	< 0.050 ppmv	< 0.100 ppmv
Thiophene	< 0.050 ppmv	< 0.100 ppmv
iso-Butyl Mercaptan	< 0.050 ppmv	< 0.100 ppmv
Diethyl Sulfide	< 0.050 ppmv	< O.1OO ppmv
n-)3utyl Mercaptan	< 0.050 ppmv	< O.1OO ppmv
Dimethyl Disulfide	< 0.050 ppmv	< O.1OO ppmv
2-Methylthiophene	< 0.050 ppmv	< 0.100 ppmv
3-Methylthiophene	<0.050ppmv	< O.1OO ppmv
Tetrahydrothiophene	< 0.050 ppmv	< O.1OO ppmv
Bromothiophene	< 0.050 ppmv	< O.1OO ppmv
Thiophenol	< 0.050 ppmv	< O.1OO ppmv
Diethyl disulfide	< 0.050 ppmv	< 0.100 ppmv
Total Unidentified Sulfur	< 0.050 ppmv	< 0.100 ppmv
Total Reduced Sulfurs as H_2S	< 0.050 ppmv	203 ppmv

All compound's concentrations expressed m terms of H2S (TRS does not include COS and S02). Sample Reporting Limit (SRL) is equal to Reporting Limit x Canister Dil. Fac. x Analysis Dil. Fac.

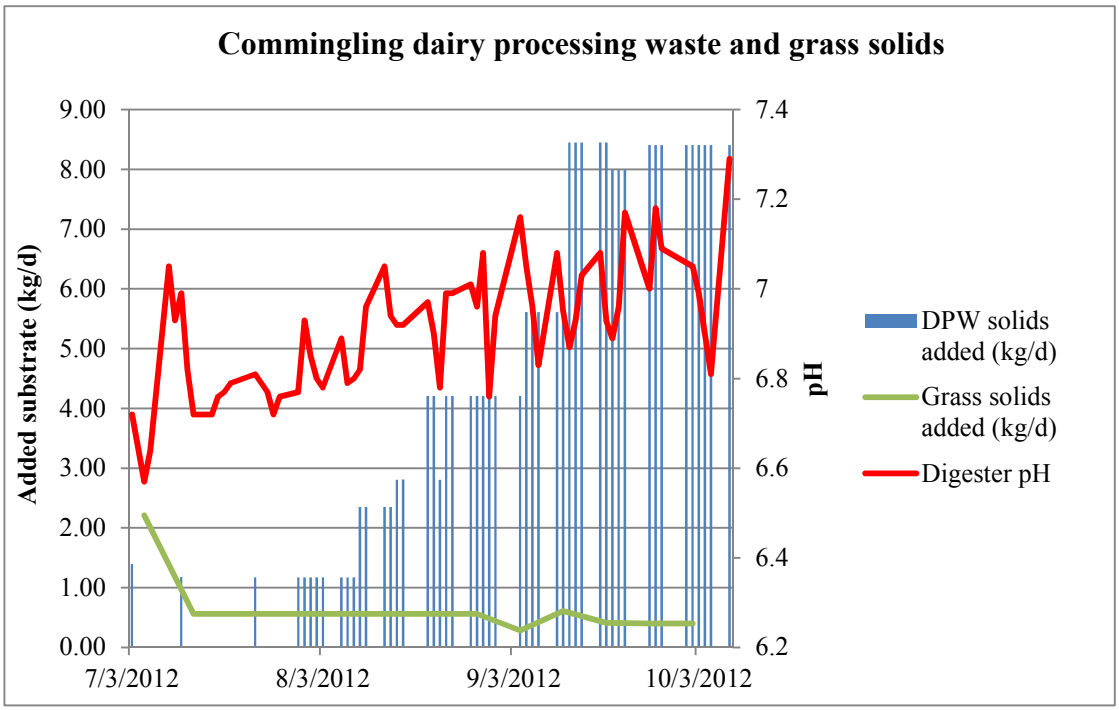

Figure 4.1-Chart showing the effect of adding grass slurry to dairy processing waste solids in stabilizing pH in pilot scale (3,800 L) IBR anaerobic digester.

CHAPTER 5

OPTIMIZATION OF ANAEROBIC HYDROGEN AND METHANE PRODUCTION FROM DAIRY PROCESSING WASTE USING A TWO-STAGE DIGESTION IN INDUCED BED REACTORS (IBR)

Abstract

This study investigated the effects of pH, temperature and hydraulic retention time (HRT) and organic loading rate (OLR) on hydrogen production from dairy processing waste (DPW) in semi-continuous 60 L pilot induced bed reactors (IBR). Results show pH played a key role on hydrogen production and the optimal pH range was in 4.8-5.5. Digestion under thermophilic temperatures (60 °C) had advantages of gaining higher hydrogen yield and suppressing the growth of methanogens. The optimal OLR was 32.9 g-COD/l-d at HRT of 3 days. Under optimal conditions, highest hydrogen yield was 160.7 ml/g-$COD_{removed}$ with 44.6% COD removal. Two-stage digestions demonstrated more energy gain from methane production and further COD removal. The overall gas production in two-stage digestion was 71.7 ml hydrogen and 61.0 ml methane per gram DPW COD. The overall COD removal under optimal conditions was 88.2%.

Highlights

- The optimal pH range of anaerobic hydrogen production from dairy processing waste (DPW) was 4.8-5.5.

- Thermophilic digestion can gain higher hydrogen yield and suppress the growth of methanogens.
- Optimum DPW loading rate was 32.9 g-COD/l-d at a hydraulic retention time (HRT) of 3 days for hydrogen production.
- Two-stage induced bed reactors (IBR) produced mixed gas with higher heating value and COD removal than single-stage.

Keywords:
Hydrogen production, Dairy processing waste (DPW), Induced bed reactor, Two-stage digestion

Introduction

Hydrogen is considered an alternative fuel of great potential. It is environmentally friendly because only water is produced when it is combusted and was identified as a clean energy carrier for the future at the first World Hydrogen Conference [1]. Hydrogen has an energy density of 143 MJ/kg, which is 2.6 times higher than methane and 3.3 times higher than gasoline. Hydrogen has great potential to reduce the use of fossil fuels. However, in industry, the majority of hydrogen is produced from fossil-fuel sources such as natural gas, oil, and coal [2].

Dairy processing waste (DPW) is the waste produced from manufacturing dairy products: cheese, yogurt, ice cream, milk, butter, etc. DPW is high in chemical oxygen demand (COD) due to its lactose, fat and protein content, and therefore needs to be

appropriately treated. The discharge of excess amounts of DPW, such as cheese whey, onto land can damage the chemical and physical structure of soil, pollute groundwater and may also affect air quality [3, 4]. Now there are more and more whey protein concentrate and isolate products [5], which has reduced DPW quantities. However, finding a novel, cost-effective disposal or utilization technology for waste has been an important issue for the dairy industry because of:

1) still high lactose content in DPW;

2) high investment cost in whey protein processing equipment;

3) increased volume of dairy processing byproducts;

4) increasingly strict legislative requirements.

Anaerobic digestion is a potential technology for both hydrogen production and food waste management. Anaerobic digesters can produce energy from inexpensive and renewable energy sources such as food processing waste. Recent research proved that certain strains of bacteria (e.g., bacteria from the genus *Clostridium*) are particularly effective at producing hydrogen as a by-product during anaerobic digestion of organic waste material [6]. Although various studies have been done on producing hydrogen with anaerobic digestion, there are still several obstacles that must be overcome before applying this technology economically at an industrial level. Induced bed reactors (IBR) are designed specifically for anaerobic digestion[7], and IBR has the ability to handle short HRT digestion of many organic wastes that may contain high un-dissolved solids [8,

9]. This may make it a very good digester for hydrogen production because studies show low HRT is typically required for hydrogen production[10, 11].

Recent studies have shown production of hydrogen and methane anaerobically from wastes organics in two-stage systems; for example, [12, 13] show hydrogen and methane can be produced using cheese whey. Other materials such as cassava wastewater[14] and sweet sorghum[15] can also be used to produce hydrogen and methane. In those studies, UASB or CSTR digesters were used. However, no research has been reported using the substrate DPW. In the dairy industry large amounts of DPW, which has high content of fat, protein and lactose and may contain cleaning chemicals, is produced. And no published anaerobic hydrogen production study is based on IBR digester. In this research, single-stage digestions were performed first in 60 L pilot IBRs to explore the optimal conditions of pH, temperature and HRT/OLR for hydrogen production from DPW. Later under optimal hydrogen production conditions, a second unit IBR was added for testing the performance of methane production from the effluent of the hydrogen reactor.

Material and Methods

Substrate and Seed

DPW was provided by Aggie Creamery (Utah State University, Logan). DPW is a mix of dairy production wastes. About 40-50% (by volume) of DPW is cheddar cheese whey; 50-55% of DPW is the waste from the production of ice cream, yogurt and milk;

and about 5% is rinsing wastewater. DPW was stored at 4 °C before use. Its characteristics are presented in Table 5.1.The inoculum was from the sludge of an anaerobic digester that was used for biogas production from algae in Logan Wastewater Treatment Facility (Logan, UT).

Experiment Set-Up

Two 60-L IBRs were constructed and installed. To enrich hydrogen-producing bacteria and inactivate methanogens, a 25-L inoculum was mixed with 20-25 L DPW in a 60-L IBR to reach the pH of 5.0-5.5. Then the mixture (inoculum sludge and DPW) was heat-treated (65 °C) overnight.

Single-stage digestion: two IBRs were used for optimization of hydrogen production from DPW (Figure 5.1). Three different parameters: temperature, pH, and HRT were examined. One IBR was set at temperature of 40 °C and another was set at 60 °C (due to the heat loss during transfer, liquid temperatures in the central digester areas were 37-38 °C and 55-58°C, respectively). Three pH ranges (4.0-4.5, 4.8-5.3 and 5.5-6.0) and three HRT values (1, 3 and 5 days) were tested in a 3-factor full factorial design.

The two-stage digestion setup is illustrated in Figure 5.1. The two-stage system had a 60 L hydrogen IBR and a 60 L methane IBR. The effluent of the hydrogen IBR was used as influent for the methane IBR. The hydrogen IBR was operated under the optimal pH/HRT/temperature conditions found in the single-stage digestion preliminary trials.

The second stage methane digester was operated at pH of 6.8-7.5 and temperature of 40 °C. The inoculum for the methane IBR was from the same sludge source (Logan wastewater treatment facility), but without heat treatment.

IBR Operation

The temperature was monitored and controlled with Cole-Parmer 16B-33 controllers (Vernon Hills, IL) and heating cable (Mor Electric Heating Assoc., Comstock Park, MI). pH could be controlled with controllers (Model 350, Cole-Parmer, Vernon Hills, IL) and associated peristaltic pumps (Cole-Parmer 7553-80) to keep pH within ± 0.1 of the set point in the digesters by adding sodium hydroxide solution (1 mole/L) in the hydrogen IBR. No chemical was needed for pH adjustment in the methane IBR. Feed rate was automated with timers (Model R-94400-62, Cole Parmer, Vernon Hills, IL) and electrically-controlled valves (Model P251SS-120-A, Ingersoll Rand, Dublin, Ireland) that regulated air supply to a diaphragm pump (ARO 1", Ingersoll Rand, Dublin Ireland). The IBRs were fed every four hours.

Analytical Methods

Total solids (TS) and volatile solids (VS) were measured by standard methods (APHA, 1998). Total chemical oxygen demand (COD) (Hach Method 8000), total organic carbon (TOC) (Hach Method 10128), total nitrogen (TN) (Hach Method 10072) and total ammonia (NH_4-N) (Hach Method 10031) were analyzed using Hach test kits (Hach DR/870). Hydrogen and biogas production were measured by mass flow meters (Model 822-L, Sierra, Monterey, CA). Data for gas flow rate were saved every five

minutes using a data logger (CR 1000, Campbell Scientific, Logan, UT). Biogas and hydrogen composition were analyzed in an Agilent 6890 GC using an RT-M sieve 5A Plot capillary column (Restek) (Agilent, Santa Clara, CA). The statistics analyses were performed in R software (version 3.0.3) [16].

Results and Discussion

Single-Stage Digestion

Three parameters temperature, pH, and HRT were examined for optimization of hydrogen production from DPW. Values of temperature, pH, HRT and OLR in each experimental run are listed in Table 5.2. The results of COD removal, hydrogen yield, hydrogen content, and methane content in nine experimental runs are shown in Figure 5.2. The main effect of these three parameters on COD removal, hydrogen yields, hydrogen content, and methane content was analyzed by analysis of variance (ANOVA) and the results are listed in Appendix A (Table A-1).

Effects of pH on hydrogen production. Keeping pH in a certain range is crucial during semi-continuous or continuous digestion operation. pH should be in the range 6.8-7.5 for single stage methane anaerobic digestion [17]. Three pH ranges (4.0-4.5, 4.8-5.5 and 5.6-6.0) were tested to examine the effect on hydrogen production from DPW. As listed in Table 5.2, experiments I, II and III were run in the pH range of 4.0-4.5; experiments IV, V and VI were run in the pH range of 4.8-5.5; and experiment VII, VIII, and IX were run in the pH range 5.6-6.0. In general, as shown in Figure 5.2, digestions in

the pH range 4.8-5.5 had higher COD removal/hydrogen yield/hydrogen content and lower methane content than the digestions in the other two pH ranges. The highest hydrogen yield-160.7 ml H2/g-COD removed; highest COD removal-44.6%, and highest hydrogen content-50.2% and lowest methane content-2.8% were all obtained in the 4.8-5.5 pH range. Statistical main effect results show pH had significant impacts on COD removal, hydrogen yield, hydrogen content and methane content (ANOVA, $p<0.05$) (Table A-1). Such results were expected because the H^+ in the extracellular environment selects the bacteria that can survive at this pH range, and at the same time suppresses or kills other organisms that cannot grow [18, 19]. Different pH ranges may result in different pathways during the complex digestion process. Although methanogens were killed or inactivated during seed preparation (see Material and Methods), in the pH range 5.6-6.0 some methanogens eventually grew in the later period of our semi-continuous digestions. That might be why the methane content was higher than for pH ranges 4.0-4.5 and 4.8-5.5 (Figure 5.2). Growing methanogens may have rapidly consumed the produced hydrogen, which led to a low hydrogen yield. At pH 4.0-4.5 methane content was relatively low, which suggests a good suppression of methanogens. However, low COD removal and hydrogen yield may suggest that this pH range also suppresses the growth of hydrogen-producing bacteria. 4.8-5.5 was the optimal pH range for not only the growth of hydrogen producing bacteria but also the suppression of methanogens. Similar results were obtained by other studies using different substrates [20-23]. In order to increase the pH sodium hydroxide was used, which is expensive for applying to full-

scale digestion. Cheap pH buffer addition should be considered and investigated in future research.

Effects of HRT/OLR on hydrogen production. Organic concentration of DPW was 98.7 g-COD/l. When HRTs were kept at 1, 3 and 5 days, the OLR rates were 98.7 g-COD/l-d, 32.9 g-COD/l-d and 19.74 g-COD/l-d, respectively. As listed in Table 5.2, experiments I, IV and VII were run with 1 day HRT (98.7 g-COD/l-d); experiments II, V and VIII were run with 3 days HRT (32.9 g-COD/l-d), and experiments III, VI and IX were run with 5 days HRT (19.74 g-COD/l-d). Statistical main effect analyses showed HRT/OLR had significant impacts on COD removal and hydrogen yield (ANOVA, $p<0.05$), but not on hydrogen content and methane content (ANOVA, $p>0.05$) (Table A-1). As shown in Figure 5.2,under the optimal pH range 4.8-5.5, when HRT increased from 1 to 3 days (OLR decreased from 98.7 to 32.9 g-COD/l-d) the hydrogen yield increased from 111.4 to 160.7 ml H_2/g-COD removed at 60 °C ; when HRT increased from 3 to 5 days (OLR decreased from 32.9 to 19.74 g-COD/l-d) the hydrogen yield decreased from 160.7 to 131.5 ml H_2/g-COD removed at 60 °C. Accordingly, the highest COD removal – 44.6% was obtained in 3 days HRT compared to 40.2 % in 1 day HRT and 42.6% in 5 days HRT. These results suggest that too high or too low HRT/OLR is not optimal for hydrogen production.

Different substrates may affect the optimal HRT/OLR because of their characteristics. Here, DPW has high organic content (mainly lactose) and very low non-biodegradable solids. When using another substrate that has relatively low organic

content, the optimal HRT/OLR may be different [18,24-26]. Other factors that might be considered are feeding type: batch, continuous or semi-continuous operation. In our study, DPW was pumped every 4 hours. The optimal HRT/OLR might be different due to less impact on sludge bed when fed continuously.

Effects of temperature on hydrogen production. Each experimental run was performed under both mesophilic 40 °C and thermophilic temperatures 60 °C. Statistics analyses show temperature had significant impact on hydrogen yield, COD removal and methane content ($p<0.05$), but not in hydrogen content. As shown in Figure 5.2, hydrogen yields were higher at 60 °C than at 40 °C expect experiment I (no significant difference in I). The largest difference in COD removal was obtained in experiment VI, where 131.5 ml H2/g-COD removed at 60 °C versus 116.5 ml H2/g-COD removed at 40 °C. Also in experiment VI, COD removal was 8% higher at 60 °C than at 40 °C. These results were expected because increasing the temperature typically can enhance the activity of the enzymes until the optimal temperature is reached[27]. Another advantage of thermophilic digestion is increased solubility of some polymeric substrates. Thermophilic temperature digestion is suitable for some biomass containing substances that are difficult to hydrolyze, e.g., lignocelluloses[28]. Moreover, the methane content was lower at 60°C than at 40 °C (Figure 5.2), especially in the pH range 5.6-6.0. This result indicates that 60 °C had a better suppression of methanogens than 40°C. Combined with the previous pH results, it is concluded that pH and temperature are two important factors that keep methane content low during hydrogen production. In the pH range 4.8-

5.5 and 3 days HRT, our results showed 40 °C digestion had low methane content and relatively high hydrogen yield and COD removal as well. Thus, 40 °C is also feasible for hydrogen production from DPW when the digestions are operated under the optimal pH range and HRT/OLR.

Two-Stage Digestion

Methane production performance. In single-stage hydrogen production the highest COD removal was 44.6 %. There is still high COD in the effluent of hydrogen production. Further treatment is necessary before being discharged. Two-stage digestion was performed to produce both hydrogen and methane. The hydrogen IBR was operated under optimal conditions (3 days HRT/32.9 g/l-d OLR/pH of 4.8-5.5/60 °C) that were found in previous single-stage digestion. The effluent of the hydrogen production was used as influent for methane production (Figure 5.1). Table 5.3 lists the results of methane production in the second stage. Four different HRTs (8, 12, 15 and 20 days) were tested. COD removals in the four different HRTs were all above 50%. With the increase in HRT the COD removal increased. Over 70% COD removal was achieved when HRT reached 15 days or higher. pH inside the IBR were all 6.8-7.5, which was the optimal range for methane production. No chemical was needed for controlling pH in the methane IBR. Two-stage digestion had an advantage because chemical or buffer is usually required to increase pH when digesting DPW in single-stage digestion[29]. The methane yields were 168.8-178.1 ml CH_4 ml/g-COD removed. The highest methane yield was found with HRT 15 of days. The methane content was in the range of 60-65%.

Overall performance. The overall performance of two-stage digestion was evaluated. Table 5.4 summarizes the two-stage digestion under the optimal condition for hydrogen production and 15 days HRT for methane production. COD removal of 88.2% was reached in overall two-stage digestion. The effluent COD was as low as 11.7 g/l compared to the original DPW- 98.7g/l. Furthermore, after thermophilic treatment in the hydrogen IBR the effluent will have less pathogens [30]. It is safer to reuse or dispose the effluent. It should be noted that the methane IBR's operation is very flexible. Many HRTs/OLRs can be set without affecting pH inside the digester (Table 5.3). Higher HRT in the methane IBR means more complete digestion. For getting maximal energy and COD removal a larger volume methane IBR size compared to the hydrogen IBR is recommended in order to make a correspondingly higher HRT. As listed in Table 5.4 one gram COD of DPW can produce 71.7 ml H_2 and 61.0 ml CH_4. Compared to hydrogen single-stage digestion, 238.9 kJ more energy in heating value was produced in two-stage digestion of one gram DPW COD. Thus, it can be concluded for hydrogen production that using two-stage anaerobic process provides higher energy than using a single-stage anaerobic digestion.

Table 5.5 compares this study with some other previous research. It should be noticed that among all the results listed in Table 5.5 [13,14,31], our 60 L pilot-scale research had much larger digester size. Thus, our studies was closer to large-scale digestion. Compared to other cheese whey two-stage digestions, this research had significantly higher hydrogen yield than that of two-phase mesophilic UASB process at

an organic loading rate of 47.4 g COD/l-d (160.7 ml-H_2/g-COD removed versus 41 ml-H_2/g-COD removed)[31]. The hydrogen production rate from this study was lower than that of mesophilic CSTR process at an organic loading rate of 182 g COD/l-d; because the CSTR hydrogen process had much higher OLR (182 g COD/l-d versus 32.8 g COD/l-d) and much lower HRT (0.25 d versus 3 d)[13]. Within the same thermophilic hydrogen two-stage digestion, our results were very similar to the two-stage cassava wastewater digestion in terms of hydrogen yield, methane yield and overall COD removal[14].

Conclusions

In this study, optimization of anaerobic hydrogen production from DPW was explored in semi-continuous pilot-scale (60 L) IBRs. We found the optimal conditions for hydrogen production from DPW were: HRT 3 days / OLR 32.9 g-COD/l-d, pH range of 4.8-5.5, and 60 °C. Under these conditions, the highest hydrogen yield was 160.7 ml/g-COD removed, highest COD removal was 44.6% and highest hydrogen content 50.2% was achieved. Two-stage digestions were tested later for further energy extraction and COD removal. Results show two-stage production of hydrogen and methane can greatly increase the amount of energy harvested and will increase COD removal. With an HRT of 15 days in the methane IBR, methane yield was 178.1 ml methane per gram COD removed and COD removal was 73.1%. The overall gas production in two-stage digestion was 71.7 ml H_2 and 61.0 ml CH_4 per gram COD loaded. The overall COD removal was as high as 88.2%. This study demonstrated that the production of both hydrogen and methane can be efficiently coupled in a two-stage IBR digestion system.

The pilot-scale research here provides the data and design requirements for full-scale application.

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Table 5.1-Characteristics of dairy processing waste (DPW)

Dairy Processing Waste (DPW)	
pH	4.5±0.6
Total COD (g/L)	98.7±4.0
Soluble COD (g/L)	75.6±2.6
Total Solids (TS, g/L)	61.1±4.8
Volatile Solids (VS, g/L)	55.3±3.7
Total Organic Carbon (TOC, g/L)	24.8 ± 3.3
Total nitrogen (g/L)	4.06±1.21
NH_3-N(g/L)	2.30±0.81

Table 5.2-Values of temperature, pH, HRT and OLR in each experiment run

Experiment run number	Temperature (°C)	pH	HRT (days)	OLR (g-COD/l-d)
I	40 or 60	4.0-4.5	1	98.7
II	40 or 60	4.0-4.5	3	32.9
III	40 or 60	4.0-4.5	5	19.74
IV	40 or 60	4.8-5.3	1	98.7
V	40 or 60	4.8-5.3	3	32.9
VI	40 or 60	4.8-5.3	5	19.74
VII	40 or 60	5.5-6.0	1	98.7
VIII	40 or 60	5.5-6.0	3	32.9
IX	40 or 60	5.5-6.0	5	19.74

Table 5.3-Methane production performance in two-stage digestion

HRT (days)	OLR (g/l-d)	Influent pH	COD Removal (%)	pH inside digester	Methane Yield (ml/g-COD removed)	Methane content (%)
8	7.0	5.7±0.2	53.4±0.3	7.0±0.2	168.8±3.2	61.3±0.4
12	5.6	5.7±0.2	61.2±0.8	7.2±0.3	170.4±1.8	62.5±0.3
15	3.7	5.7±0.2	73.1±1.2	7.2±0.2	178.1±2.9	62.7±0.5
20	2.8	5.7±0.2	75.7±0.6	7.3±0.2	145.2±3.2	64.7±1.1

Table 5.4-Overall performance of two-stage digestion

	Overall two-stage IBR digestion
Hydrogen production unit	
optimal HRT(days)	3
OLR(g-COD/l-d)	32.9
optimal pH	4.8-5.3
Temperature (°C)	60
hydrogen yield ($ml\text{-}H_2/g\text{-}COD_{removed}$)	160.7
hydrogen production rate($ml\text{-}H_2/l\text{-}d$)	2358.3
COD removal (%)	44.6
Methane production unit	
methane yield ($ml\text{-}CH_4/g\text{-}COD_{removed}$)*	178.1
methane production rate($ml\text{-}CH_4/l\text{-}d$)	483.3
COD removal (%)*	73.1
Overall gas production ($ml\text{-}H_2$:$ml\text{-}CH_4$ /COD_{loaded})*	71.7:61.0
Overall COD removal (%)*	88.2

*: Calculation based on 15 days HRT in the methane IBR.

Table 5.5-Comparison of two-stage digestion to some other studies

	Two-stage IBRs (present study)	Two-stage CSTR+UASB[13]	Two-stage UASBs [31]	Two-stage UASBs[14]
Substrate	DPW	Cheese whey powder	Cheese Whey	Cassava wastewater
Temperature (°C)	60	37	35	55
HRT(days)	3	0.25	1	-
OLR(g-COD/l-d)	32.9	182	47.4	12
Hydrogen production unit				
Digester size (L)	60	2(CSTR)	3	4
hydrogen yield (ml-H_2/g-COD removed)	160.7	-	41	169
hydrogen production rate(ml-H_2/l-d)	2358.3	25000	2510	-
COD removal (%)	44.6	-		35
Methane production unit				
Digester size (L)	60	0.79(UASB)	15	24
methane yield (ml-CH_4/g-COD $_{removed}$)*	178.1	210	-	164.8
methane production rate(ml-CH_4/l-d)	483.3	-	-	-
COD removal (%)*	73.1			72
Overall gas production (ml-H_2:ml-CH_4 /COD $_{loaded}$)*	71.7:61.0	-	-	-
Overall COD removal (%)*	88.2	82	94	86.4

*: Calculation based on 15 days HRT in the methane IBR

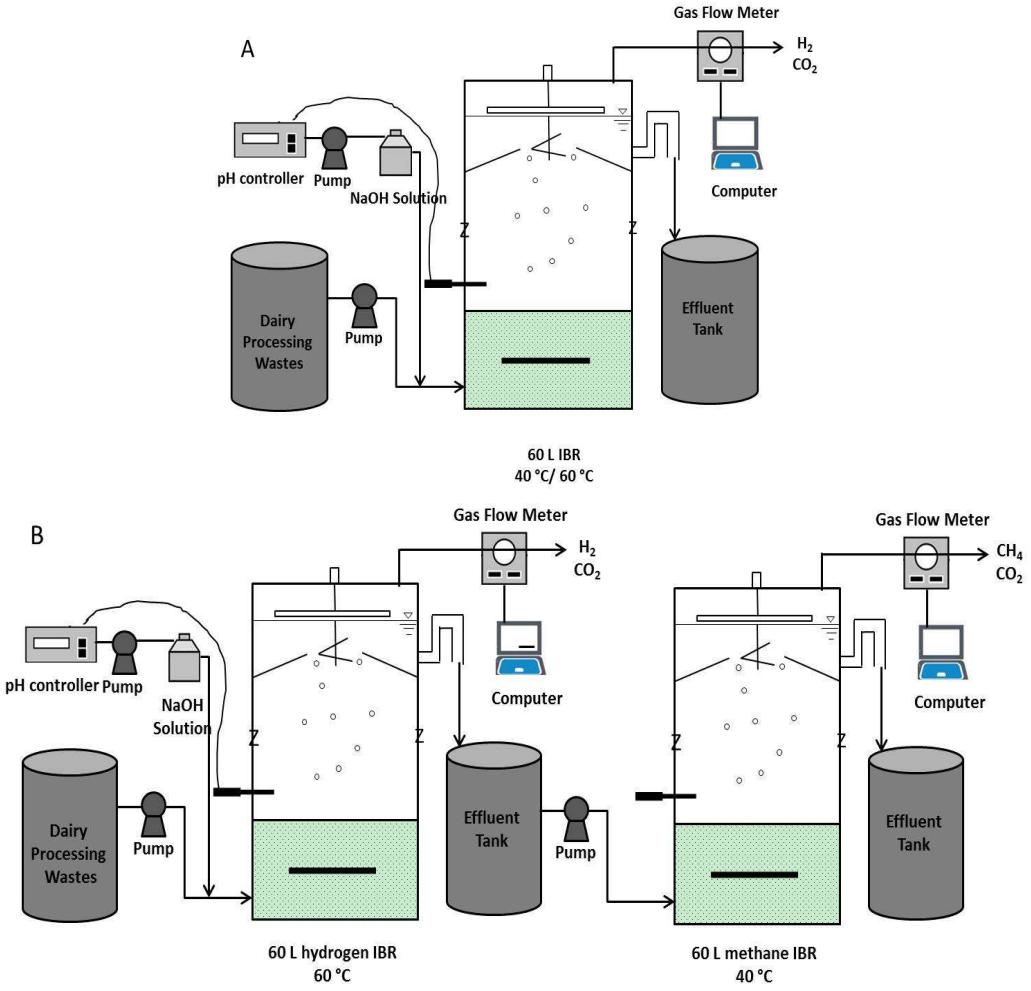

Figure 5.1-Schematic of single-stage and two-stage induced bed reactor (IBR) digestion. (A) single-stage IBR digestion. (B) two-stage IBR digestion

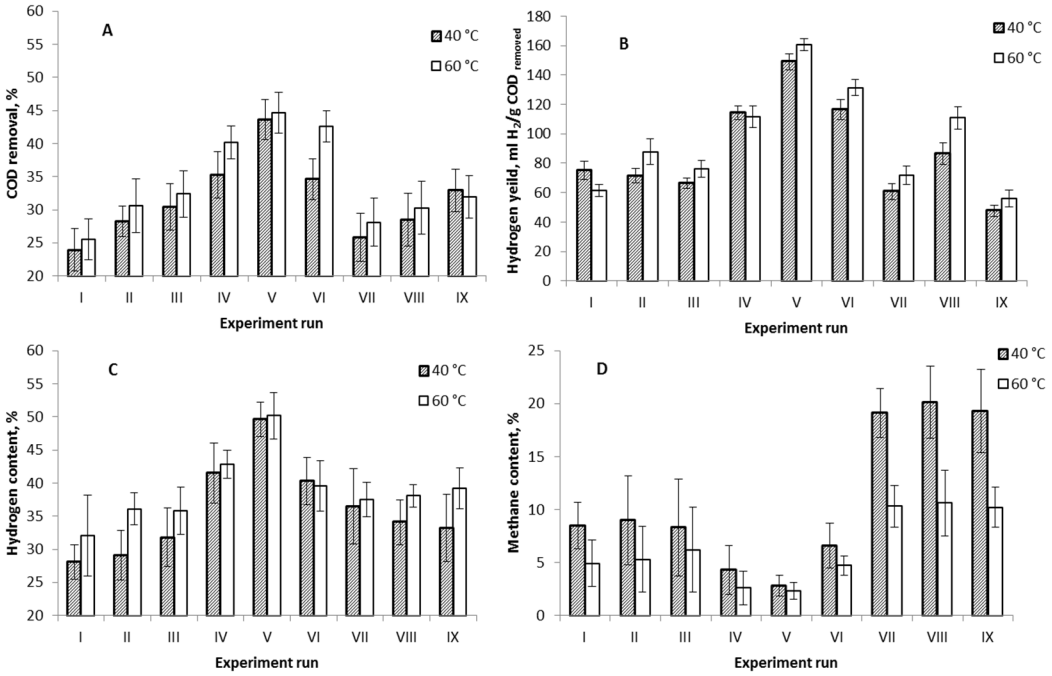

Figure 5.2-Results of (A) COD removals, (B) hydrogen yields, (C) hydrogen content and (D) methane content in nine experiment runs. Error bar represents standard deviation.

CHAPTER 6

MODELING OF ANAEROBIC HYDROGEN PRODUCTION FROM DAIRY PROCESSING WASTE USING A MODIFIED ADM1

Abstract

In this study, a mathematic model was built and implemented in R based on Anaerobic Digestion Model No. 1 (ADM1) for predicting and describing the anaerobic hydrogen production process. Modifications in the ADM1 include changes in biochemical process rate, inhibition factors, and dynamic parameters. The modified ADM1 was then validated by comparing the predictions with observations of anaerobic hydrogen production from dairy processing waste. The model successfully predicted hydrogen production, hydrogen content, methane content, VFA concentration, and digestion system stability. This research provides a useful mathematical model to investigate anaerobic hydrogen production process and stability.

Keywords:

Hydrogen production, Dairy processing waste (DPW), Anaerobic digestion model No.1 (ADM1)

Introduction

The technology of anaerobic digestion for biogas production was established a long time ago and is now widely applied. But the process is not fully understood due to

the complexity of microbial metabolism. For example, hydrogen anaerobic digestion, which is a promising method to produce hydrogen economically, faces several problems due to limited understanding of its microbial metabolism. An accurate mathematical model is needed to analyze and further understand the microbial metabolism process, especially in hydrogen anaerobic digestion, which is very attractive for future hydrogen production but faces several limitations at present. A few models have been developed to describe the hydrogen production process, but all are limited in scope. The Monod model was used to describe the relationship between the organic substrate degradation rate and the growth rate of hydrogen-producing bacteria [1]. The Andrews model is usually used to show the impacts of pH on the specific hydrogen production rate, although it is sometimes also used to describe the effects of temperature on the hydrogen production process [2-5]. A modified Gompertz model was specifically developed to examine the batch hydrogen fermentation process [6, 7]. The Luedeking–Piret model and its modified version were developed to describe the correlation between hydrogen production rate and the growth rate of hydrogen-producing bacteria [8, 9]. However, none of the above models describes the whole process of hydrogen production and the effects of inhibition factors, such as hydrogen partial pressure and fatty acid concentration.

Since the International Water Association in 2002 developed the anaerobic digestion model No. 1 (ADM1), this model has attracted wide attention in the field of research and practical application of anaerobic digestion[10, 11]. The ADM1 is a mathematical model that is often used as a framework that investigators can modify and

choose coefficients according to their specific substrates and digester. The reactions occurring in anaerobic digestion are very complex because of many sequential and parallel steps. The ADM1 divides those reactions into two main types during model development: biochemical reactions and physicochemical reactions[10, 11].

(1) Biochemical reactions

Microorganisms play a key role in this process. The ADM1 starts the biochemical reactions at hydrolysis; that is, the conversion of complex organic compounds to carbohydrates, proteins and lipids, and further to simple sugars, amino acids and long-chain fatty acids. This process is treated as first order kinetics in ADM1and is rate-limiting in the model development. Acidogenesis and methanogenesis are also included in the model.

(2) Physicochemical reactions

The ADM1 model also describes gas-liquid transfer and ion association and dissociation. An additional reaction, not included in ADM1 is precipitation.

Implemented as a differential equation system, the ADM1 model describes 19 processes and 24 components (Table A-2 and A-3). Inhibition functions contain pH (all groups), hydrogen (acetogenic groups) and free ammonia (aceticlastic methanogens). In this research, a mathematic model was built based on ADM1. The modified model was tested by comparing simulations to experimental observations. The objective of this

research was trying to establish a tool to describe, monitor and predict the anaerobic hydrogen process from dairy processing waste (DPW).

Materials and Methods

Digestion Experiment

The simulation experiments were done in two 60 L duplicate Induced Bed Reactors (IBRs). Three different parameters: temperature, pH, and HRT were examined. One IBR was set at the temperature of 40 °C and another was set at 60 °C. Three pH ranges (4.0-4.5, 4.8-5.3 and 5.5-6.0) and three HRT values (1, 3 and 5 days) were tested in a 3-factor full factorial design. Each digestion run lasted at least one week to obtain stable hydrogen production. The DPW characteristics (Table A-1), digester set-up, digestion operation and measurements methods can be found in a previously published paper [12].

Modeling Approach

In this study, ADM1 physicochemical processes were implemented as algebraic equations. Differential and (implicit) algebraic equations (DAE) were established to describe the change of 24 components' concentrations during 19 processes. Those equations were built based on the following mass balance:

$$\frac{dS_{liq,i}}{dt} = \frac{q_{in} * S_{liq,i}}{V_{liq}} - \frac{S_{liq,i} * q_{out}}{V_{liq}} + \sum_{j=1-19} \rho_j * \nu_{i,j}$$

where S is the concentration of a constituent, q is the flow rate, V is the digester volume, ρ are the rates of the processes that affect S, and ν is the stoichiometric coefficient for the constituent in each of those processes. Details of the nomenclature and units for each term are listed in Table 1.

The process rates (include biochemical process rates, acid-base rates and gas transfer rates), process inhibition, as well as differential equations, were written in R code and then implemented in R programming software (version 3.2.3) [13].

In order to predict and describe anaerobic hydrogen production instead of methane production, modifications were made to the original ADM1. The modifications were made in three aspects: biochemical process, process inhibitors, and parameters. It was assumed that methanogens were killed or inactivated during seed preparation and hydrogen production process. Thus, aceticlastic methanogenesis (uptake acetate to produce methane) and reductive methanogenesis (uptake hydrogen to produce methane) were negligible. Biochemical process rate for the uptake of acetate (process rate #11 in

the original ADM1) was removed, which leads to changes in the differential equations in the expression of methane, acetate, and inorganic nitrogen (Table 3). Also, the biochemical process rate for the uptake of hydrogen (process rate #12 in the original ADM1) was removed, which leads to changes in the differential equations for expression of methane, hydrogen, inorganic carbon, and inorganic nitrogen.

The reported optimal pH for hydrogen production is around 5.3 [14-16]. Based on our previously experiments of hydrogen production from DPW, hydrogen yield was significantly lower when pH was in the ranges of <4.3 or >5.8. Thus, additional pH inhibition was added when the pH was in the range of <4.3 or >5.8.

$$I_{pH\_N} = \begin{pmatrix} e^{-3*\left(\frac{pH-4.3}{1.5}\right)^2} & when\ pH < 4.3 \\ e^{-3*\left(\frac{5.8-pH}{1.5}\right)^2}, & when\ pH > 5.8 \\ 1, & else \end{pmatrix}$$

High hydrogen partial pressure and volatile fatty acids (VFA) concentration inside the digester have a negative effect on fermentation through feedback inhibition on the microbial hydrogen production process. Moreover, high hydrogen partial pressure not only affects hydrogen production but also triggers a shift of metabolic pathways towards the accumulation of acetate, ethanol, acetone, and butanol [17-19]. Thus, an inhibition factor for total VFA and an inhibition factor for hydrogen partial pressure were developed and added.

$$I_{vfa\_N} = \frac{1}{1 + S_{vfa}/K_{i\_vfa}}$$

where S_{vfa} is the total VFA concentration, kg COD / m^3 ;

K_{i_vfa} is the VFA inhibition factor, kg COD / m^3.

$$I_{h2\_N} = \frac{1}{1 + p_{gas\_h2}/K_{i\_h2}}$$

where p_{gas_h2} is the hydrogen partial pressure, bar;

K_{i_vfa} is the hydrogen partial inhibition factor, bar.

These additional inhibition factors affected the mass balances for methane, hydrogen, acetate, inorganic carbon, long chain fatty acids, valerate, propionate, butyrate, and valerate & butyrate degraders. The details of modifications in each component's differential equations are presented in Table 3.

The original ADM1's dynamic parameters or constants were based on the anaerobic digestion of sewage sludge substrate. Most of them were modified in order to more accurately model digestion of dairy processing waste. The modified parameter values are listed in the Appendix (Table A-4, A-5, and A-6).

Results and Discussion

The simulation experiments were carried out for hydrogen production based on different temperature, pH, and HRT. Values of temperature, pH, HRT and OLR in each experimental run are listed in Table 2. The modified ADM1 model was run to predict hydrogen production, hydrogen content, methane content, total VFA concentration, and stability. Unlike municipal sewage sludge, DPW has much higher carbohydrate content and lower protein and fat content [14]. Thus, the modified model assumed that the COD was divided to 75% carbohydrates, 15% proteins, 8% lipids, and 2% inert (non-

biodegradable) in DPW compared to 20% carbohydrates, 20% proteins, 25% lipids, and 35% inert in municipal sewage sludge. The experimental digester was operated until hydrogen content and methane content reached stable hydrogen production stage at which time data collection began for purposes of model/observations comparison. The results are shown in Figures 6.1, 6.2, 6.3, 6.4 and 6.5, respectively.

Hydrogen Yield Prediction

The model was run for each experiment's different operational conditions in Table 2. The output of predicted total gas production was reported as m^3 per day; the outputs of hydrogen, methane, and carbon dioxide gas were listed as partial pressure (bar) in the model. Predicted hydrogen yield; shown as ml H_2/g-COD $_{removed}$, was calculated based on the COD loading (Figure 6.1) and the above predictions (total gas production and hydrogen partial pressure). The results of observed and predicted hydrogen yield in nine experiment runs are shown in Figure 6.1.

The modified ADM1 model predicted hydrogen yield under both mesophilic 40 °C and thermophilic temperatures 60 °C. The observed hydrogen yields in the nine runs were in the range of 42-165 ml H_2/g-COD $_{removed}$. The predicted values were all within this range. This indicates that the modified ADM1 model was suitable for predicting hydrogen production. Figure 6.1 also shows the range of predictions (84-152 ml H_2/g-COD $_{removed}$) was smaller than that in observations (42-165 ml H_2/g-COD $_{removed}$), which could be explained by the complexity and uncertainty in actual digestions. The model overestimated the hydrogen yield except in experiments V and VIII. When considering

the model performance in different pH ranges, predictions within the pH range of 4.8-5.5 (experiments IV, V, and VI) were more accurate than those in pH ranges of 4.0-4.5 or 5.6-6.0, as indicated by the smaller value and equal distribution of residuals at the range of 4.8-5.5. There are possibly other unknown inhibitions in the pH range of 4.0-4.5 and 5.6-6.0 that the modified model doesn't reflect.

As shown in Figure 6.2, the model also shows better prediction in hydrogen production when the OLR was 32.9 g-COD/l-d (experiments II, V, and VIII) with 3 days HRT. The residuals at this ORL had smaller values and more even distribution compared to the other two COD loadings. It was also noticed that this modified ADM1 was very sensitive to the HRT, because when the HRT increased to 5-10 days, the hydrogen yield was significantly reduced (data not shown).

Hydrogen Content Prediction

The results of observed and predicted hydrogen content in nine experiment runs are shown in Figure 6.2. The model overestimated the hydrogen content in experiments I, II, III, VIII, and IX, while underestimating it in experiment V. The model predicted well in experiments IV, VI, and VII. The hydrogen content predictions were all in the small range of 36-48% while the observations varied from 25% to 55%. In the simulation experiments, the digesters were fed semi-continuously (every 4 hours) and gas samples were collected randomly at different times. Samples were either collected right after feeding, hours after feeding or right before feeding. We observed that hydrogen concentrations were much higher at between 30 minutes and one hour after feeding than

that at two hours after feeding. However, the model assumed the intakes were continuous, and thus, couldn't reflect this variation in semi-continuous feeding. Previous results showed temperature had no significant impact on hydrogen content [12]. The model verified this, as indicated by no significant difference in predicted hydrogen content between 60 °C and 40 °C (ANOVA, $p>0.05$, data not shown).

Methane Content Prediction

Methane concentration is an important indicator of the methanogens growth in hydrogen digestion. Growing methanogens may have rapidly consumed the produced hydrogen, resulting in a low hydrogen yield [20]. The results of observed and predicted methane content in the nine experimental runs are shown in Figure 6.3. Overall predicted methane concentrations were lower than observed except for experimental runs IV and V. All the predictions were below 10% methane, whereas observations had higher variations. The reason could be the feeding and sampling schedule during the experiment as discussed above for hydrogen content. Figure 6.3 shows the model predicted better in 4.8-5.5 pH range than in 4.0-4.5 and 5.6-6.0. The model also shows that the methane concentrations were higher in the pH range of 5.6-6.0 than the other two ranges, which was in accord with the experimental observations [12]. Both predictions and observations proved that methanogenesis was less suppressed in the pH range of 5.6-6.0. Growing methanogens may have rapidly consumed the produced hydrogen, which led to a low hydrogen yield. The residuals analysis showed that the model predicted methane content better at 60°C than at 40°C.

Total VFA concentration and stability prediction

The total VFA concentrations were measured and predicted at four different stages – before start of the stage (DPW feeding started but hydrogen hadn't been produced), start of stage (hydrogen started to be produced and the production was increasing), stable stage (hydrogen was produced stably) and collapse stage (hydrogen production decreased or stopped). The results shown in Figure 6.4 are the average measurements/predictions in nine experiments both at 60 °C and 40°C. As shown in Figure 6.4, the measured VFA concentrations increased over the four stages of hydrogen production. The model predicted a VFA peak at the start stage where the observations didn't show this high of level of VFA. The authors couldn't find a good solution to avoid this peak in the model and this is one aspect that could be improved in the future. The model did successfully predict the highest measured VFA peak (~5,000 mg/L as HOAc) when failure occurred. According to the model, the collapse was predicted to happen during the second peak when the VFA concentration was close to 5,100 mg/L (as HOAc). This can help to predict the time when collapses occur and to take actions to avoid collapse.

The results of observed and predicted stability (shown in days until collapse) in nine experimental runs are shown in Figure 6.5. All model predicted values showed collapse events sooner than observed values. The largest underestimation was found in experiment V. The observed stable hydrogen production days were on average 4.5 days longer that the model predictions. It is probably attributed to the IBR digester that we

used in the simulation experiments. The model assumed the digestion was under simple complete mixed reactor condition (CSTR). However, the IBRs used here has the advantage to effectively capture the bacteria/sludge in the bottom to form a thick active bed [21-23]. This special design may help to lessen the negative effect of high level accumulated VFA. Similar results were observed when using IBR to digest bakery waste for methane production (paper in preparation) - the actual failure days were much later than the original ADM1 predicted failure days.

Conclusions

The modified ADM1 successfully predicted reasonable hydrogen production, hydrogen content, methane content, VFA concentration, and stability. We found that this model predicted hydrogen production and methane content best in the pH range of 4.8-5.5. At 60 °C the model more accurately predicted methane content than at 40 °C. Overall the model tended to underestimate the hydrogen and methane content because it didn't consider the semi-continuous feeding used experimentally. More importantly, this model accurately predicted collapse to happen when the second peak of VFA concentration occurs (close to 5,100 mg/L). This information makes the model a useful tool in the investigation of anaerobic hydrogen production and will help in applying this technology at large-scale.

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Table 6.1- Nomenclature and units used in this paper

Symbol	Description	Units
i	Component index (Appendix A-2 and A-3)	N/A
j	Process index (Appendix A-2 and A-3)	N/A
Si	Soluble component *i* concentration	kg COD m^{-3}
Xi	Particle component *i* concentration	kg COD m^{-3}
Si_in	Soluble component *i* input concentration	kg COD m^{-3}
Xi_in	Particle component *i* input concentration	kg COD m^{-3}
ρ_j	Rate for process *j*	kg COD m^{-3}
t	Time	day
q_{in}	Flow rate	m^3 day^{-1}
V_{liq}; V_{gas}	Liquid volume of digester; Gas volume of digester	m^3
λ	Hydraulic retention time (Vliq/Q)	day
I	Inhibition function(various, see Table)	N/A
Ni	Nitrogen content of component i	Kmole N/kg COD
Ci	Carbon content of component i	kmole C/kg COD
$Y_{substrate}$	Yield of biomass on substrate	(kgCOD_X) m^{-3}(kg COD_S) m^{-3}
$F_{product\_substrate}$	Yield of product on substrate	N/A
s*j*	Sum of carbon rate coefficients in process j	N/A
$\nu_{i,j}$	rate coefficients for component i in process j	kg COD $(m^3)^{-1}$
p_{gas}	Pressure of gas	bar
$k_{A/B,i}$	Acid-base rate constant for component i	kmole $(m^3)^{-1}$ day^{-1}
k_{dec}	First order decay rate for biomass death	day^{-1}
k_La	Gas-liquid transfer coefficient	day^{-1}
k_m	Specific Monod maximum uptake rate	(kgCOD_X) m^{-3}*(kg COD_S) m^{-3}* day^{-1}
K_a	Acid-base equilibrium constant	kmole m^{-3}
K_H	Henry's law constant	kmole m^{-3}* bar^{-1}

Table 6.2-Values of temperature, pH, HRT and OLR in each experiment run

Experiment run number	Temperature (°C)	pH	HRT (days)	OLR (g-COD/l-d)
I	40 or 60	4.0-4.5	1	98.7
II	40 or 60	4.0-4.5	3	32.9
III	40 or 60	4.0-4.5	5	19.74
IV	40 or 60	4.8-5.3	1	98.7
V	40 or 60	4.8-5.3	3	32.9
VI	40 or 60	4.8-5.3	5	19.74
VII	40 or 60	5.5-6.0	1	98.7
VIII	40 or 60	5.5-6.0	3	32.9
IX	40 or 60	5.5-6.0	5	19.74

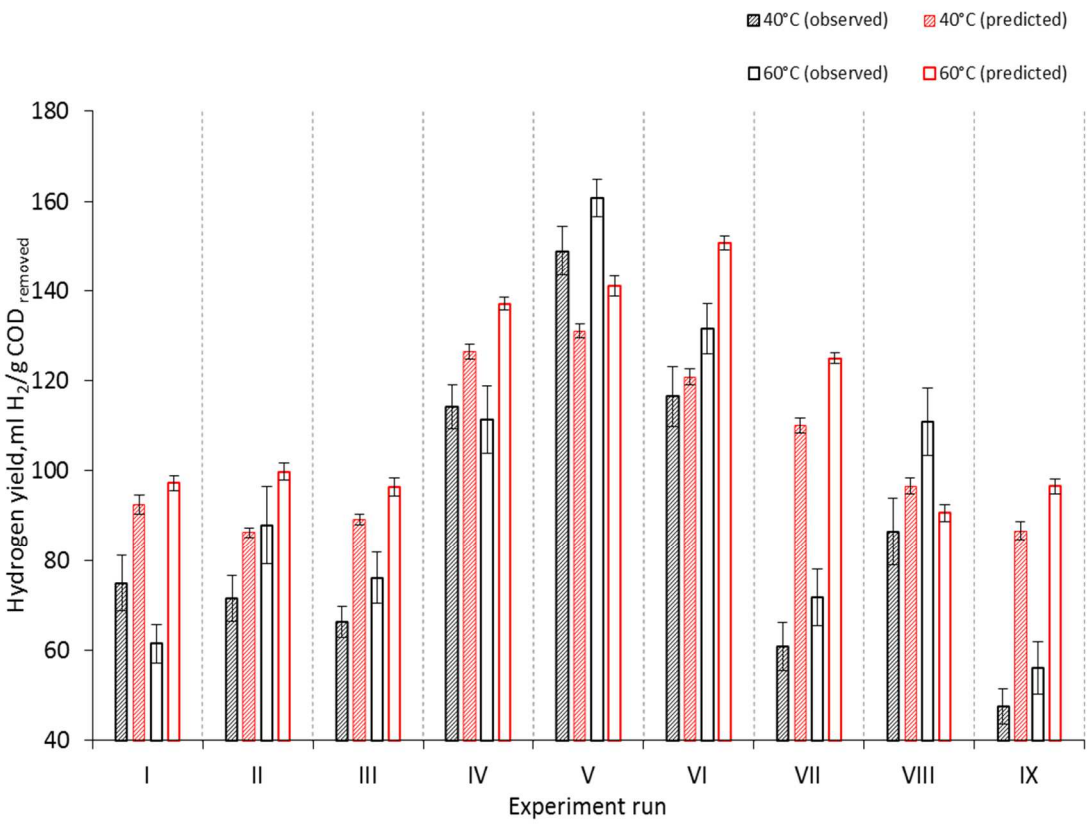

Figure 6.1- Results of observed and predicted hydrogen yield in nine experiment runs. Observed error bars represent standard deviation from 8 observations at each set of conditions. Predicted error bars represent standard deviation of the model hydrogen production outputs.

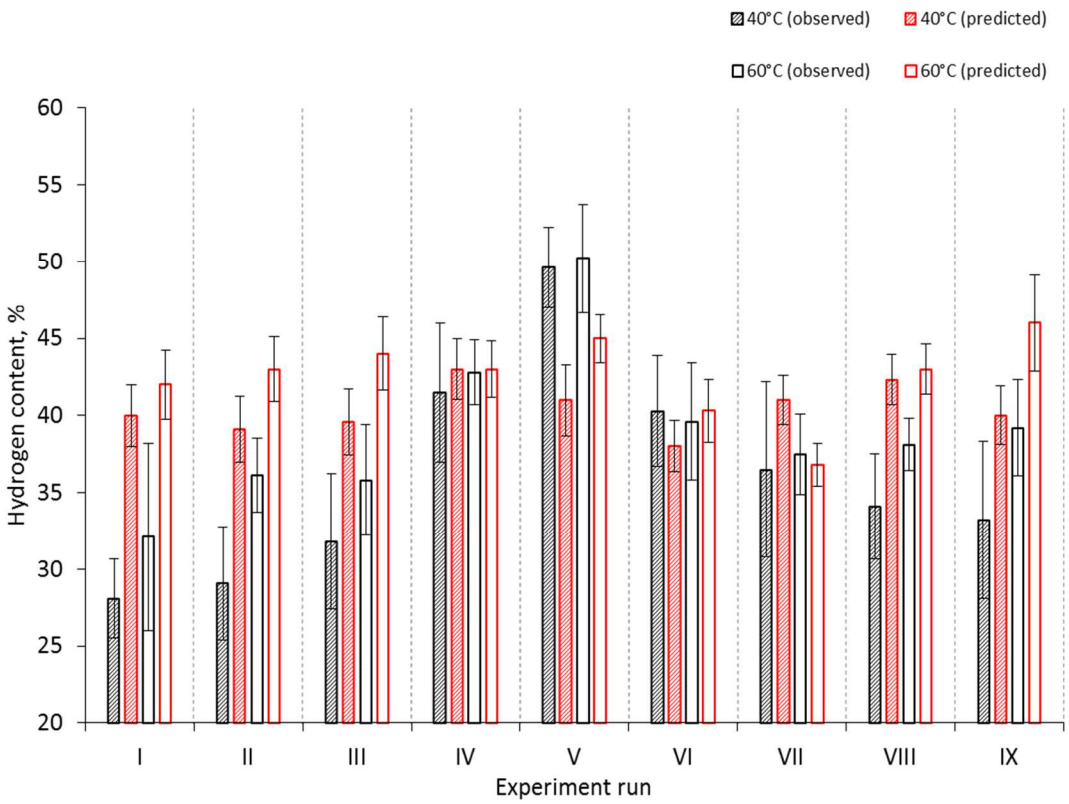

Figure 6.2- Results of observed and predicted hydrogen content in nine experiment runs. Observed error bars represent standard deviation from 16 observations at each set of conditions. Predicted error bars represent standard deviation of the model hydrogen concentration outputs.

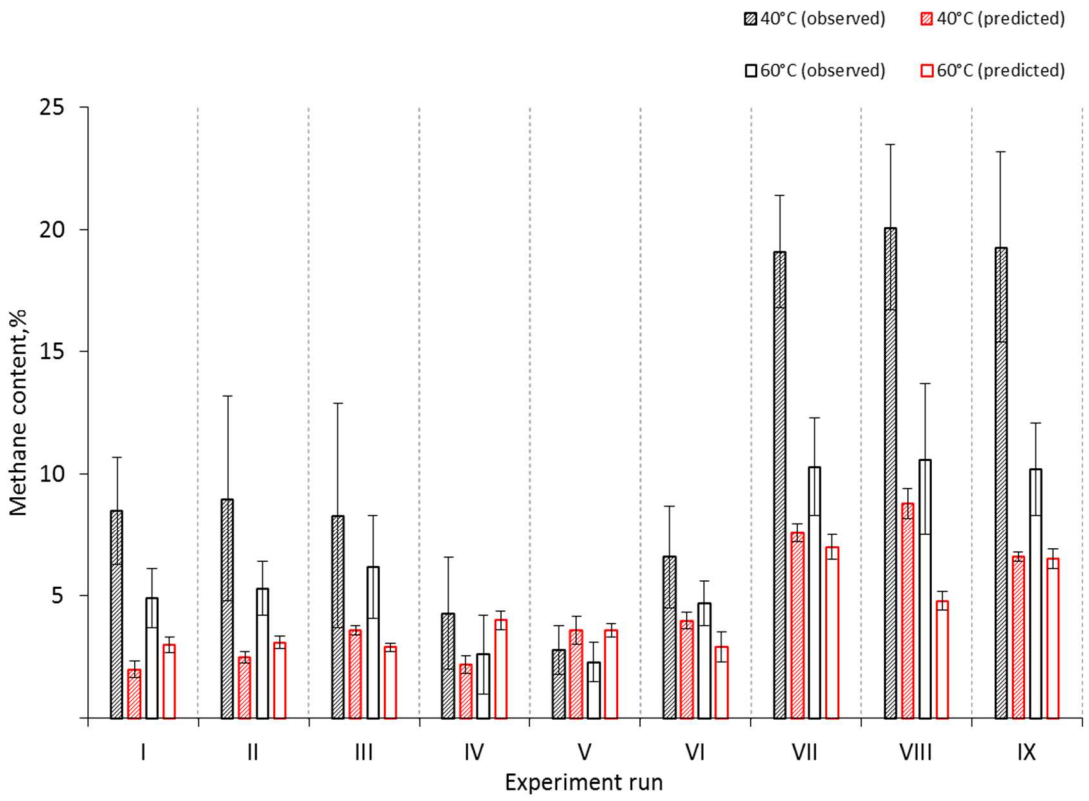

Figure 3- Results of observed and predicted methane content in nine experiment runs. Observed error bars represent standard deviation from 16 observations at each set of conditions. Predicted error bars represent standard deviation of the model methane content outputs.

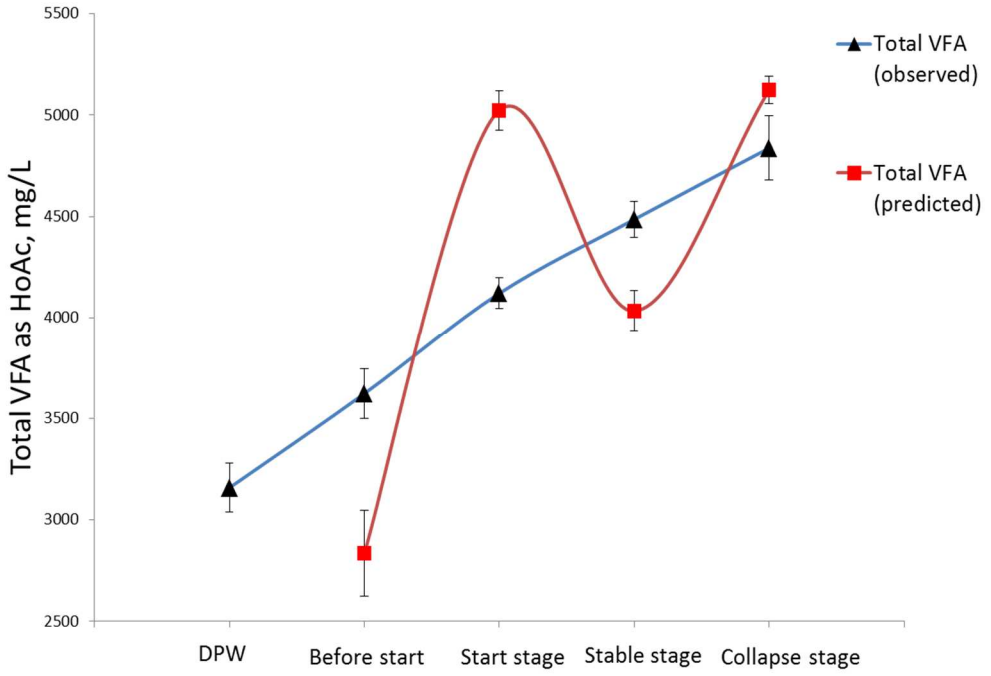

Figure 6.4 - Results of observed and predicted total VFA concentration in nine experiment runs. Observed error bars represent standard deviation from 27 observations at each stage; predicted error bars represent standard deviation from 9 experiments runs at each stage.

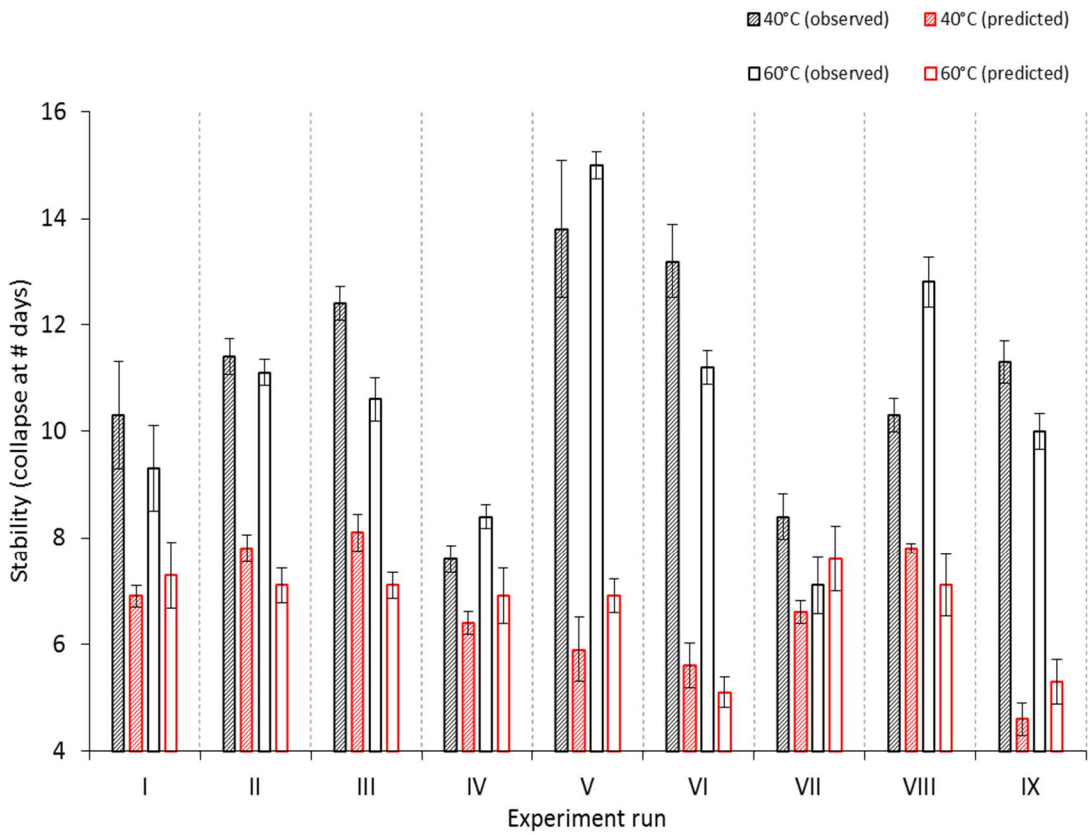

Figure 6.5– Results of observed and predicted stability in nine experiment runs. Observed error bars represent standard deviation from 3 observations at each set of conditions. Predicted error bars represent standard deviation of the model predicted time when stable hydrogen production stopped.

CHAPTER 7

CONCLUSIONS

This research first tried to find an effective method for stable methane production from DPW without adding chemicals. Under the conditions of the study, commingling algae or grass with DPW made it possible to avoid the addition of pH control chemicals. When treated alone, COD removal from algae was about 45% with a hydraulic retention time (HRT) of 24 day and specific SLR of 0.9 g total solids (TS) $L^{-1}d^{-1}$. Adding up to about 92% (solids basis) DPW that included hard and soft cheese whey and milk processing and yogurt waste (COD = 107 g L^{-1}) to the algae improved COD removal to as high as 87% with SLR = 2.3 g L^{-1} d^{-1}. Under these conditions, biogas yield was 0.37 L (SATP (T = 25°C P = 100 kPa)) g-1 of COD loaded. The pH of commingled influent was 3.5 – 5.4. When algae were no longer available, fresh grass clippings were slurried and commingled with DPW. Adding 1.61% grass to DPW (solids basis) resulted in COD removal of 94% with SLR = 1.21 g $L^{-1}d^{-1}$. Biogas yield was 0.37 L (SATP) g^{-1} of COD loaded.

Optimization of anaerobic hydrogen production from DPW was explored in semi-continuous pilot-scale (60 L) IBRs. We found the optimal conditions for hydrogen production from DPW were: HRT 3 days / OLR 32.9 g-COD/l-d, pH range of 4.8-5.5, and 60 °C. Under these conditions, the highest hydrogen yield was 160.7 ml/g-COD removed, highest COD removal was 44.6% and highest hydrogen content 50.2% was

achieved. Two-stage digestions were tested later for further energy extraction and COD removal. Results show two-stage production of hydrogen and methane can greatly increase the amount of energy harvested and will increase COD removal. With an HRT of 15 days in the methane IBR, methane yield was 178.1 ml methane per gram COD removed and COD removal was 73.1%. The overall gas production in two-stage digestion was 71.7 ml H_2 and 61.0 ml CH_4 per gram COD loaded. And the overall COD removal was as high as 88.2%. This study demonstrated that the production of both hydrogen and methane can be efficiently coupled in a two-stage IBR digestion system. The pilot-scale research here provides the data and design requirements for full-scale application.

The modified mathematical model successfully predicted hydrogen production, hydrogen and methane concentration, VFA concentration and stability. We found that this model predicted hydrogen production and methane concentration better in the pH range of 4.8-5.5. The model predicted methane concentration more accurately at 60 °C than at 40 °C. Overall the model underestimated the hydrogen and methane concentrations because the model did not consider semi-continuous feeding. More importantly, this model predicted collapse; which happens when the second peak of VFA concentration occurs (close to 5,100 mg/L). Thus, the model provides a good tool to predict collapse. It will help bring this technology; anaerobic hydrogen production, to large scale commercialization.

Engineering significance

This research provides a new method for the dairy industry to not only manage wastes but also produce clean energy (hydrogen). The applications will, however, not be limited to the dairy industry. The methods and conclusions of this study might also be applicable to management of many other wastes with high organic content; for example, meat processing waste, sugar processing waste, agricultural wastes like manure, and wastewater sludge.

For decades, anaerobic digestion has proved to be an effective method to reduce pollution and produce methane. However, to date, there are relatively few digesters being installed in the United States. One major reason for the limited number of anaerobic digesters is the minimal return on investment. The value of methane is low and most of the biogas produced by anaerobic digestion generally is used to generate electricity. Anaerobic digesters are expensive to build and commodity-priced electrical generation is not producing enough revenue to achieve a decent rate of return on investment. The anaerobic technology developed here will produce significant hydrogen and methane. Hydrogen is a much more valuable gas than methane.

The produced hydrogen is not limited to fuel cells technology. A mixture of hydrogen and methane as a fuel can be much more valuable than using methane alone. Thus, the technology developed here may provide a good means of renewable and clean energy production in facing a shortage of fossil fuels in the future.

The model developed here may help in the practical application of anaerobic hydrogen production technology. It can give operators a tool to monitor anaerobic digesters when producing hydrogen. Operators may know what happens inside the digester so that they can take actions before the digester collapses. Also, the model provides a tool for engineers to potentially improve anaerobic hydrogen production technology.

APPENDICES

Appendix A

Supplementary information for Chapter 5: OPTIMIZATION OF ANAEROBIC HYDROGEN AND METHANE PRODUCTION FROM DAIRY PROCESSING WASTE USING A TWO-STAGE DIGESTION IN INDUCED BED REACTORS (IBR).

Table A-1 Results of analysis of variance (ANOVA)

	COD removal p-value	Hydrogen yield p-value	Hydrogen content p-value	Methane content p-value
Temperature	0.048	0.034	0.061	0.004
pH	4.28e-6	3.58e-7	8.91e-5	8.76e-6
HRT/OLR	0.013	0.001	0.195	0.841

Appendix B

Supplementary information for Chapter 6: MODELING OF ANAEROBIC HYDROGEN PRODUCTION FROM DAIRY PROCESSING WASTE USING A MODIFIED ADM1.

Table B-1 Characteristics of dairy processing waste

Dairy Processing Waste	
pH	4.5±0.6
Total COD (g/L)	98.7±4.0
Soluble COD (g/L)	75.6±2.6
Total Solids (TS, g/L)	61.1±4.8
Volatile Solids (VS, g/L)	55.3±3.7
Total Organic Carbon (TOC, g/L)	24.8 ± 3.3
Total nitrogen (g/L)	4.06±1.21
NH_3-N(g/L)	2.30±0.81

Table B-2 Biochemical rate coefficients ($v_{i,j}$) and kinetic rate equations (ρ_j) for soluble components (i = 1 – 12, j = 1 – 19)[10]

	Component → i	1	2	3	4	5	6	7	8	9	10	11	12	Rate (ρ_j, kg COD.m^{-3}.d^{-1})
j	Process ↓	S_{su}	S_{aa}	S_{fa}	S_{va}	S_{bu}	S_{pro}	S_{ac}	S_{h2}	S_{ch4}	S_{IC}	S_{IN}	S_I	
1	Disintegration												$f_{sI,xc}$	$k_{dis}X_c$
2	Hydrolysis carbohydrates	1												$k_{hyd,ch}X_{ch}$
3	Hydrolysis of proteins		1											$k_{hyd,pr}X_{pr}$
4	Hydrolysis of lipids	$1-f_{fa,li}$		$1-f_{fa,li}$										$k_{hyd,li}X_{li}$
5	Uptake of sugars	−1				$(1-Y_{su})f_{bu,su}$	$(1-Y_{su})f_{pro,su}$	$(1-Y_{su})f_{ac,su}$	$(1-Y_{su})f_{h2,su}$		$-\sum_{i=9,11-24} C_i v_{i,5}$	$-(Y_{su})\,N_{bac}$		$k_{m,su}\frac{S_{su}}{K_S+S}X_{su}I_1$
6	Uptake of amino acids		−1		$(1-Y_{aa})f_{va,aa}$	$(1-Y_{aa})f_{bu,aa}$	$(1-Y_{aa})f_{pro,aa}$	$(1-Y_{aa})f_{ac,aa}$	$(1-Y_{aa})f_{h2,aa}$		$-\sum_{i=1-9,11-24} C_i v_{i,6}$	$N_{aa}-(Y_{aa})\,N_{bac}$		$k_{m,aa}\frac{S_{aa}}{K_S+S_{aa}}X_{aa}I_1$
7	Uptake of LCFA			−1				$(1-Y_{fa})$ 0.7	$(1-Y_{fa})$ 0.3			$-(Y_{fa})\,N_{bac}$		$k_{m,fa}\frac{S_{fa}}{K_S+S_{fa}}X_{fa}I_2$
8	Uptake of valerate				−1		$(1-Y_{c4})$ 0.54	$(1-Y_{c4})$ 0.31	$(1-Y_{c4})$ 0.15			$-(Y_{c4})\,N_{bac}$		$k_{m,c4}\frac{S_{va}}{K_S+S_{va}}X_{c4}\frac{1}{1+S_{bu}/S_{va}}I$
9	Uptake of butyrate					−1		$(1-Y_{c4})$ 0.8	$(1-Y_{c4})$ 0.2			$-(Y_{c4})\,N_{bac}$		$k_{m,c4}\frac{S_{bu}}{K_S+S_{bu}}X_{c4}\frac{1}{1+S_{va}/S_{bu}}I$
10	Uptake of propionate						−1	$(1-Y_{pro})$ 0.57	$(1-Y_{pro})$ 0.43		$-\sum_{i=1-9,11-24} C_i v_{i,10}$	$-(Y_{pro})\,N_{bac}$		$k_{m,pr}\frac{S_{pro}}{K_S+S_{pro}}X_{pro}I_2$
11	Uptake of acetate							−1		$(1-Y_{ac})$	$-\sum_{i=1-9,11-24} C_i v_{i,11}$	$-(Y_{ac})\,N_{bac}$		$k_{m,ac}\frac{S_{ac}}{K_S+S_{ac}}X_{ac}I_3$
12	Uptake of hydrogen								−1	$(1-Y_{h2})$	$-\sum_{i=1-9,11-24} C_i v_{i,12}$	$-(Y_{h2})\,N_{bac}$		$k_{m,h2}\frac{S_{h2}}{K_S+S_{h2}}X_{h2}I_1$
13	Decay of X_{su}													$k_{dec,Xsu}X_{su}$
14	Decay of X_{aa}													$k_{dec,Xaa}X_{aa}$
15	Decay of X_{fa}													$k_{dec,Xfa}X_{fa}$
16	Decay of X_{c4}													$k_{dec,Xc4}X_{c4}$
17	Decay of X_{pro}													$k_{dec,Xpro}X_{pro}$
18	Decay of X_{ac}													$k_{dec,Xac}X_{ac}$
19	Decay of X_{h2}													$k_{dec,Xh2}X_{h2}$
		Monosaccharides (kgCOD·m^{-3})	Amino acids (kgCOD·m^{-3})	Long chain fatty acids (kgCOD·m^{-3})	Total valerate (kgCOD·m^{-3})	Total butyrate (kgCOD·m^{-3})	Total propionate (kgCOD·m^{-3})	Total acetate (kgCOD·m^{-3})	Hydrogen gas (kgCOD·m^{-3})	Methane gas (kgCOD·m^{-3})	Inorganic carbon (kmoleC·m^{-3})	Inorganic nitrogen (kmoleN·m^{-3})	Soluble inerts (kgCOD·m^{-3})	Inhibition factors: $I_1 = I_{pH}I_{IN,lim}$ $I_2 = I_{pH}I_{IN,lim}\cdot I_{h2}$ $I_3 = I_{pH}I_{IN,lim}\cdot I_{NH3,Xac}$

Table B-3 Biochemical rate coefficients ($\nu_{i,j}$) and kinetic rate equations (ρ_j) for soluble components (i = 13 – 24, j = 1 – 19)[10]

j	Component → i / Process ↓	13 X_c	14 X_{ch}	15 X_{pr}	16 X_{li}	17 X_{su}	18 X_{aa}	19 X_{fa}	20 X_{c4}	21 X_{pro}	22 X_{ac}	23 X_{h2}	24 X_I	Rate (ρ_j, kg COD.m^{-3}.d^{-1})
1	Disintegration	-1	$f_{ch,xc}$	$f_{pr,xc}$	$f_{li,xc}$								$f_{xl,xc}$	$k_{dis}X_c$
2	Hydrolysis carbohydrates		-1											$k_{hyd,ch}X_{ch}$
3	Hydrolysis of proteins			-1										$k_{hyd,pr}X_{pr}$
4	Hydrolysis of lipids				-1									$k_{hyd,li}X_{li}$
5	Uptake of sugars					Y_{su}								$k_{m,su}\frac{S_{su}}{K_S+S}X_{su}I_1$
6	Uptake of amino acids						Y_{aa}							$k_{m,aa}\frac{S_{aa}}{K_S+S_{aa}}X_{aa}I_1$
7	Uptake of LCFA							Y_{fa}						$k_{m,fa}\frac{S_{fa}}{K_S+S_{fa}}X_{fa}I_2$
8	Uptake of valerate								Y_{c4}					$k_{m,c4}\frac{S_{va}}{K_S+S_{va}}X_{c4}\frac{1}{1+S_{bu}/S_{va}}I$
9	Uptake of butyrate								Y_{c4}					$k_{m,c4}\frac{S_{bu}}{K_S+S_{bu}}X_{c4}\frac{1}{1+S_{va}/S_{bu}}I$
10	Uptake of propionate									Y_{pro}				$k_{m,pr}\frac{S_{pro}}{K_S+S_{pro}}X_{pro}I_2$
11	Uptake of acetate										Y_{ac}			$k_{m,ac}\frac{S_{ac}}{K_S+S_{ac}}X_{ac}I_3$
12	Uptake of hydrogen											Y_{h2}		$k_{m,h2}\frac{S_{h2}}{K_S+S_{h2}}X_{h2}I_1$
13	Decay of X_{su}	1				-1								$k_{dec,Xsu}X_{su}$
14	Decay of X_{aa}	1					-1							$k_{dec,Xaa}X_{aa}$
15	Decay of X_{fa}	1						-1						$k_{dec,Xfa}X_{fa}$
16	Decay of X_{c4}	1							-1					$k_{dec,Xc4}X_{c4}$
17	Decay of X_{pro}	1								-1				$k_{dec,Xpro}X_{pro}$
18	Decay of X_{ac}	1									-1			$k_{dec,Xac}X_{ac}$
19	Decay of X_{h2}	1										-1		$k_{dec,Xh2}X_{h2}$
		Composites (kgCOD·m^{-3})	Carbohydrates (kgCOD·m^{-3})	Proteins (kgCOD·m^{-3})	Lipids (kgCOD·m^{-3})	Sugar degraders (kgCOD·m^{-3})	Amino acid degraders (kgCOD·m^{-3})	LCFA degraders (kgCOD·m^{-3})	Valerate and butyrate degraders (kgCOD·m^{-3})	Propionate degraders (kgCOD·m^{-3})	Acetate degraders (kgCOD·m^{-3})	Hydrogen degraders (kgCOD·m^{-3})	Particulate inerts (kgCOD·m^{-3})	Inhibition factors: $I_1 = I_{pH}I_{IN,lim}$; $I_2 = I_{pH}I_{IN,lim}\cdot I_{h2}$; $I_3 = I_{pH}I_{IN,lim}\cdot I_{NH3,Xac}$

Table B-4 Stoichiometric parameter values used in modified ADM1

parameter	value	unit	parameter	value	unit
Ffa_li	0.95	N/A	Ni	0.003	Kmole N(kgCOD)$^{-1}$
Yaa	0.08	N/A	FsI_xc	0.1	N/A
Fva_aa	0.23	N/A	Fpr_xc	0.2	N/A
Ysu	0.1	N/A	Fch_xc	0.2	N/A
Fbu_su	0.1328	N/A	Fli_xc	0.3	N/A
Fbu_aa	0.26	N/A	Cxc	0.02786	Kmole C(kgCOD)$^{-1}$
Fpro_su	0.2691	N/A	CsI	0.03	Kmole C(kgCOD)$^{-1}$
Fpro_aa	0.05	N/A	Cch	0.0313	Kmole C(kgCOD)$^{-1}$
Yc4	0.06	N/A	Cpr	0.03	Kmole C(kgCOD)$^{-1}$
Fac_su	0.40755	N/A	Cli	0.022	Kmole C(kgCOD)$^{-1}$
Fac_aa	0.4	N/A	CxI	0.03	Kmole C(kgCOD)$^{-1}$
Yfa	0.06	N/A	Csu	0.0313	Kmole C(kgCOD)$^{-1}$
Ypro	0.04	N/A	Caa	0.03	Kmole C(kgCOD)$^{-1}$
Fh2_su	0.19	N/A	Cbu	0.025	Kmole C(kgCOD)$^{-1}$
Fh2_aa	0.06	N/A	Cpro	0.0268	Kmole C(kgCOD)$^{-1}$
Yac	0.05	N/A	Cac	0.0313	Kmole C(kgCOD)$^{-1}$
Yh2	0.06	N/A	Cbac	0.0313	Kmole C(kgCOD)$^{-1}$
Nbac	0.00625	Kmole N(kgCOD)$^{-1}$	Cva	0.024	Kmole C(kgCOD)$^{-1}$
Naa	0.007	Kmole N(kgCOD)$^{-1}$	Cfa	0.0217	Kmole C(kgCOD)$^{-1}$
Nxc	0.005	Kmole N(kgCOD)$^{-1}$	Cch4	0.0156	Kmole C(kgCOD)$^{-1}$
FxI_xc	0.2	N/A			

Table B-5 biochemical parameter values used in modified ADM1

parameter	value	unit	parameter	value	unit
Kdis	0.4	d^{-1}	Kdec_xaa	0.02	d^{-1}
Khyd_ch	10	d^{-1}	Kdec_xfa	0.02	d^{-1}
Khyd_pr	10	d^{-1}	Kdec_xc4	0.02	d^{-1}
Khyd_li	10	d^{-1}	Kdec_xpro	0.02	d^{-1}
Km_su	30	d^{-1}	Kdec_xac	0.02	d^{-1}
Ks_su	0.5	kg COD m^{-3}	Kdec_xh2	0.02	d^{-1}
Km_aa	50	d^{-1}	pHuL_aa	5.5	N/A
Ks_aa	0.3	kg COD m^{-3}	pHlL_aa	4	N/A
Km_fa	6	d^{-1}	pHuL_ac	7	N/A
Ks_fa	0.4	kg COD m^{-3}	pHlL_ac	6	N/A
Km_c4	20	d^{-1}	pHuL_h2	6	N/A
Ks_c4	0.2	kg COD m^{-3}	pHlL_h2	5	N/A
Km_pro	13	d^{-1}	Ks_in	1e-4	kg COD m^{-3}
Ks_pro	0.1	kg COD m^{-3}	Ki_h2_fa	5e-6	kg COD m^{-3}
Km_ac	8	d^{-1}	Ki_h2_c4	1e-5	kg COD m^{-3}
Ks_ac	0.15	kg COD m^{-3}	Ki_h2_pro	3.5e-6	kg COD m^{-3}
Km_h2	35	d^{-1}	Ki_nh3	0.0018	kg COD m^{-3}
Ks_h2	7e-6	kg COD m^{-3}	Ki_vfa	1.6e-6	kg COD m^{-3}
Kdec_xsu	0.02	d^{-1}	Ki_h2	2.7e-5	kg COD m^{-3}

Table B-6 Physicochemical parameter values used in modified ADM1

parameter	value	unit	parameter	value	unit
Ka_bva	1e10	M^{-1} d^{-1}	Ka_in	1.11e-9	kmole m^{-3}
Ka_bbu	1e10	M^{-1} d^{-1}	Kw	2.08e-14	M 10^{-14}
Ka_bpro	1e10	M^{-1} d^{-1}	KL	200	d^{-1}
Ka_bac	1e10	M^{-1} d^{-1}	Kh_h2	7.38e-4	M bar^{-1}
Ka_bco2	1e10	M^{-1} d^{-1}	Kh_ch4	0.00116	M bar^{-1}
Ka_bin	1e10	M^{-1} d^{-1}	Kh_co2	0.0271	M bar^{-1}
Ka_va	1.38e-5	M	kp	5e4	$M^3 d^{-1}$ bar^{-1}
Ka_bu	1.5e-5	kmole m^{-3}	R	0.083145	bar M^{-1} K^{-1}
Ka_pro	1.32e-5	kmole m^{-3}	Top	308.15	K
Ka_ac	1.74e-5	kmole m^{-3}	Patm	1.013	bar
Ka_co2	4.94e-7	kmole m^{-3}	Pgas_h2o	0.0557	bar

Appendix C

An example of R code in experiment run I at 40 °C.

(t:independent variable, state: list of state variables, par:constants)

```
ADM1_C<-function(t,state,parameters){

With (as.list(c(state,parameters)), {

#Algebraic equ.

    Snh4=Sin-Snh3

    Sco2=Sic-Shco3_m

    Z=Scation+Snh4-Shco3_m-Sac_m/64-Spro_m/112-Sbu_m/160-Sva_m/208-
Sanion

    Kw=(exp(55900/(R*100)*(1/Tbase-1/Top)))*(10^(-14))

    Sh=-Z*.5+.5*sqrt(Z^2+4*Kw)

    Svfa= Sac_m+Spro_m+ Sbu_m+Sva_m

    Pgas_h2=Sgas_h2*R*Top/16

#pH

     pH = -log10(Sh)
```

#inhibition factors

IpH_aa<- if (pH<pHuL_aa) exp(-3*((pH-pHuL_aa)/(pHuL_aa-pHlL_aa))^2) else 1

IpH_ac<- if (pH<pHuL_ac) exp(-3*((pH-pHuL_ac)/(pHuL_ac-pHlL_ac))^2) else 1

IpH_h2<- if (pH<pHuL_h2) exp(-3*((pH-pHuL_h2)/(pHuL_h2-pHlL_h2))^2) else 1

Iin_lim = 1/(1+Ks_in/Sin)

Ih2_fa = 1/(1+Sh2/Ki_h2_fa)

Ih2_c4 = 1/(1+Sh2/Ki_h2_c4)

Ih2_pro = 1/(1+Sh2/Ki_h2_pro)

Inh3 = 1/(1+Snh3/Ki_nh3)

IpH_N = if (pH<4.3) exp(-3*((4.3-pH)/1.5)^2) if (pH>5.8) exp(-3*((pH-pHuL_h2)/1.5)^2) else 1

Ih2_N = if (Pgas_h2>0.58) 1/(1+Pgas_h2/Ki_h2) else 1

Ivfa = if (Svfa>9.5) 1/(1+Svfa/Ki_vfa) else 1

I5=I6=IpH_aa*Iin_lim

```
        I7=IpH_aa*Iin_lim*Ih2_fa*Ih2*Ivfa

        I8=I9=IpH_aa*Iin_lim*Ih2_c4*Ih2*Ivfa

        I10=IpH_aa*Iin_lim*Ih2_pro*Ih2*Ivfa

        I11=IpH_ac*Iin_lim*Inh3

        I12=IpH_h2*Iin_lim

#process rates

          P1=Kdis*Xc

          P2=Khyd_ch*Xch

          P3=Khyd_pr*Xpr

          P4=Khyd_li*Xli

          P5=Km_su*Ssu/(Ks_su+Ssu)*Xsu*I5

          P6=Km_aa*Saa/(Ks_aa+Saa)*Xaa*I6

          P7=Km_fa*Sfa/(Ks_fa+Sfa)*Xfa*I7

          P8=Km_c4*Sva/(Ks_c4+Sva)*Xc4*Sva/(Sbu+Sva+1e-6)*I8

          P9=Km_c4*Sbu/(Ks_c4+Sbu)*Xc4*Sbu/(Sva+Sbu+1e-6)*I9

          P10=Km_pro*Spro/(Ks_pro+Spro)*Xpro*I10
```

P13=Kdec_xsu*Xsu

P14=Kdec_xaa*Xaa

P15=Kdec_xfa*Xfa

P16=Kdec_xc4*Xc4

P17=Kdec_xpro*Xpro

P18=Kdec_xac*Xac

P19=Kdec_xh2*Xh2

#inorganic carbon

S1=-Cxc+FsI_xc*CsI+Fch_xc*Cch+Fpr_xc*Cpr+Fli_xc*Cli+FxI_xc*CxI

S2=-Cch+Csu

S3=-Cpr+Caa

S4=-Cli+(1-Ffa_li)*Csu+Ffa_li*Cfa

S5=-Csu+(1-Ysu)*(Fbu_su*Cbu+Fpro_su*Cpro+Fac_su*Cac)+Ysu*Cbac

S6=-Caa+(1-
Yaa)*(Fva_aa*Cva+Fbu_aa*Cbu+Fpro_aa*Cpro+Fac_aa*Cac)+Yaa*Cbac

S7=-Cfa+(1-Yfa)*0.7*Cac+Yfa*Cbac

```
        S8=-Cva+(1-Yc4)*.54*Cpro+(1-Yc4)*.31*Cac+Yc4*Cbac

        S9=-Cbu+(1-Yc4)*.8*Cac+Yc4*Cbac

        S10=-Cpro+(1-Ypro)*.57*Cac+ Ypro*Cbac

        S11=-Cac+(1-Yac)*Cch4+ Yac*Cbac

        S12=(1-Yh2)*Cch4+ Yh2*Cbac

        S13=-Cbac+Cxc

#acid-base rates:

      Pa_4=Ka_bva*(Sva_m*(Ka_va+Sh)-Ka_va*Sva)

      Pa_5=Ka_bbu*(Sbu_m*(Ka_bu+Sh)-Ka_bu*Sbu)

      Pa_6=Ka_bpro*(Spro_m*(Ka_pro+Sh)-Ka_pro*Spro)

      Pa_7=Ka_bac*(Sac_m*(Ka_ac+Sh)-Ka_ac*Sac)

      Ka_co2=10^(-6.35)*exp(7646/(R*100)*(1/Tbase-1/Top))

      Pa_10=Ka_bco2*(Shco3_m*(Ka_co2+Sh)-Ka_co2*Sic)

      Ka_in=10^(-9.25)*exp(51965/(R*100)*(1/Tbase-1/Top))

      Pa_11=Ka_bin*(Snh3*(Ka_in+Sh)-Ka_in*Sin)

#gas transfer equ&as transfer rates
```

```
        Pgas_h2=Sgas_h2*R*Top/16

        Pgas_ch4=Sgas_ch4*R*Top/64

        Pgas_co2=Sgas_co2*R*Top

        Kh_h2=(7.8e-4)*exp(-4180/(R*100)*(1/Tbase-1/Top))

        Pt_8=KL*(Sh2-16*Kh_h2*Pgas_h2)

        Kh_ch4=0.0014*exp(-14240/(R*100)*(1/Tbase-1/Top))

        Pt_9=KL*(Sch4-64*Kh_ch4*Pgas_ch4)

        Kh_co2=0.035*exp(-19410/(R*100)*(1/Tbase-1/Top))

        Pt_10=KL*(Sco2-Kh_co2*Pgas_co2)

        Pgas_h2o=0.0313*exp(5290*(1/Tbase-1/Top))

        Qgas = R*Top/(Patm-Pgas_h2o)*Vliq*(Pt_8/16+Pt_9/64+Pt_10)

#Components dff equ.

dSsu = tau*Ssu_in-tau*Ssu+(P2+(1-Ffa_li)*P4-P5)#C1  components

dSaa = tau*Saa_in-tau*Saa+(P3-P6)#C2

dSfa = tau*Sfa_in-tau*Sfa+(Ffa_li*P4-P7)#C3

dSva = tau*Sva_in-tau*Sva+((1-Yaa)*Fva_aa*P6-P8)#C4
```

dSbu = tau*Sbu_in-tau*Sbu+((1-Ysu)*Fbu_su*P5+(1-Yaa)*Fbu_aa*P6-P9)#C5

dSpro= tau*Spro_in-tau*Spro+((1-Ysu)*Fpro_su*P5+(1-Yaa)*Fpro_aa*P6+(1-Yc4)*.54*P8-P10)#C6

dSac = tau*Sac_in-tau*Sac+((1-Ysu)*Fac_su*P5+(1-Yaa)*Fac_aa*P6+.7*(1-Yfa)*P7

+.31*(1-Yc4)*P8+.8*(1-Yc4)*P9+.57*(1-Ypro)*P10)#C7

dSh2 = tau*Sh2_in-tau*Sh2+((1-Ysu)*Fh2_su*P5

+(1-Yaa)*Fh2_aa*P6+.3*(1-Yfa)*P7+.15*(1-Yc4)*P8+.2*(1-Yc4)*P9+.43*(1-Ypro)*P10

-Pt_8)#C8

dSch4 = tau*Sch4_in-tau*Sch4-Pt_9 #C9

dSic=tau*Sic_in-tau*Sic-(sum(S1*P1,S2*P2,S3*P3,S4*P4,S5*P5,S6*P6,S7*P7,S8*P8,S9*P9,S10*P10)+S13*(P13+P14+P15+P16+P17+P18+P19))-Pt_10 #C10

dSin = tau*Sin_in-tau*Sin-Ysu*Nbac*P5+(Naa-Yaa*Nbac)*P6-Yfa*Nbac*

P7-Yc4*Nbac*P8-Yc4*Nbac*P9-Ypro*Nbac*P10+(Nbac-Nxc)*sum(P13,P14,P15,P16,P17,P18,P19)+(Nxc-FxI_xc*Ni-FsI_xc*Ni-Fpr_xc*Naa)*P1 #C11

```
dSi   =   tau*Si_in-tau*Si+FsI_xc*P1   #C12

dXc   =  tau*Xc_in-tau*Xc +(-P1+sum(P13,P14,P15,P16,P17,P18,P19))  #C13

dXch   =  tau*Xch_in-tau*Xch +(Fch_xc*P1-P2)   #C14

dXpr =   tau*Xpr_in-tau*Xpr +(Fpr_xc*P1-P3)  #C15

dXli =   tau*Xli_in-tau*Xli +(Fli_xc*P1-P4)  #C16

dXsu =   tau*Xsu_in-tau*Xsu +(Ysu*P5-P13)  #C17

dXaa =   tau*Xaa_in-tau*Xaa +(Yaa*P6-P14)  #C18

dXfa =   tau*Xfa_in-tau*Xfa +(Yfa*P7-P15)  #C19

dXc4 =   tau*Xc4_in-tau*Xc4 +(Yc4*P8+Yc4*P9-P16)  #C20

dXpro =   tau*Xpro_in-tau*Xpro +(Ypro*P10-P17)  #C21

dXac =   tau*Xac_in-tau*Xac   #C22

dXh2 =   tau*Xh2_in-tau*Xh2   #C23

dXi =   tau*Xi_in-tau*Xi +(FxI_xc*P1)  #C24

dScation =   tau*Scation_in-tau*Scation  #C25  cations and anions

dSanion =   tau*Sanion_in-tau*Sanion    #C26

dSva_m = -Pa_4  #C27   ion states
```

```
dSbu_m = -Pa_5  #C28

dSpro_m = -Pa_6  #C29

dSac_m = -Pa_7   #C30

dShco3_m = -Pa_10 #C31

dSnh3 = -Pa_11   #C32

dSgas_h2 =-Sgas_h2*Qgas/Vgas+Pt_8*Vliq/Vgas     #33    gas phase differential equ.

dSgas_ch4 =-Sgas_ch4*Qgas/Vgas+Pt_9*Vliq/Vgas    #34

dSgas_co2 =-Sgas_co2*Qgas/Vgas+Pt_10*Vliq/Vgas    #35

list(c(dSsu,dSaa,dSfa,dSva,dSbu,dSpro,dSac,dSh2,dSch4,dSic,dSin,dSi,dXc,dXch,dXpr,

dXli,dXsu,dXaa,dXfa,dXc4,dXpro,dXac,dXh2,dXi,dScation,dSanion,dSva_m,dSbu_m,dSpro_m

,dSac_m,dShco3_m,dSnh3,dSgas_h2,dSgas_ch4,dSgas_co2))

# calculate pH Pgas_h2,Pgas_ch4,Pgas_co2?

})

}
```

```
require(deSolve) # external package1

Q =  0.054; Vliq=0.054; Vgas=0.006 # flow rate

tau=Q/Vliq;

#parameters' values, change values based on different digestion

Ffa_li=0.95;Yaa=0.15;Fva_aa=0.23;Ysu=0.15;Fbu_su=0.13;Fbu_aa=0.26;Fpro_su=0.27;Fpro_aa=0.05;Yc4=0.06;

Fac_su=0.41;Fac_aa=0.4;Yfa=0.05;Ypro=0.05;Fh2_su=0.19;Fh2_aa=0.06;Yac=0.05;Yh2=0.05;Nbac=0.08/14;Naa=0.007;
Nxc=0.0376/14;FxI_xc=0.2;Ni=0.06/14;FsI_xc=0.1;Fpr_xc=0.2;Fch_xc=0.2;Fli_xc=0.3;

Kdis=0.25;Khyd_ch=10;Khyd_pr=10;Khyd_li=10;Km_su=27;Ks_su=0.05;Km_aa=27;Ks_aa=0.05;Km_fa=12;Ks_fa=1;

Km_c4=14;Ks_c4=0.03;Km_pro=11;Ks_pro=0.02;Km_ac=13;Ks_ac=0.04;Km_h2=44;Ks_h2=1e-6;

Kdec_xsu= Kdec_xaa= Kdec_xfa=Kdec_xc4= Kdec_xpro= Kdec_xac= Kdec_xh2=0.02;Cxc=0.02786;CsI=0.03;

Cch=0.0313;Cpr=0.03;Cli=0.022;CxI=0.03;Csu=0.0313;Caa=0.03;Cbu=0.025;Cpro=0.0268;Cac=0.0313;
```

Cbac=0.0313;Cva=0.024;Cfa=0.0217;Cch4=0.0156;pHuL_aa=5.5;pHlL_aa=4;pHuL_ac=7;pHlL_ac=6;

pHuL_h2=6;pHlL_h2=5;Ks_in=1e-4;Ki_h2_fa=5e-6;Ki_h2_c4=1e-5;Ki_h2_pro=3.5e-6;Ki_nh3=0.0018;

Ka_bva= Ka_bbu= Ka_bpro= Ka_bac= Ka_bco2= Ka_bin=1e10;Ka_va=10^(-4.86);

Ka_bu=10^(-4.82);Ka_pro=10^(-4.88);Ka_ac=10^(-4.76);KL=200;Ki_h2=2.2e-6;Ki_vfa=1.4e-6;

R=0.083145;Tbase=298.15;Top=308.15;Patm=1.013;

constants

Kw=(exp(55900/(R*100)*(1/Tbase-1/Top)))*(10^(-14))

Ka_co2=10^(-6.35)*exp(7646/(R*100)*(1/Tbase-1/Top))

Ka_in=10^(-9.25)*exp(51965/(R*100)*(1/Tbase-1/Top))

Kh_h2=(7.8e-4)*exp(-4180/(R*100)*(1/Tbase-1/Top))

Kh_ch4=0.0014*exp(-14240/(R*100)*(1/Tbase-1/Top))

Kh_co2=0.035*exp(-19410/(R*100)*(1/Tbase-1/Top))

Pgas_h2o=0.0313*exp(5290*(1/Tbase-1/Top))

```
#input values

Ssu_in=0.03;Saa_in=0.001;Sfa_in=0.002;Sva_in=0.002;Sbu_in=0.002;
Spro_in=0.002;

Sac_in=0.001; Sh2_in=1e-8; Sch4_in=1e-5; Sic_in=0.04; Sin_in=0.01; Si_in=1;
Xc_in=5;

Xch_in=73.35; Xpr_in=14.80; Xli_in=7.90; Xsu_in=0.03; Xaa_in=0.01;
Xfa_in=0.002; Xc4_in=0.01;

Xpro_in=0.01; Xac_in=0.1; Xh2_in=0.01; Xi_in=16; Scation_in=0.04;
Sanion_in=0.02

#states initial condition, liquid within the digester, not the input

state=c(Ssu=0.3,Saa=0.001,Sfa=0.3, Sva=0.3, Sbu=0.3, Spro=0.3, Sac=0.3,
Sh2=1e-6,

Sch4=1e-5, Sic=0.04, Sin=0.01, Si=0.02, Xc=0.3, Xch=0.026, Xpr=0.3, Xli=0.03,
Xsu=0.4, Xaa=1.1,

Xfa=0.20, Xc4=0.41, Xpro=0.137, Xac=0.7, Xh2=0.01, Xi=5, Scation=0.04,
Sanion=0.02,

Sva_m=0.0601, Sbu_m=0.0905,Spro_m=0.13, Sac_m=0.159, Shco3_m=0.0090,

Snh3=0.0165, Sgas_h2=0.03, Sgas_ch4=0.029, Sgas_co2=0.0378)
```

```
#parameters

parameters=c(Ffa_li= Ffa_li,Yaa= Yaa,Fva_aa= Fva_aa,Ysu= Ysu,Fbu_su=
Fbu_su,Fbu_aa= Fbu_aa,

Fpro_su= Fpro_su,Fpro_aa= Fpro_aa,Yc4= Yc4,Fac_su= Fac_su,Fac_aa=
Fac_aa,Yfa= Yfa,Ypro= Ypro,

Fh2_su= Fh2_su,Fh2_aa= Fh2_aa,Yac= Yac,Yh2= Yh2,Nbac= Nbac,Naa =
Naa,Nxc= Nxc,FxI_xc= FxI_xc,

Ni= Ni,FsI_xc= FsI_xc,Fpr_xc= Fpr_xc,Fch_xc= Fch_xc,Fli_xc= Fli_xc,Kdis=
Kdis,Khyd_ch= Khyd_ch,

Khyd_pr= Khyd_pr,Khyd_li= Khyd_li,Km_su= Km_su,Ks_su= Ks_su,Km_aa=
Km_aa,Ks_aa= Ks_aa,Km_fa= Km_fa,

Ks_fa= Ks_fa,Km_c4= Km_c4,Ks_c4= Ks_c4,Km_pro= Km_pro,Ks_pro=
Ks_pro,Km_ac= Km_ac,Ks_ac= Ks_ac,

Km_h2= Km_h2,Ks_h2= Ks_h2,Kdec_xsu=Kdec_xsu, Kdec_xaa= Kdec_xaa,
Kdec_xfa= Kdec_xfa,

Kdec_xc4= Kdec_xc4, Kdec_xpro= Kdec_xpro, Kdec_xac=Kdec_xac,
Kdec_xh2= Kdec_xh2,Cxc= Cxc,

CsI= CsI,Cch= Cch,Cpr= Cpr,Cli= Cli,CxI= CxI,Csu= Csu,Caa= Caa,Cbu= Cbu,
```

```
Cpro= Cpro,Cac= Cac,Cbac = Cbac,Cva= Cva,Cfa= Cfa,Cch4= Cch4,pHuL_aa=
pHuL_aa,pHlL_aa= pHlL_aa,

pHuL_ac= pHuL_ac,pHlL_ac= pHlL_ac, pHuL_h2= pHuL_h2,pHlL_h2=
pHlL_h2,Ks_in= Ks_in,Ki_h2_fa= Ki_h2_fa,

Ki_h2_c4= Ki_h2_c4,Ki_h2_pro= Ki_h2_pro,Ki_nh3= Ki_nh3, Ka_bva=
Ka_bva, Ka_bbu= Ka_bbu,

Ka_bpro= Ka_bpro, Ka_bac= Ka_bac, Ka_bco2= Ka_bco2, Ka_bin= Ka_bin,
Ka_va= Ka_va,

Ka_bu= Ka_bu,Ka_pro= Ka_pro,Ka_ac= Ka_ac,KL= KL,R= R,

Tbase=Tbase,Top= Top,Patm= Patm,

Kh_h2= Kh_h2, Kh_ch4= Kh_ch4,Kh_co2= Kh_co2,Ka_in=
Ka_in,Pgas_h2o=Pgas_h2o)

#extract pH

getpH <- function(state) {

  with(as.list(c(state,parameters)), {

    Snh4=Sin-Snh3
```

```
    Z=Scation+Snh4-Shco3_m-Sac_m/64-Spro_m/112-Sbu_m/160-Sva_m/208-
Sanion

    Sh=-Z*0.5+0.5*sqrt(Z^2+4*Kw)

  pH <- -log10(Sh*0.6)})

}



#extract Qgas

getQgas <- function(state) {

  with(as.list(c(state,parameters)), {

    Sco2=Sic-Shco3_m

    Pgas_h2=Sgas_h2*R*Top/16

    Pgas_ch4=Sgas_ch4*R*Top/64

    Pgas_co2=Sgas_co2*R*Top

    Pt_8=KL*(Sh2-16*Kh_h2*Pgas_h2)

    Pt_9=KL*(Sch4-64*Kh_ch4*Pgas_ch4)

    Pt_10=KL*(Sco2-Kh_co2*Pgas_co2)
```

```
   Qgas = R*Top/(Patm-Pgas_h2o)*Vliq*(Pt_8/16+Pt_9/64+Pt_10) })
}
# extract Pgas_h2/ch4/co2
getPgas_h2 <- function(state) {
  with(as.list(c(state,parameters)), {
    Pgas_h2=Sgas_h2*R*Top/16})
}
getPgas_ch4 <- function(state) {
  with(as.list(c(state,parameters)), {
    Pgas_ch4=Sgas_ch4*R*Top/64})
}


getPgas_co2 <- function(state) {
  with(as.list(c(state,parameters)), {
    Pgas_co2=Sgas_co2*R*Top})
}
```

```
state.pH <- getpH(state=state)

state.Qgas <- getQgas(state=state)

state.Pgas_h2 <- getPgas_h2(state=state)

state.Pgas_ch4 <- getPgas_ch4(state=state)

state.Pgas_co2 <- getPgas_co2(state=state)

  # number of iterations

nIt = 10000

# create place for results

mc.out <- data.frame()    # for just one time

mc.all.out <- list()      # for all the output

# define distributions for the parameters

p.test.Xc_in <- rnorm(nIt,Xc_in,0.12)

p.test.Xch_in <- rnorm(nIt,Xch_in,4.23)

p.test.Xpr_in <- rnorm(nIt,Xpr_in,1.53)

p.test.Xli_in <- rnorm(nIt,Xli_in,0.95)
```

```
p.test.Ffa_li <- rnorm(nIt,Ffa_li,0.05)

p.test.Yaa <- rnorm(nIt,Yaa,0.05)

p.test.Fva_aa <- rnorm(nIt,Fva_aa,0.05)

p.test.Ysu <- rnorm(nIt,Ysu,0.05)

p.test.Fbu_su <- rnorm(nIt,Fbu_su,0.05)

p.test.Fbu_aa <- rnorm(nIt,Fbu_aa,0.05)

p.test.Fpro_su <- rnorm(nIt,Fpro_su,0.004)

p.test.Fpro_aa <- rnorm(nIt,Fpro_aa,0.002)

p.test.Yc4 <- rnorm(nIt,Yc4,0.001)

p.test.Fac_su <- rnorm(nIt,Fac_su,0.03)

p.test.Fac_aa <- rnorm(nIt,Fac_aa,0.02)

p.test.Yac <- rnorm(nIt,Yac,0.006)

p.test.Yh2 <- rnorm(nIt,Yh2,0.0001)

p.test.FxI_xc <- rnorm(nIt,FxI_xc,0.02)

p.test.Fpr_xc <- rnorm(nIt,Fpr_xc,.003)

p.test.Fch_xc <- rnorm(nIt,Fch_xc,.003)
```

```
p.test.Fli_xc <- rnorm(nIt,Fli_xc,.003)

p.test.Kdis <- rnorm(nIt,Kdis,.05)

p.test.Km_su <- rnorm(nIt,Km_su,.003)

p.test.Ks_su <- rnorm(nIt,Ks_su,.003)

p.test.Km_aa <- rnorm(nIt,Km_aa,2.5)

p.test.Km_fa <- rnorm(nIt,Km_fa,.3)

p.test.Ks_c4 <- rnorm(nIt,Ks_c4,0.002)

p.test.Km_ac <- rnorm(nIt,Km_ac,2)

p.test.Km_h2 <- rnorm(nIt,Km_h2,4)

# copy the parameters to modify

pars <- parameters[c('Xc_in','Xch_in','Xpr_in','Xli_in','Xpr_in','Xli_in','Ffa_li',

'Yaa','Fva_aa','Ysu','Fbu_su','Fpro_su','Fpro_aa','Yc4','Fac_su','Fac_aa','Yac','Yh2',

'FxI_xc','Fpr_xc','Fch_xc','Fli_xc','Kdis','Km_su','Ks_su','Ks_c4','Km_ac','Km_h2'

)]

# start the Monte Carlo iteration

for(iIt in 1:nIt){
```

```
# put the 'new' values in a named vector

p.test <- c(p.test.Km_h2[iIt],p.test.Fac_aa[iIt],p.test.Ks_fa[iIt])

names(p.test) <- names(pars)



# copy the 'old' parameter list

parms <- parameters

# replace the 'old' with the 'new' in the new list

parms[names(pars)] <- p.test



# run the model

out <- as.data.frame(ode(y = state,times = times,func=ADM1_C,parms=
parms,method='lsoda'))

out$pH       <- getpH(state=out)

out$Qgas     <- getQgas(state=out)

out$Pgas_h2  <- getPgas_h2(state=out)

out$Pgas_ch4 <- getPgas_ch4(state=out)
```

```
  out$Pgas_co2 <- getPgas_co2(state=out)

  # put the SS value in a data frame

  mc.out <- rbind(mc.out,out[nrow(out),])



  # put entire output table in a list

  mc.all.out[[iIt]] <- out}

# plot the output

par(mfrow=c(4,5),mar=c(0,0,0,0),mgp=c(2.5,.5,0),oma=c(5,4,2,1),las=1,tcl=.25,c
ex.axis=.75)

iplt <<- 1          ##1:n, 1 is the time

lapply(2:21,function(ix) {

  x = out[,ix]; tx <- names(out)[ix]

  plot(x~out$time,type='l',xaxt='n',ylab='')

  if(iplt > 15) {

    axis(1,labels=T)

  }
```

```
  else {
    axis(1,labels=F)
  }
  u <- par('usr'); dy = diff(u[3:4])/10
  text(0,par('usr')[4]-dy,labels=tx,pos=4)
  cat(iplt,tx,'\n')          ###\n huanhang,
  iplt <<- iplt + 1
} )
mtext('Time, days',side=1,outer=T,line=3)
mtext('Constituent value',side=2,line=2,las=0,outer=T)
windows()
par(mfrow=c(4,5),mar=c(0,0,0,0),mgp=c(2.5,.5,0),oma=c(5,4,2,1),las=1,tcl=.25,c
ex.axis=.75)
iplt <<- 1          ##1:n, 1 is the time
lapply(22:41,function(ix) {
  x = out[,ix]; tx <- names(out)[ix]
```

```
 plot(x~out$time,type='l',xaxt='n',ylab='')
 if(iplt > 15) {
  axis(1,labels=T)
 }
 else {
  axis(1,labels=F)
 }
 u <- par('usr'); dy = diff(u[3:4])/10
 text(0,par('usr')[4]-dy,labels=tx,pos=4)
 cat(iplt,tx,'\n')          ###\n huanhang,
 iplt <<- iplt + 1
} )
mtext('Time, days',side=1,outer=T,line=3)
mtext('Constituent value',side=2,line=2,las=0,outer=T)
x <- data.frame(a = I("a \" quote"), b = pi)
write.table(out, file = "outD.csv", sep = ",", col.names = NA,
```

```
qmethod = "double")
```

CURRICULUM VITA

JIANMING ZHONG

Dept. of Civil and Environmental Engineering
Utah State University
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Specialties and Skills

Water Quality and Wastewater Management		Anaerobic Digestion
Biochemistry Techniques	Statistical Data Analysis	Dynamic Model Development

Education

Ph. D. Environmental Engineering 02/2016

Utah State University, Logan, UT GPA 3.8

M. S. Biochemistry 06/2010

Nanchang University, Nanchang, Jiangxi, China GPA 3.6

B. E. Biological Engineering 06/2007

Nanchang University, Nanchang, Jiangxi, China GPA 3.4

Experience

Engineering Intern Glanbia Foods Twin Falls, ID 05/2015-08/2015

- Optimized the energy usage in Glanbia's WWTP to help the company save $60,000/year
- Improved anaerobic digestion performance in Glanbia's WWTP to help the company save $30,000/year.
- Solved holding pond overflow issue in Glanbia's WWTP to avoid breaking permit

Research Assistant Utah Water Research Lab Logan, UT 09/2010-02/2016

- Developed a mathematic model for anaerobic hydrogen production from wastewater
- Conducted testing and developed models for anaerobic digestion using cow manure and cheese whey for waste treatment in local farms
- Conducted research project on anaerobic digestion of dairy industry waste for both biogas and hydrogen production
- Established methods to improve algae anaerobic digestion in Logan Wastewater Treatment Facility (Lagoon)
- Guided the establishment of 6000-gallon hydrogen anaerobic digester in Logan Wastewater Treatment Facility

Resident Assistant Utah State University Logan, UT 06/2011-12/2011

- Actively participated in the development of a comprehensive Residence Life program
- Conducted residential programs in a team for thousands of residents
- Led a 20-member resident assistant team in placing and organizing large resident events

Research Assistant Nanchang University Nanchang, China 09/2007-01/2010

- Conducted research project on protein-Tra1's function in gene transcription and translation in brewer's yeast (*S. cerevisiae*)
- Established the yeast two-hybrid system for protein function identification in yeast
- Conducted research project on yeast RPL-36B protein's role in gene translation in yeast
- Proficient in Plasmids Construct, DNA/RNA Extraction, RT-PCR and other biological techniques.

Engineering Intern Nanchang Beer Co.Ltd Nanchang, China 09/2006-12/2006

- Assisted the quality control in the beer fermentation
- Help solve engineering problems in the plant

Publication

Zhong, J., D. Stevens, and C. Hansen. 2015. Modeling of anaerobic hydrogen production from dairy processing waste using a modified ADM1. International Journal of Hydrogen Energy, (Submitted, under reviewing)

Demistry, M., **J. Zhong**, C. Hansen, and M. McFarland. 2015. Modifying the ADM1 model to predict the operation of an anaerobic digester co-digesting municipal sludge

with bakery waste. Environment and pollution,4(4):38-57.

Zhong, J., D. Stevens, and C. Hansen. 2015. Optimization of anaerobic hydrogen and methane production from dairy processing waste using a two-stage digestion in induced bed reactors (IBR). International Journal of Hydrogen Energy,40(45): 15470–15476.

Hansen, C., **J. Zhong**, and J. Hansen. 2014. Anaerobic digestion of dairy processing waste, algae & grass in pilot and full scale. Transactions of the ASABE, 57(2): 609-614.

Zhong, J., Y. Lu, X. Liu, and Y. Zhang. 2010. NuA 4 complex and its cellular functions. Jiangxi Science, 6(24):855~857.

Liu, X., **J. Zhong**, Y. Zhang, and X. Zhou. 2010. Insulator anti-gene silencing function and application. Jiangxi Science, 2(24):567~571.

Zhang, Y., **J. Zhong**, X. Liu, and X. Zhou. 2010. Noncoding RNAs and their regulation. Jiangxi Science, 2(24):630~633.

Professional Progress

Build Dairy Project, 2014

40-hour Training of Hazardous Waste Operations and Emergency Response, 2013

Fundamental Engineering Certificate, 2012

Training and Practice of Pollution Prevention Process, 2012

Computer and Language Skills

- Proficient in using software including: Microsoft Office, SAS, Matlab, and R
- Proficient in speaking and writing Chinese and English