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Title	International biogas applications
Authors	Chan Gutierrez, Enrique
Publication date	2018
Original Citation	Chan Gutierrez, E. A. 2018. International biogas applications. PhD Thesis, University College Cork.
Type of publication	Doctoral thesis
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Download date	2026-11-07 02:57:30
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Energy Engineering

School of Engineering

& Environmental Research Institute

University College Cork

International biogas applications

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Thesis submitted for the degree of Doctor of Philosophy to the National University of Ireland, Cork

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May 2018

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Declaration

It is stated that this thesis is my original research work and that it has not been submitted for another degree, either at University College Cork or elsewhere. Any other sources of supporting information have been acknowledged.



Enrique Abraham Chan Gutierrez

May, 2018

Acknowledgements

This thesis would have not been possible without the following:

My supervisor Professor Jerry Murphy for his support, guidance and encouragement during my time as a PhD student.

My co-supervisor Dr.Brian Ó'Gallachóir for all the interesting conversations and support.

Dr. David Wall for all his help and advice during these years.

Professor Sarah Culloty director of the Environmental Research Institute for providing the facilities to conduct this research.

CONACYT-CONCITEY for the scholarship provided to undertake this research.

To all my colleagues in the biofuels group, Richie, Magdalena, Amita, Markus, Shane Gallagher, Shane McDonagh, Eoin Allen, Eoin Ahern, Truc, Muhammad, Christiane, Aoife, Richen, karthik, Lingkan, Barbara and especially to Dr. Ao Xie for his valuable help.

My parents, Jorge and Paty and my brothers Jorge, Chris and Memo for their continuous support.

Finally, to my dear wife Sol for having followed me to this part of the world and to my beautiful baby Fernando for all the joy and extra work he has brought into our lives

Abstract

AD plants for the production of biogas have grown steadily in recent years. Animal slurry and sewage sludge have been traditionally used as feedstock for AD, however, substrates such as the organic fraction of municipal solid waste (OFMSW) and energy crops are preferred due to their higher methane yields. Concerns about the sustainability of the use energy crops for biogas production have reduced the use of these substrates worldwide, which has encouraged the research on algae feedstock for AD. In the first part of this thesis, the biogas and economic potential from wastes and energy crops in a Mexican context are analysed. In Mexico AD from food waste has the potential of producing 42 PJ and to reduce 17.9 $MtCO_2e$ on a yearly basis. An urban centralised plant for the co-digestion of food waste and sewage sludge for the production of biomethane as a transport fuel can be economically feasible. The co-digestion of food waste and pig slurry appear less profitable due to higher operational costs and reduced gate fees. The implementation of economic instruments can help increasing the economic attractiveness of these plants. In the case of rural digesters, on-site co-digestion of pig slurry and grass can be economically feasible if the biogas produced is used to generate electricity. Centralised biogas plants appear less profitable, requiring higher tariffs to break even. The production of biomethane as a transport fuel is not economically viable in rural plants. In the second part of the thesis the biogas potential of algae in an Irish context is analised through laboratory research. A special reactor was design and build for the co-digestion of *Ulva lactuca*, dairy slurry and grass silage. Continuous co-digestion of 25% *U.lactuca*, 5% dairy slurry and 70% grass silage based on volatile solids (VS) yielded 288 LCH_4/kgVS, at an organic loading rate (OLR) of 2 kgVS/m^3/d equating to 89% of the biomethane potential test value. When digested at an OLR of 3 kgVS/m^3/d, methane yields decreased due to an accumulation of volatile fatty acids. A biomethane plant in the coastal town of Timoleague, Cork co-digesting beach cast *U.lactuca*, dairy slurry and grass silage would have a methane production of approximately 1.19 Mm^3/year (42 TJ/year). A two-stage continuous fermentative hydrogen and methane co-production using macro-algae (*Laminaria digitata*) and micro-algae (*Arthrospira platensis*) was tested in laboratory trials. The hydraulic retention time (HRT) of the H_2 reactor was set at 4 days. The highest hydrogen yield was obtained at an OLR of 6 kgVS/m^3/d. In the second-stage Methane reactor a HRT of 12 days was used, reaching a specific methane yield of 245 L/kgVS at an OLR of 2kgVS/m^3/d. The energy yield was calculated at 9.4 kJ/gVS. Nevertheless, when the OLR was increased a reduction in H_2 and methane was observed. A one-stage anaerobic reactor was run as a comparison to the two-stage system at an HRT of 16 days. At an OLR of 1 a methane yield of 204 L/kgVS was achieved. When the OLR was increased to 2 kgVS/m^3/d, the yields decreased due to volatile fatty acid accumulation. The two-stage system offered better performances in both energy return and process stability. The energy potential of this algal mixture may reach 213 GJ/ha/yr.

Output of the thesis and contribution to the papers

Chapter 3: I was the first author of the paper and was responsible for the data collection, processing and interpretation of results. I was advised by Dr.David Wall and Professor Jerry Murphy. This thesis chapter is identical to the published paper.

Gutierrez, E.C., Wall, D.M., O'Shea, E., Mendez, R., Moreno, M.,Murphy, J.D. 2016. An economic and carbon analysis of biomethane production from food waste to be used as a transport fuel in Mexico. Journal of Cleaner Production, 20, 852-862. https://doi.org/10.1016/j.jclepro.2018.06.051

Chapter 4: I was the first author of the paper and was responsible for the data collection, processing, data analysis and interpretation of results. I was advised by Dr.Ao Xia and Professor Jerry Murphy. This thesis chapter is identical to the published paper.

Gutierrez, E. C., Xia, A., & Murphy, J. D. (2016). Can slurry biogas systems be cost effective without subsidy in Mexico? Renewable Energy, 95, 22–30. https://doi.org/10.1016/j.renene.2016.03.096

Chapter 5: I was the first author of the paper and was responsible for the experimental design, sample collection, operation of the continuous digester and data analysis. I was advised by Dr. David M. Wall and Professor Jerry Murphy. This chapter is under preparation for publication.

Chapter 6:I was the first author of the paper Jointly with Lingkan Ding. I was responsible for laboratory work, data collection and analysis. Sample collection and preparation was undertaken with the aid of my colleagues. I was responsible for the writing of the paper jointly with Lingkan Ding. I was advised by Professor Jerry Murphy. This chapter is identical to the published paper.

Ding, L., Gutierrez, E.C., Cheng, J., Xia, A., Shea, R. O., Jacob, A., & Murphy, J. D. (2018). Assessment of continuous fermentative hydrogen and methane co-production using macro- and micro-algae with increasing organic loading rate. Energy, 151, 760–770. https://doi.org/10.1016/j.energy.2018.03.103

Nomenclature

AD	Anaerobic Digestion
AFB	Anaerobic fermentative bacteria
BI	Biodegradability index
BMP	Biomethane potential assay
BOC	Biofuel Obligation Certificate
CAPEX	Capital Cost
CDM	Clean Development Mechanism
CEC	Clean Energy Certificates
CH_4	Methane
CNG	Compressed Natural Gas
CO_2	Carbon Dioxide
CSTR	Continuously Stirred Tank Reactor
DS	Dry Solids
ENCC	Estrategia Nacional de Cambio Climatico (National Estrategy for Climate Change)
FOS:TAC	Flüchtige organische säuren /totales anorganisches carbonat (volatile organic acids/total inorganic carbon)
FW	Food Waste
GHG	Greenhouse gas emission
H_2	Hydrogen
HDPE	High Density Polyurethane
HPWS	High Pressure Water Scrubbing
HRT	Hydraulic Retention Time

LCOE	Levelised Cost of Energy
LGCC	Ley General de Cambio Climatico (General Law for Climate Change)
NGV	Natural Gas Vehicle
NPV	Net Present Value
OFMSW	Organic Fraction of Municipal Solid Waste
OLR	Organic Loading Rate
OPEX	Operational Cost
SEMARNAT	Secretaria de Medio Ambiente y Recursos Naturales (Environment and Natural Resources Department)
SENER	Secretaria de Energia (National Secretariat of Energy)
SHY	Specific Hydrogen Yield
SMY	Specific Methane Yield
TAN	Total Ammonical Nitrogen
UAP	Unit of Animal Population
VFA	Volatile Fatty Acids
VS	Volatile Solids
WW	Wastewater

List of units

kWh	kilowatt hour
kWhe	kilowatt hour of electric power
MJ	Mega Joule
GJ	Giga Joule
PJ	Peta Joule
TJ	Tera Joule
m^3	Cubic metre
Mm^3	Million cubic metre
ha	Hectare
Mt	Million tonne
Gg	Gigagram
t	Tonne
kg	Kilogram
mg	milligram
km	Kilometre
L	Litre
DGE	Diesel gallon equivalent
GGE	Gallons of gasoline equivalent

1. Introduction

1.1. Introduction and background to thesis

Anaerobic digestion is a fermentation process that occurs in the absence of oxygen which produces a gas that consists mostly of methane (50-70%) and carbon dioxide. This so called biogas can be cleaned from impurities to upgrade it into natural gas quality (97% methane on average) to be used as a fuel for transport, electricity generation and heat production. Biomass containing high quantities of carbohydrates, fats, proteins, cellulose and hemicellulose can be used as feedstock for the process. Traditionally anaerobic digestion has been used to treat wastewater (municipal and industrial), sewage sludge and animal slurries, however, with the oil crisis in the 1970's the interest in using this process as a mean to produce biogas for energy applications (transport fuel, heat and power) has increased (Abbasi, Tauseef, and Abbasi 2012). Nowadays sewage sludge and animal slurries are commonly co-digested with other substrates such as harvest residues, organic waste from industries, food waste, maize, sugar beet and grass. This technology has been widely adopted in Europe where Germany leads with more than 10,000 biogas plants, digesting mostly animal slurries and energy crops (EBA 2017). The upgrading of biogas to biomethane is gaining more attention due to the European Energy Directive that requires 10% of transport fuel to be renewable by 2020. Nevertheless, the amount of first generation biofuels (biofuels based on edible crops) has been limited to 7% of total energy in transport (EC 2017). This could favour the use of animal slurries as a feedstock for anaerobic digestion and the production of second generation biofuels based on lignocellulosic material such as grass and third generation biofuels based on macro and micro algae. In the case of Mexico and Latin America anaerobic digestion has been traditionally used to treat sewage sludge and industrial wastewater, however, in the mid 2000's, there was a surge of anaerobic digesters in rural areas mainly because of the Kyoto protocol that allowed anaerobic digestion developers to sell carbon credits to foreign

stakeholders. Mexico is the second ranked country in Latin American for clean development mechanism projects behind Brazil. The majority of projects consist of anaerobic digestion plants that treat slurries from intensive pig farming (Lokey 2009). There is an increasing interest in using other substrates such as slaughter house waste, dairy slurry, food waste and energy crops such as grass, however, the technical issues reported by the operators of the first constructed plants and the uncertainty of their economic feasibility have stalled the use of these feedstocks in large scale anaerobic digestion projects (Lokey 2009). Nevertheless, this trend is due to change as a new law promoting the use of renewables was published by the federal government in 2013 (DOF 2013). Life cycle assessment of biogas production has been shown to have lesser negative environmental impacts as compared to conventional energy sources. On top of that, if wastes are used as feedstock, significant reductions on greenhouse gases can be achieved (Hijazi et al. 2016). In the case Mexico, most wastes are disposed without an adequate treatment, creating a source of air, water and soil pollution. If digested in anaerobic reactors a reduction on greenhouse gases and environmental pollution can be obtained.

In an international context, the use of grass for anaerobic digestion and algae for the production of advanced biofuels is gaining more attention. Grass is considered a potential feedstock for anaerobic digestion given its high dry solids content and high specific methane yields. Mono digestion of grass has been described as difficult and unstable in long term digestion (Thamsiriroj and Murphy 2010; Thamsiriroj, Nizami, and Murphy 2012). To overcome this, co-digestion with animal slurry has been recommended in order to stabilise the anaerobic digestion process. In rural areas a biodigester treating slurry and grass can help create a circular economy which can be beneficial for farmers and the environment.

Algae is considered a third generation biofuel and is believed to possess several advantages over land based feedstocks such as higher production rates, and a higher rate of carbon dioxide fixation (Maity, Bundschuh, et al. 2014). On top of that, algae do not need arable land to be produced neither do they need fresh water for their cultivation, avoiding the food vs fuel debate. Algae both micro and macro have been anaerobically digested and co-digested at laboratory scale finding promising results (Tabassum, Wall, and Murphy 2016; Eoin Allen et al. 2014), nevertheless, there are still some major bottlenecks that prevent the use of this feedstock on a larger scale. Organic substances such as polyphenols in macro algae and triglycerides in micro algae are not easily digested under anaerobic conditions hence decreasing the biodegradability of the process. To overcome this, some authors have suggested a two-stage process, separating the hydrolysis and the methanation stages, therefore producing hydrogen and methane (Ding et al. 2016a; Guneratnam, Xia, and Murphy 2017).

In the first part of this thesis the feasibility of the production of biomethane for power generation and for transport fuel using pig slurry, grass and food waste as a feedstock is analysed on a Mexican context. Potential methane yields as well as the economic feasibility is assessed. In the second part of this thesis the anaerobic co-digestion of grass silage, dairy slurry and *Ulva lactuca* is analysed. *Ulva lactuca,* also known as sea lettuce is an edible green algae that belongs to the phylum chlorophyta and is commonly found in the majority of oceans. These substrates were co-digested on a pilot scale Continuously Stirred Tank Reactor (CSTR). Aspects such as organic loading rates, substrates mixes and reactor performance were assessed. The continuous fermentative production of hydrogen and methane from macro and micro algae (*Laminaria digitata* and *Arthrospira platensis*) was assessed. For this purpose, biomethane potential tests and biohydrogen potential tests were performed. *Laminaria digitata* is a brown algae that belongs to

the phylum Phaeophyceae and it is commonly found in the north Atlantic ocean. *Arthrospira platensis* is a bacteria also known as blue-green algae that belongs to the phylum cyanobacteria. *Arthrospira platensis* is generally used as a food supplement. *Laminaria digitata* and *Arthrospira platensis* were used as a feedstock in a two stage continuous fermentative hydrogen and methane system using 5 litre CSTR's. Optimum organic loading rates and reactor performance amongst other parameters were analysed.

1.2. Thesis aims and objectives

The aims and objectives of this thesis are:

- To estimate the resource and biomethane potential of pig slurry and food waste in Mexico.
- To analyse the technical and economic feasibility of a biomethane plant co-digesting pig slurry and grass in Mexico.
- To analyse the technical and economic feasibility of a biomethane plant for a Mexican city which digests food waste as a main substrate for the production of biomethane.Two different scenarios were analysed using pig slurry and sewage sludge as co-substrates respectively.
- To estimate the potential of reducing greenhouse gas emissions by displacing the use of diesel with biomethane in transport in Mexico.
- To evaluate the anaerobic co-digestion of *Ulva lactuca*, grass silage and dairy slurry at different mixes in biomethane potential tests and continuous digestion and to analyse what parameters may affect the process.

- To evaluate the co-generation of hydrogen and methane using a mixture of *Laminaria digitata* and *Arthrospira platensis* with increasing organic loading rates and to evaluate the performances of two-stage and one-stage systems based on energy conversion.

1.3. Thesis outline

The thesis consists of 7 chapters. Chapter 1 consists of a brief introduction to the thesis. Chapter 2 analyses the role of anaerobic digestion as a viable technology for the production of biomethane, as well as the future trends in biogas production in an international context. Chapter 3 and 4 focuses on Mexico and its potential to produce biomethane using several waste streams (pig slurry, grass and food waste) and energy crops (grass). The economic feasibility of a biomethane plant is evaluated using the net present value methodology. Chapter 5 and 6 analyses the laboratory work carried out over the research period. Each chapter is structured as an individual paper with introduction, methodology, results and discussion and conclusions. Chapters 3, 4 and 6 have been published in scientific peer reviewed journals while chapter 5 is currently in preparation for submission. A summary of chapters 2 to 6 is given as follows:

Chapter 2: A review of the current status of biogas technologies and future trends.
This chapter is a literature review of the status of anaerobic digestion plants. It includes a review of the preferred feedstocks for biomethane production, from first generation biofuels to advanced biofuels. This chapter highlights the vast array of waste streams and energy crops that can be used to produce renewable heat, electricity and transport fuels as well as the different technologies available in the market. The potential for the production of biomethane as a transport fuel is

discussed in detail. The chapter explores the anaerobic digestion applications on an international context, focusing mostly in the Americas and Europe. The European union has stipulated that 20% of all the energy used in Europe has to be renewable by 2020. This same directive requires that 10% of the energy used in transport has to be produced from a renewable source, favouring the use of wastes, non-food cellulosic materials and ligno-cellulosic materials. In the case of the Americas, anaerobic digestion has been less employed as compared to Europe. The use of biogas within the energy matrix in Mexico is particularly discussed. The costs of producing biomethane from wastes and energy crops as well as the capital costs of biomethane plants are presented in this chapter. The potential of third generation biofuels is also discussed. A literature review of the use of algae and micro algae is given, highlighting the advantages and bottlenecks of algae feedstock. It is suggested that the production and use of biomethane can be beneficial in order to decarbonise energy production.

Chapter 3. An economic and carbon analysis of biomethane production from food waste to be used as a transport fuel in Mexico.

Chapter 3 analyses the potential of food waste digestion in Mexico on a technical and economic outlook. It is the first part of two papers that study the feasibility of biomethane projects in Mexico. The contribution of greenhouse gas savings by displacing diesel with biomethane and its contribution to the CO_2 emissions reduction target is also examined in this paper. A review of the most recent literature at the time was undertaken in order to calculate the biomethane potential of food waste. The results calculated in this chapter are valid within the proposed conditions (feedstock, biomethane sale price, gate fees etc) and using the costs that were available at the time this study was conducted. Two scenarios were proposed to assess the economic feasibility of a

biomethane plant in a Mexican city. Scenario 1 consisted of a co-digestion plant using sewage sludge as a co-substrate. For scenario 2 pig slurry was chosen as co-substrate. The volatile solids ratio was taken at 80% food waste, 20% sewage sludge/pig slurry. By anaerobically digesting all the food waste generated in the country, 42 PJ per annum can be produced. If diesel consumption in transport is displaced by biomethane, 17.91 $MtCO_2e$ per annum can be prevented from being released to the atmosphere. The levelised cost of energy of scenario 1 and scenario 2 were calculated at $US 11.32/GJ and $US 14.38/GJ respectively. Scenario 1 achieved a positive net present value. The net present value for scenario 2 was negative. Gate fees are vital to the economic feasibility of biomethane projects. Currently there are no incentives for the production of gaseous biofuels in Mexico, therefore it is vital to find mechanisms that financially support these projects. It is suggested that the clean energy certificate scheme should be applied to increase the production of biomethane as a transport fuel.

Chapter 4. Can slurry biogas systems be cost effective without subsidies in Mexico?

This paper analyses the feasibility of biogas plants co-digesting pig slurry and grass without economic incentives as well as the resource and biogas potential of pig slurry. Biomethane projects in Mexico started in the mid 2000's encouraged by the clean development mechanism scheme proposed within the Kyoto protocol, however most biogas plants have reported low biogas production and several issues in design and maintenance. In this paper it was assumed that biogas is used in a power generator to upload electricity to the national grid. Three different scenarios were assessed. Scenario 1 consisted of an on-site anaerobic digester digesting pig slurry as is the common practice in the region. Scenario 2 consisted of an on-site biogas plant co-digesting pig slurry and grass. Scenario 3 consisted of a centralised biogas plant co-digesting pig

slurry and grass. A mix of 1:1 based on volatile solids was chosen. To analyse the economic feasibility the concept of net present value was used as in chapter 3. A review of the most recent literature at the time was undertaken in order to calculate the biomethane potential of pig slurry. The results calculated in this chapter are valid within the proposed conditions (feedstock, electricity sale price, gate fees etc) and using the costs that were available at the time this study was conducted. The design and operation of the proposed systems are given in detail.
The findings of this paper suggest that, on-site biogas plants are more profitable as compared to centralised plants. This is due to greater operational costs such as slurry transportation and grass harvest and ensiling. To increase economic feasibility, subsidies for the production of biogas are recommended. The energy potential of pig slurry was found to be quite significant at 21 PJ per annum. This paper also highlights the importance of co-digestion as a way to increase methane yields.

Chapter 5. Anaerobic co-digestion of *Ulva lactuca*, dairy slurry and grass silage for the production of gaseous biofuel in coastal regions.
This chapter examines the methane potential of *U.lactuca* co-digested with dairy slurry and grass silage in batch reactors (BMP) and continuous digestion. This green seaweed represents a recurring environmental issue in many countries where it is stranded on shores and left to decompose. A possible solution to this problem is to retrieve *Ulva* from beaches to be used as a feedstock for anaerobic digestion. *U.lactuca* has been suggested as a good feedstock for anaerobic digestion given its low lignin content and high methane yields. In this experiment, *U.lactuca* was collected in the summer months was used. The effects of increasing *U.lactuca* in the mix were analysed. Three different mixes based on volatile solids were tested finding that as the content of

U.lactuca increases the methane yield increases as well. Best mix was found at 70% grass silage, 25% ulva lactuca and 5% dairy slurry based on volatile solids. A 50 litre pilot scale CSTR was operated for a period of 16 weeks for the co-digestion of the three substrates at an organic loading rate of 2 kgVS/m^3/d. When the organic loading rate was increased to 3 kgVS/m^3/d methane yields decreased and levels of FOS:TAC (ratio of acidity to alkalinity) and volatile fatty acids increased. A content of *U.lactuca* no greater than 25% based on volatile solids in the mix at a loading rate of 2 kgVS/m^3/d is suggested as ideal for continuous co-digestion.

Chapter 6. Assessment of continuous fermentative hydrogen and methane co-production using macro- and micro-algae with increasing organic loading rate.

The application of a two stage continuous fermentative hydrogen and methane co-production using *Laminaria digitata* (macro algae) and *Arthrospira platensis* (micro algae) with increasing organic loading rates was examined in this chapter. Anaerobic digestion of algae biomass poses some major bottlenecks that have hindered its application on a larger scale. Organic substances such as polyphenols in macro algae and triglycerides in micro algae are not easy to degrade under anaerobic conditions. To overcome this a two-stage process is proposed in this work. Batch tests were performed to estimate the biohydrogen and biomethane potential of the feedstock. Four CSTRs with a capacity of 4 litres each one were used. The hydrogen reactor was operated at a retention time of 4 days. The effluent from the hydrogen reactor was fed into methane rectors A and B with a retention time of 12 and 24 days respectively. Reactor C worked as a stand-alone reactor with a retention time of 16 days to match the overall retention time of the first two-stage system, comprised of the hydrogen reactor and methane reactor A. Total volatile fatty acids (VFA), FOS:TAC and pH levels were analysed in detail. A retention time of 16 days allowed an

efficient process in hydrogen reactor at 6 kgVS/m^3/d and a steady anaerobic digestion process in methane reactor A at an OLR of 2 kgVS/m^3/d. Reactor C methane yields started to decrease at a loading rate of 2 kgVS/m^3/d, at the same time there was an increase in FOS:TAC and volatile fatty acids. At a loading rate of 3 kgVS/m^3/d the reactor failed. It was found that a two-stage system optimised hydrolysis and acidification of macro and micro algae, enabling methane production and improving stability in the second-stage anaerobic digestion.

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2. A review of the current status of biogas technologies and future trends

2.1. Development of anaerobic digestion

Anaerobic digestion (AD) is a biological process where organic matter is broken down by micro-organisms in the absence of oxygen, producing a combustible gas (biogas) and a rich nutrient substance (digestate) as end products. This process consists of four distinct phases: hydrolysis, acidogenesis, acetogenesis and methanogenesis. The first step is the hydrolysis of complex organic matter (carbohydrates, proteins and lipids) to basic monomers such as aminoacids, long chain fatty acids and monosaccharides. These products are further broken down by acidogenic bacteria (acidogenesis) to produce carbon dioxide (CO_2), hydrogen (H_2), volatile fatty acids (VFAs) and alcohols. In the third phase, acetogenic bacteria convert the products of acidogenesis to acetic acid, carbon dioxide and hydrogen. In methanogenesis, acetoclastic methanogens and hydrogenotrophic metanogens use acetic acid and hydrogen to produce methane and carbon dioxide. The produced biogas has a methane content of typically 50% to 60%.

AD is a mature technology that has been traditionally used to treat municipal wastewater and sewage sludge since the late 1800's. The first anaerobic reactors employed to treat these wastes were septic tanks that had low treatment efficiencies as compared to modern digesters. The main goal of these first plants was to treat and stabilise organic wastes rather than to produce biogas (Abbasi et al., 2012). It was until the 1970's during the oil crisis that this process started gaining the attention of researchers and the industry sector. This lead to a development of new digester technologies and designs, however the use of wastewater, sewage sludge and animal slurry continued to be the main sources of feedstock. In Europe in the 1990's, organic source segregation in municipal solid waste initiated the implementation of AD for the treatment of urban biosolids. At present AD is widely used in Europe as a mean to produce biogas for

electricity generation and more recently for gaseous biofuel (Torrijos, 2016). For this purpose, a wide array of substrates are used, ranging from organic wastes to energy crops. In developing countries, AD of animal slurry has been encouraged by the Kyoto's protocol clean development mechanism (CDM) (Lokey, 2009). The biogas produced in these AD digesters is recovered and burnt using industrial flare stacks or used in combined heat and power plants. This allows the biogas producers the sale of certificates of emissions reductions (CER) to a third party, increasing the economic attractiveness of these projects (Gutierrez et al., 2016).

In 2015, the number of biogas plants in Europe was 17,240 with 367 biomethane plants. Germany is the country that leads the sector with more than 10,000 biogas plants in operation (EBA). Most of these biogas plants use agricultural wastes as their main source of feedstock (Torrijos, 2016). The use the organic fraction of municipal solid waste (OFMSW) as a feedstock for AD is also growing in European countries with approximately 127 plants in operation. Germany and Spain lead in this category with 60% of all biogas plants that use OFMSW as the main substrate (Arsova, 2010). In the US there are approximately 239 biogas plants treating animal slurry and 1,241 wastewater treatment plants using AD. Unlike Europe, AD of energy crops and the organic fraction of municipal solid waste (OFMSW) has been less employed. In the Latin American region, Brazil and Mexico lead the biogas sector with approximately 57% of all of the CDM projects. Currently, there are more than 900 biodigesters in Mexico working under the CDM scheme, with most of these digesters using pig and cow slurry as its source feedstock. In the majority of these plants, the biogas produced is commonly burnt using industrial flare stacks in order to combust methane and release CO_2 ,which has a less global warming potential as compared to methane. During this process the energy produced in the form of heat is not recovered.

2.2. Biogas plants systems classification

In Europe there are basically two models of biogas plants operation, centralised and farm scale (Holm-Nielsen et al., 2009). On a centralised plant, feedstock is sent from different sources such as farms, canteens and wastewater plants via pipe or haulage to a single digester. After the AD process digestate is sent back to farms and used as fertilizer (Holm-Nielsen et al., 2009). Denmark started developing this concept in the 1980's, building their first centralised plant in 1984 in Vester Hjermitslev. This first plant was originally based on animal manure and slurry. The biogas produce was used for power and heat production (Raven and Gregersen, 2007). Farm scale digestion is commonly used in pig and dairy farms where the slurry is pumped to the biogas plant for treatment. This model is widely used in Germany where manure and slurry is usually co-digested with agricultural waste and energy crops (Wilkinson, 2011). There are approximately 9,000 farm scale plants in operation in this country and this number is expected to increase in the coming years (EBA).

There are different types of technologies and reactor configurations that are used in biogas plants. These configurations depend heavily on the feedstock used, varying from simple designs such as small scale farm scale digesters to complex systems such as OFMSW digesters, that are highly automated with special cleaning and size reduction requirements (Arsova, 2010).

According to their feeding scheme, AD reactors can be categorized as batch or continuous.

- In batch digesters feedstock is fed all at once and left inside for a set period of time for the reactions to occur. After this, the digester is emptied and filled again.
- In continuous digestion, feedstock is constantly fed and digestate is removed continuously (EPA, 2016). Most commercial AD OFMSW digesters use continuous digestion.

AD systems can be classified according to their solids content in wet and dry digestion.

- In wet digestion the content of dry solids (DS) inside the reactor is less than 15%. AD of animal slurry and sewage sludge are good examples of low-solids content digestion, given the liquid nature of these feedstocks.
- In dry digestion the content of DS is between 25 and 30%. AD of OFMSW is commonly carried out at high-solids content, however digestion of OFMSW can be carried out in wet systems through use of adequate water or leachate recirculation (Arsova, 2010).

According to their operating temperature AD digesters can be classified as mesophilic and thermophilic:

- Mesophilic processes operate between 30°C and 37°C.
- Thermophilic processes operate at 50°C and 65°C

Mesophilic digestion is considered easier to operate and to require less energy as compared to thermophilic digestion, nevertheless its efficiency is lower than that of thermophilic processes. Many modern biogas plants operate at high temperatures as this provides some advantages over mesophilic digestion. It produces higher biogas yields and has shorter retention times. However, this process also poses several disadvantages such as higher energy demands, and a higher sensitivity of thermophilic bacteria to toxins and environmental changes that affect the AD process (Arsova, 2010; Jain et al., 2015). According to the number of reactors employed, the systems can be classified in single stage and two stage.

- In a single stage digester, the four phases of AD (hydrolysis, acidogenesis, acetogenesis and methaneogenesis) occur at the same time in the same reactor vessel.
- In a two stage digester the AD phases occur in different rector vessels. In the first reactor hydrogen, alcohols and VFA's are produced favoured by a suitable pH for bacteria to

degrade carbohydrates and proteins. The effluent from the first reactor is subsequently fed to the second reactor where methanogenic archaea consumed them producing methane and carbon dioxide.

2.3. Operational parameters

The organic loading rate (OLR) is defined as the amount of substrate fed to a reactor per day per unit of volume. High OLRs could reduce the size of a digester, helping reducing its cost as a consequence, however high OLRs could also lead to process instability due to an accumulation of volatile fatty acids (VFAs), reducing methane yields and cause a drop in pH (Xie et al., 2011). Most sewage sludge AD plants operate at a loading rate of between 0.5 to 1.6 kgVS/m^3/d (Jain et al., 2015). For mono-digestion of grass silage OLRs of between 2 to 4 kgVS/m^3/d have been reported in the literature. However as the OLR increases the methane yield decreases (Lehtomäki et al., 2007; Wall et al., 2014), the most suitable OLRs are in the range 2 to 3 kgVS/m^3/d. AD of OFMSW finds its best performance at an OLR of 2 kgVS/m^3/d given that an increase in this OLR reduces methane yields (Browne et al., 2014).

Another critical parameter to take into account is the hydraulic retention time which is defined as the time that the feedstock remains inside a reactor. The longer the substrate stays in the reactor under ideal conditions the better the breakdown of organic matter, however the degradation rate decreases if the retention time is too long, signifying that there is an ideal retention time (Arsova, 2010). A short retention time may "washout" bacteria, impeding an efficient process thus reducing methane production (Jain et al., 2015). The appropriate retention time depends on the feeding regime and the type of feedstock used (Arsova, 2010; Jain et al., 2015).

2.4. Feedstock for AD

There is a vast range of organic material that can be used as feedstock for AD. The most common substrates used for AD are:

- Animal slurry and manure;
- Agricultural waste;
- Organic waste from industries (breweries, food processing industries);
- OFMSW;
- Wastewater and sewage sludge;
- Energy crops (grass silage, maize, sugar beet amongst others).

The use of a specific substrate is determined by its geographical location and accessibility. Some of these feedstocks require special pre-treatments in order to enhance their anaerobic digestibility. Basic concepts of AD of OFMSW, food waste, animal slurry, sewage sludge and grass silage are briefly discussed as follows:

2.4.1. OFMSW and food waste digestion

OFMSW consists of food scraps produced in households, restaurants and hotels,garden waste paper and contaminated food packaging. In AD of OFMSW screening for non-degradable material such as plastics and size reduction before AD is needed, increasing capital costs, maintenance and operation (Vasco-correa et al., 2018). OFMSW and food waste have an intrinsic variability and its composition depends on the location, type of collection and time of the year when it is generated. Solids content in OFMSW are between 25% and 34% and methane yields of between 274 LCH_4/kgVS and 367 LCH_4/kgVS have been reported in literature for waste not containing paper and packaging (Browne et al., 2014). In the case of food waste, methane yields

of 467 LCH_4/kgVS to 529 LCH_4/kgVS have been reported (Browne and Murphy, 2013; Zhang et al., 2007). As compared to OFMSW, food waste has higher methane yields given it does not contain garden waste which has a lower biodegradability index (Browne and Murphy, 2013). However, low C:N ratios in food waste and OFMSW can lead to process instability due to high total ammonia levels (Browne et al., 2014).

2.4.2. Animal slurry

AD of animal slurry has some advantages as compared to other substrates as they already contain anaerobic bacteria and a high water content (92%-96%) that acts as a solvent and ensures proper mixing and flow (Seadi et al., 2008). It also contains trace elements that are necessary in AD (Dennehy et al., 2016; Wall et al., 2014). Nevertheless, specific methane yields (SMY) are lower as compared to that of energy crops and OFMSW given that the organic matter in slurry has already been partially digested in the digestive system of the animal . In the case of dairy slurry, this substrate can have a methane yield of between 112 LCH_4/kgVS to 143 LCH_4/kgVS (Allen et al., 2014; Wall et al., 2013). For pig slurry yields of between 244 LCH_4/kgVS to 343 LCH_4/kgVS have been reported in the literature (Vedrenne et al., 2008). These yields are affected by several factors such as the time of the year when slurry is collected and the type feed the animal is given amongst others. The C:N ratio for dairy slurry is close to 20 (Wall et al., 2013) which makes it a good feedstock for AD . Unlike dairy slurry, pig slurry has a low C:N ratio of between 10 to 14.5, that could lead to ammonia nitrogen inhibition (W. Zhang et al., 2014).

2.4.3. Energy Crops

The use of energy crops is quite popular in European countries, where it is commonly co-digested with animal slurry (Rechberger and Lötjönen, 2009). Crops such as forage maize, willow and grass are particularly used in AD plants as co-substrates. Grass is considered to be a good substrate for AD given its high methane yields in the range of 245 LCH_4/kgVS to 400 LCH_4/kgVS and high dry solids yields per hectare (Gutierrez et al., 2016). Currently, around 50% of biogas plants in Germany use grass as feedstock (Wall et al., 2013).

2.5. Advanced biofuels

Biofuels from edible crops have been regarded by some as unsustainable, given that they compete with farmland for the production of food. For this reason, research on new substrates for the production of biofuels has developed. Third generation biofuels (also known as advanced biofuels) based on macro and micro algae may overcome issues associated with land based crop biofuels. Different algae species are found in temperate waters that could be collected in the open sea, bays and from beaches to be used to produce biofuels. Species such as *laminaria digitata*, *ascophyllum nodosum* and *ulva lactuca* have been used to produce biomethane in laboratory trials (Laurens 2017). However, the use of algae from natural sources involves several difficulties as they may be linked with variable climatic conditions that directly affect the characteristics and taxonomy of algae (Dębowski et al. 2013). A possible solution to this is to grow algae under controlled conditions either in open or closed installations, nevertheless, the cost effectiveness of these systems still being unfavourable (Dębowski et al. 2013). Algae, both micro and macro have higher yields as compared to agricultural crops, this is due to the more efficient photosynthesis of

algae that is three or four times greater than terrestrial crops, which leads to a higher organic matter production as compared to terrestrial plants (Rowbotham et al. 2012). Furthermore, they do not require arable land to be produced (Ding et al., 2016b; Dismukes et al., 2008). On top of that if anaerobically digested, the low lignin content in algae simplifies and facilitates the conversion to biogas (Jones and Mayfield, 2012). Like organic wastes and energy crops, algae composition varies throughout the year, having a direct impact on methane yields. DS from macro algae are in the range of 12% to 21% and SMY's are between 100 LCH_4/kgVS and 342 LCH_4/kgVS depending on the seaweed species, time of collection and pre-treatment applied (Tabassum et al., 2017). Despite these advantages, AD of algae has not been carried out on a large scale due to some significant issues that still restrict its application. High salt content in algae may inhibit the AD process (Tabassum et al., 2016) . Polyphenols and triglycerides are not easily digestible under anaerobic conditions and on top of that, rigid algae cell wall structures prevent an efficient break down. A possible solution to this is to split the process into hydrolysis and methanation using a two stage reactor for the production of hydrogen and methane (Ding et al., 2016b). This can improve the energy yield as compared to the solely AD process.

Algae is a promising feedstock for AD that can potentially surpass biogas produced from energy crops and wastes, however to achieve this more research is needed.

2.6. Future of biogas systems

Despite a recent stagnation in the biogas market in Germany given to changes that cut incentives for biogas production, AD is expected to grow globally. Countries like the UK where in recent years the number of AD plants has almost doubled, still maintain a policy that encourages biogas

production. In France the government has set a target of 1500 biogas plants by 2020, of which 1000 will be based on agricultural feedstock (Torrijos, 2016). The number of large-scale digesters is expected to increase not only in Europe but also in North America where the biogas industry is constantly growing (Vasco-correa et al., 2018). In Africa and Latin America the biogas industry will grow as well, although the use of small scale digesters will predominate in these regions (Torrijos, 2016). It is well understood that in order to boost the biogas industry favourable policies are needed. In the case of Mexico, new policies that encourage the use of renewables have been published by the federal government which could potentially increase the use of AD (DOF, 2008). The EU landfill directive has helped the implementation of AD of OFMSW, however incentives are still needed as AD technologies present financial challenges such as high operation and capital costs (Vasco-correa et al., 2018); these high capital costs are ameliorated by landfill taxes, which allow the biogas facility charge a gate fee . Biogas used in CHP plants will continue to be the most cost-effective self contained end use in the short term, however biomethane production (biogas cleaned and upgraded to natural gas quality typically with grid injection) could be significant in the long term (Vasco-correa et al., 2018). Currently this option is limited to some regions specially located in Europe. Regarding substrates, the use of ligno-cellulosic material and algae will be steadily more employed as biofuel production shifts towards these feedstcoks (Ward et al., 2014). Research on two stage digestion for the production of hydrogen and methane will continue to be assessed as this process has been identified as suitable for a variety of substrates such as algae, OFMSW and sugarcane bagasse amongst others (Baêta et al., 2016; Ding et al., 2018; Silva et al., 2018). Furthermore, the production of hydrogen and methane in a two stage process can increase overall energy yields as compared to single stage anaerobic digestion (Ding

et al., 2018). Nevertheless, despite all the above merits, its application on a greater scale remains a challenge.

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3. An economic and carbon analysis of biomethane production from food waste to be used as a transport fuel in Mexico

An economic and carbon analysis of biomethane production from food waste to be used as a transport fuel in Mexico

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Abstract

Biomethane produced from food waste is a potential fuel for urban buses in Mexico to reduce greenhouse gas (GHG) emissions in transport. Biomethane from food waste can potentially produce 42.32 PJ per year, equivalent to 6.5% of the energy content of diesel used in transport in 2015. By replacing diesel with biomethane from food waste, a reduction of 17.91 $MtCO_2e$ can be effected, 6.06% of the 2050 GHG emissions target. The economic feasibility of a biomethane plant for a Mexican city was investigated using two scenarios: co-digestion of food waste and sewage sludge (scenario 1); and co-digestion of food waste and pig slurry (scenario 2), both scenarios utilising anaerobic high density polyurethane digesters. Economic performance based on net present value (NPV) gave a positive outcome for scenario 1 with 33% of the revenue coming from gate fees. The levelised cost of energy (LCOE) for biomethane was $US 11.32/GJ ($US 40c/m^3 CH_4). Scenario 2 has a negative NPV; to break even (LCOE) biomethane has to be sold at $US 14.38/GJ ($US 51 c/m^3 CH_4). Biomethane from scenario 2 can be economically viable if a subsidy of $US 1.38/GJ is applied, equivalent to 5% of the cost of diesel.

Keywords: Food waste; Biomethane; Greenhouse gas emissions; Transport fuel; LCOE.

3.1. Introduction

3.1.1. Greenhouse gas emissions in Mexico

By 2050 Mexico is committed to reduce its greenhouse gas (GHG) emissions by 50% (287.92 Mt CO_2e) with respect to the emissions produced in the year 2000 (SEMARNAT, 2013a). To achieve this, a National Strategy for Climate Change (ENCC) was published in 2013 (SEMARNAT, 2013a). This strategy establishes a road map for new national policies and courses of action involving government departments and the private sector in compliance with the General Law for Climate Change (LGCC) (DOF, 2012; SEMARNAT, 2013a). The ENCC is primarily focused on the creation of economic instruments that promote the production and use of renewable electricity, sustainable development, energy efficiency and waste management (SEMARNAT, 2013a). Within the LGCC framework, large electricity consumers are obliged to purchase energy from renewable sources (clean energy certificates) equivalent to 5% of their annual electricity consumption (DOF, 2012). In 2016 the first auction of clean energy certificates (CECs) was carried out, reaching an average price of $US 45.4/MWh (KPMG, 2016). According to the National Inventory of GHG, in 2013, 665.30 Mt CO_2eq was produced in Mexico (SEMARNAT, 2015). The major contributor was the transport sector with a 26.17% share (174.15 Mt CO_2eq) followed by electricity generation with 19.02% (126.6 Mt CO_2eq) (Figure 3.1).

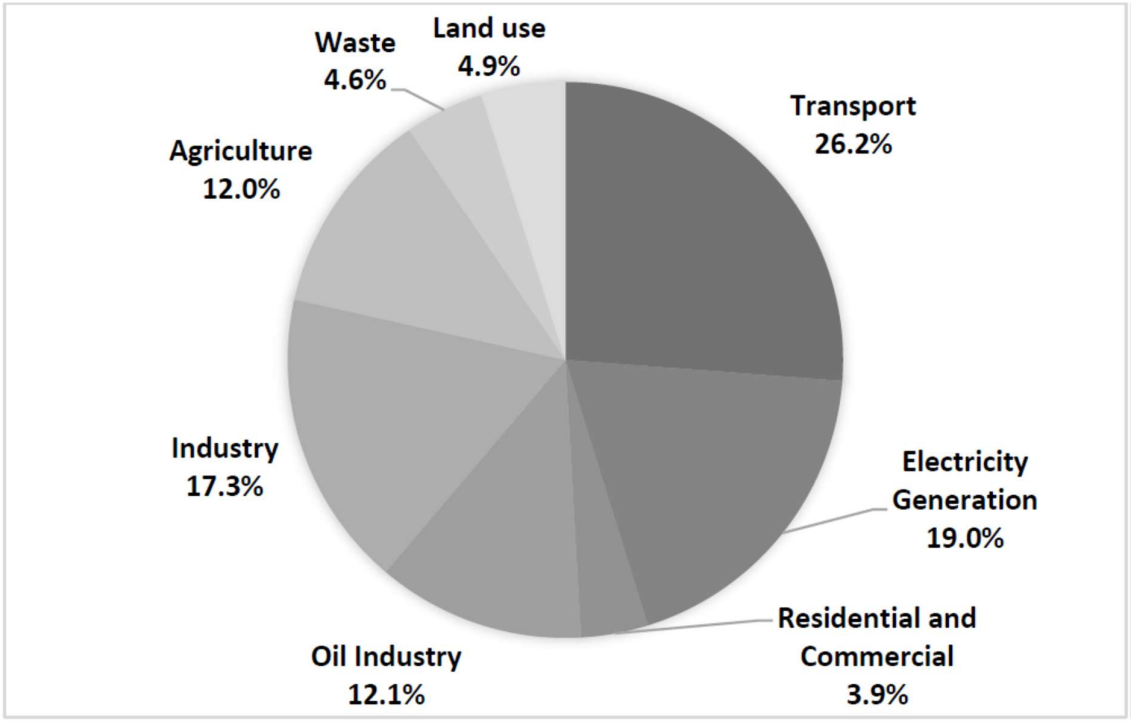


Figure 3.1. GHG emissions per sector in 2013 in Mexico (SEMARNAT, 2015)

3.1.2. Renewable energy in transport

Energy in transport accounted for 2,361.75 PJ in 2015 equating to 44.7% of the total energy consumption in the country (SENER, 2015). The three most consumed fuels were petrol (1,498.58 PJ), diesel (652.2 PJ), and liquefied petroleum gas (53.09 PJ). Natural gas consumption in this sector was only 0.83 PJ, clearly indicating the lack of natural gas vehicles (NGVs) and gas fuelling stations. As of 2015, the NGV fleet was estimated at 3,100 vehicles (SENER, 2016a). The use of renewable fuels (biodiesel and bioethanol) remains incipient. According to SENER (National Secretariat of Energy) the advances of biofuels in Mexico are focused on the design and implementation of new policies that encourage their production and use as described in the Law for the Promotion and Introduction of Bioenergy (DOF, 2008; SENER, 2016b). The introduction of ethanol within the energy matrix is set to commence in 2017 with a test trial that

proposes to introduce a 5.8% ethanol mix in petrol (SENER/CONUEE, 2016). The production of biodiesel is estimated to be 4,182 m^3/year and is undertaken in demonstration scale plants using mostly used oil as raw material (SENER, 2016b). The production of biogas reached 1.87 PJ in 2015, however, the majority of this gas was used to produce electricity (SENER, 2015). To date, no firm targets have been set for the production and use of biofuels, except ethanol blending.

3.1.3. Biomethane as a transport fuel

Biogas that has been upgraded (through the removal of CO_2) and cleaned from contaminants is termed biomethane. Biomethane can be obtained using biogas purification systems such as membranes, bio-filters and water scrubbers amongst other technologies. This gaseous biofuel can be used as a substitute for natural gas in NGVs. The use of natural gas as a transport fuel is growing worldwide. As of 2015 there were over 18 million vehicles running on natural gas (Khan et al., 2015). Countries such as Iran, Pakistan, China, Argentina and Brazil have developed a strong market for NGVs. In Europe natural gas is replacing traditional fuels (diesel and petrol) in buses and refuse collection trucks (Bord Gais, 2010). Sweden uses natural gas predominantly in transport, with 195 NGV refuelling stations (64 with biomethane) and ca. 60 biogas upgrading plants. The NGV fleet in Sweden is estimated at 44,000 vehicles (Fevre, 2014). In the city of Linkoping, biomethane has been used in all the urban city buses since 2002, sourced from an anaerobic digestion plant that treats a combination of household waste, animal manure, abattoir waste and industrial organic waste (IEA, 2005). In Mexico, the number of NGVs may be set to increase as SEMARNAT invests in projects to encourage the use of fuels with low carbon

footprint. An emphasis is placed on natural gas as it is considered “the cleanest and securest fuel in the country” (SEMARNAT, 2017).

3.1.4. Anaerobic digestion in Mexico: state of the art

Anaerobic digestion (AD) has been used in Mexico to treat municipal and industrial wastewater and sewage sludge since the 1980s. In the majority of these plants, the biogas produced is not recovered due to its low production and associated high costs (Monroy et al., 1998). Interest in AD has grown significantly in the last 15 years especially in the agricultural sector to treat pig, dairy and cattle slurry. This is a result of the Kyoto Protocol’s clean development mechanism (CDM), which allowed farmers to sell carbon credits (Lokey, 2009). The biogas plants are typically high-density polyurethane (HDPE) covered lagoons with no mechanical agitation. Slurry is pumped from the farms to the digesters without any pre-treatment. As of 2012, there were 966 bio-digesters treating animal slurries in Mexico (Weber et al., 2012). AD of the organic fraction of municipal solid waste (OFMSW) and food waste (FW) has been less employed. In 2015, the first OFMSW digestion facility was employed in the city of Merida. This plant mechanically separates the inorganic fraction of the municipal solid waste from the organic fraction. The biogas generated powers an internal combustion engine to produce electricity, which is sold to a third party using the national electric grid. The technology employed for this plant is similar to the aforementioned agricultural digesters in Mexico, consisting of a HDPE anaerobic pond with a cover on top to collect the biogas generated. Given the novelty of this system in the region, little information regarding design and operation is available.

3.1.5. Environmental justification for the digestion of FW

In Mexico and the Latin American region, urban waste production has become a major concern in recent years. Municipal solid waste generation in Mexico is increasing. From 2003 to 2011 it grew 25% as a result of urban growth, industrial development, prosperity of the population and a change in purchasing patterns (SNIARN, 2012). According to SEMARNAT in 2011, 41 million tons of municipal solid waste was generated in the country (0.94 kg/inhabitant/day) (SNIARN, 2012). Of this, approximately, 30% was FW (EPA, 2014); equating to 12.3 million tons of FW being generated every year. The global trend regarding this issue is to try to reduce FW at the source of origin. In Europe and the US, reduction targets for FW of 50% by 2030 have been set (Campoy-Muñoz et al., 2017; EPA, 2015). To achieve this ambitious goal, government agencies, policy makers, retailers and the food industry sector must work together to implement FW reduction strategies (ReFED, 2016). It has been reported that in the US, an initial investment of $US 1,800 M in grants, innovation technologies and project finance is needed to reduce 20% of FW within a decade (ReFED, 2016). At present, Mexico has no strategies or policies that encourage FW reduction. Furthermore, there is insufficient infrastructure for the production, distribution and commercialization of food, leading to waste. In households, consumers are not aware of the environmental benefits of reducing FW, making its reduction a complicated task. The common management practice in Mexico is to send this waste to landfills or to uncontrolled city dumps (INECC/SEMARNAT, 2012). FW decomposes slowly within the membrane cells of the landfill producing a gas with a methane content of typically 50% (EPA, 2011). As of 2012, only 17 of the 186 landfills in the country were burning the landfill gas or generating electricity in accordance with the clean development mechanism (CDM) framework (INECC/SEMARNAT,

2012). At present, only 35% of the landfills in Mexico comply with the applicable normativity (NOM-85), which results in major problems in the design, construction and operation that can lead to further air, soil and water pollution (EPA, 2011). Municipal solid waste contributed 3.2% (21.3 Mt CO_2eq) of the GHG generated in Mexico in 2013 (SEMARNAT, 2015) .Ideally, OFMSW and FW would be digested in bio-digesters, significantly reducing pollution and generating a clean, secure, sustainable gaseous biofuel.

3.1.6. Digestion of OFMSW and FW

Anaerobic digestion of OFMSW is a well-established technology in Europe. This is due to waste management policies (EU Landfill Directive) that required EU states to reduce the quantity of landfilled biodegradable material to 35% of that landfilled in 1995, by 2016 (EU Comission, 1999). FW is typically the largest component of OFMSW (Browne and Murphy, 2013).It is a readily biodegradable feedstock and its characteristics depend on eating habits, collection system, season and region (Browne and Murphy, 2013; C. Zhang et al., 2014). For the anaerobic digestion of OFMSW and FW, the feedstock has to undergo a series of pre-treatments where it is treated mechanically for separation of inorganic components such as plastics and metals. Subsequently, it is ground (or milled) to reduce its particle size and then diluted (Levis et al., 2010; J Rapport et al., 2008). Source segregated OFMSW and FW provides a feedstock of better quality with minimum contaminants (and associated reduction in pre-treatment processes required) as compared to mechanically sorted comingled OFMSW and FW (Browne and Murphy, 2013). As proposed by Browne and Murphy, source segregation may be effected by adding a specific bin for FW collection (Browne and Murphy, 2013). Specific methane yields (SMY) from FW are higher

than that of OFMSW making segregation of FW from the waste stream a more beneficial option (Browne and Murphy, 2013). Long term mono-digestion of FW and OFMSW has been described as difficult due to a lack of essential trace elements, which can lead to a failure of the process (Banks et al., 2012; C. Zhang et al., 2014) . To overcome these drawbacks, co-digestion with different substrates such as sewage sludge and animal manure have been suggested (C. Zhang et al., 2014). Pig slurry can increase biogas yields and improve process stability (through the in-built trace element content of the slurry) when used as a co-substrate in FW digestion (Zhang et al., 2011). Co-digestion with sewage sludge can increase buffering capacity, allowing the process to work at higher organic loading rates (Kim et al., 2011). In an industrial scale, OFMSW and FW are commonly co-digested with sewage sludge in wastewater treatment plants (Mata-Alvarez et al., 2011). The main driver for this is the use of wastewater treatment facilities that already have anaerobic digesters, reducing treatment costs of both wastes (Mata-Alvarez et al., 2000). At present in Mexico, digestion of sewage sludge is carried out as a way to stabilise the sludge for its final disposal. According to CONAGUA (Mexican National Commission of Water) there are approximately 2,337 wastewater treatment plants in the country, treating a flow of 111.25 m^3 of wastewater/s (34% of the total WW production) (CONAGUA, 2015). The sewage sludge generated per year is estimated to be 285,759 t dry solids (DS).

3.1.7. Feasibility of a biomethane plant

One of the major drawbacks in the employment of biogas plants is the high associated capital cost (CAPEX). Depending on the feedstock and the type of technology used, these costs can be as high as 380 €/t/a (418 $US/t/a) (Browne et al., 2011) . To increase the economic feasibility of such

plants, subsidies are required (Gebrezgabher et al., 2010). In the case of Mexico, grants by the federal government are given for the construction of new biogas plants. These grants can cover up to 50% of the plant's CAPEX provided that the feedstock is derived from an agricultural activity such as pig farming, however, it is not clear if these grants are applicable for co-digestion plants. Increasing the methane production per ton of feedstock can alleviate some of the economic constraints in developing biomethane plants. For this purpose, substrates with higher solids content and higher SMYs such as energy crops, OFMSW and FW are preferred. In the case of FW, OFMSW and sewage sludge gate fees can be accrued from waste management companies for disposal, creating an extra source of income that can help increase overall revenues. This gate fee does not apply to pig slurries, as the farmers are allowed to have their own waste treatment systems and final disposal sites in their facilities. Another important aspect in assessing the economic feasibility of biomethane is the levelised cost of energy (LCOE). A FW digester in Europe can have a LCOE of between € 59.4/MWh to € 99.9/MWh ($US 18.15/GJ to $US 30.5/GJ) (O'Shea et al., 2016), which means that biomethane has to be sold at a minimum price of $US 18.15/GJ to break even.

3.1.8. Innovation and objectives

The innovation in this study is that it is the first paper to assess the potential of biomethane as a transport fuel in Mexico based on co-digestion of food waste with sewage sludge and pig slurry and also assesses the potential GHG emissions savings. The objectives are to:

- Estimate the biogas potential of food waste in Mexico;

- Evaluate the GHG emissions reductions by replacing diesel fuel in transport with biomethane produced from the digestion of food waste;
- Analyse the economic feasibility of biomethane as a transport fuel for urban buses based on Net Present Value.

3.2. Methodology

3.2.1. Biogas yields from food waste

There are a number of methods to estimate the biogas potential of a given substrate such as FW and sludge from wastewater treatment plants without undertaking a biomethane potential test (BMP). These techniques include an ultimate analysis of feedstock and application of stoichiometric formulae (Buswell Equation), computer simulation and literature reviews (Curry and Pillay, 2012). To calculate the biogas potential from food waste in Mexico, relevant data from SEMARNAT was used along with a literature review. If the chemical composition of any given organic material is known, the quantity of methane that it can produce can be calculated from its stoichiometric formula (Buswell and Hatfield, 1936; Curry and Pillay, 2012). A typical ultimate analysis of FW is: 48% C, 6.4% H, 37.6% O, 2.6% N and 0.4 % S. The remaining 5% of DS is inert matter (Tchobanoglous et al., 1993). Using this data, the following formula for FW is obtained: $C_{21.5}H_{34.4}O_{12.6}N$. The theoretical yield of biogas can be estimated using the Buswell equation as shown in equation1:

$$C_aH_bO_cN_d + \left(\frac{4a-b-2c+3d}{4}\right)H_2O \rightarrow \left(\frac{4a+b-2c-3d}{8}\right)CH_4 + \left(\frac{4a-b+2c+3d}{8}\right)CO_2 + dNH_3 \quad \text{(Eq.1)}$$

According to the reviewed literature, FW is approximately 70% moisture and 90% of DS are volatile solids (VS) (Browne and Murphy, 2013; Zhang et al., 2007).

3.2.2. Scenario 1: Co-digestion of FW and sewage sludge

In order to analyse the feasibility of a co-digestion plant, the city of Merida in Mexico was taken as a model. Merida is located in the southeast of Mexico, with a total population of 830,732 inhabitants, making Merida the 12th most populated city in the country (SEDUMA, 2012). Approximately 832 tons of municipal solid waste are generated daily (ca. 1kg/inhabitant/day), which is collected by 126 vehicles with an average capacity of 4 tons (INEGI, 2013). The FW content in this waste for the city of Merida is 18.9 %, equating to 157.2 tons per day (EPA, 2014) . For the sake of this study it is assumed that FW is segregated at source in order to avoid the use of mechanical separation and the feedstock is provided by the municipality of Merida. In Scenario 1 FW is to be co-digested with sewage sludge. The amount of sewage sludge generated for a given wastewater treatment plant depends on the technology employed and the characteristics of the influent. In Mexico, several technologies such as activated sludge and trickling filters are used. There are 27 plants in Merida, which generate 218.2 t/day of thickened mixed sludge (see Appendix A, table A.1 for calculation). Sewage sludge has a SMY of between 160 to 310 LCH_4/kgVS (Koch et al., 2016; Nielfa et al., 2015; Rintala and Jarvinen, 1996; Yalcinkaya and Malina, 2015).For the purpose of this calculation an average of 235 LCH_4/kgVS was considered. As proposed by Rintala and Järvinen, a VS ratio of 80:20 of FW to sewage sludge will be used (Rintala and Jarvinen, 1996). Thickened sludge (8% DS content) is assumed in this calculation. The characteristics of the sewage sludge assessed is shown in Table 3.1.

Table 3.1. Sewage sludge and pig slurry generation

Feedstock	Daily production t/day [a]	DS t/day	VS t/day
Sewage sludge	218.2	17.3	13.4
Pig slurry	162.1	13.1	10.8

[a] Thickened at 8% DS content

3.2.3. Scenario 2: Co-digestion of FW and pig slurry

An area comprised of the municipalities of Merida, Conkal, Chicxulub and Tixcocob was taken into account for scenario 2, given that they are located in the most intensive pig farming area in the Yucatan state (Méndez et al., 2009). These towns have a total population of 928,924 inhabitants (INEGI, 2015). Pig farming is one of the major economic activities of this region, with 19 farms that generate 162.1 t/day of thickened pig slurry (see Appendix A, table A.2 for calculations). Similar to scenario 1, a VS ratio of 80:20 of FW to pig slurry is to be used. Pig slurry is to be transported by loading trucks within a radius of approximately 15 km. It is assumed that the slurry is thickened to 8% DS content prior to transportation to the biomethane plant (Table 3.1). The SMY of pig slurry is considered to be 307 LCH_4/kgVS on average (Gutierrez et al., 2016). As well as pig slurry, FW is to be collected and transported to the plant by loading trucks.

3.2.4. Economic analysis

The costs and revenues used in these calculations were taken from the available literature and from discussions with industry stakeholders and government officials. The price of land and the

sale of carbon credits were not taken into account. In order to analyse the economic feasibility of the different scenarios, the concept of net present value (NPV) was used as shown in equation 2:

$$NPV = -CAPEX + \sum_{t=0}^{N} \frac{R_t}{(1+i)^t} \qquad \text{(Eq. 2)}$$

where, NPV is the net present value, CAPEX is the total cost of the biogas plant, t is the period of time (years), N is the number of years considering the life span of the biogas plant (20 years), i is the discount rate (10%) and Rt is the positive cash flow in a year, (revenues-OPEX).

3.2.4.1. Capital Costs

The CAPEX of a biogas plant is related to the feedstock, plant size and technology employed (Browne et al., 2011). According to Smyth et al. a bio-digester of 50,000 t/year treating a mixture of animal slurry and grass silage costs ca. €110/t/a in Ireland (121 $US/t/a) (Smyth et al., 2010). A digester in Mexico treating grass silage and pig slurry in an anaerobic covered lagoon system can have a capital cost of $US 20/t/a, this is 16% of the capital cost of the Irish agricultural digester described by Smyth et al (Gutierrez et al., 2016). In the specific case of OFMSW and FW higher costs can be expected given the high solids content of the substrate and the need for processing of the feedstock to remove contaminants such as plastics and other inorganic materials. These activities are commonly done by the plant operators. It can be assumed that, as the content of dry solids in the feedstock increases, the CAPEX increases. In this study it is assumed that HDPE continuously stirred tank reactors are used. The cost of a digester is given in section 3.5. The CAPEX of a biogas upgrading plant varies depending on the size and the technology employed. A high pressure water scrubbing (HWPS) upgrading plant with a capacity of 250 m^3 of biogas/h can

have a cost of $US 1.29 Million (Urban et al., 2008). The CAPEX of a HWPS upgrading plant can be calculated using equation 3 (Gutierrez et al., 2016):

$$CAPEX = \left(0.0008 \times Plant\ Capacity\ \left(\frac{m3\ biogas}{h}\right)\right) + 1.0817 \quad \text{(Eq. 3)}$$

where CAPEX is given in Million $US.

In this study a compressed natural gas (CNG) service station is proposed to be built beside the biogas upgrading plant. There are several factors that directly affect the CNG cost, such as fuel demand from vehicle fleets, type of fill (fast or slow fill) equipment and installation (Smith and Gonzalez, 2014). The CAPEX of a service station can be calculated using equation 4, as proposed by the US National Renewable Energy Laboratory (NREL) (Smith and Gonzalez, 2014):

$$CAPEX = \left(39.782 \times Station\ Capacity\ \left(\frac{GGE}{Month}\right)\right) + 270031 \quad \text{(Eq.4)}$$

where CAPEX is given in $US and station capacity in gallons of gasoline equivalent (GGE) per month. For this study, a slow-fill station is considered. It is assumed that urban buses fill at night, avoiding the need of large storage tanks, thus reducing the CAPEX.

3.2.4.2. Operational Costs

The operational costs of a biogas plant vary depending on the type of technology used and size of the plant. Costs ranging from 3% to 16% of the CAPEX are suggested in the literature (Browne et al., 2011). In Ireland, according to Browne et al (2011), the OPEX for an agricultural plant is € 5/t of feedstock ($US 5.5/t), this cost increases to € 25/t of feedstock ($US 27.5/t) when the substrate

used is OFMSW. The OPEX of an agricultural digester in Mexico treating a mixture of pig slurry and grass is between $US 3.15 /t to $US 3.4 /t (Gutierrez et al., 2016). These costs include for energy consumption, wages, transportation, maintenance and grass ensiling. Similar to CAPEX, the OPEX of a biogas plant is directly related to the content of solids in the feedstock. As the dry solids content increases, the OPEX increases. The OPEX of a digester is given in section 3.5. The OPEX of a biogas upgrading plant accounts for energy consumption, maintenance, operation and labour costs (Gutierrez et al., 2016). Electricity consumption for biogas upgrading is taken at 0.25 kWh/m^3biogas and maintenance at 2% of CAPEX (B C Innovation Council, 2008; Urban et al., 2008). OPEX costs per m^3 of raw biogas are between $US c 2.04 to $US c 7.94 (without including the cost of capital) (B C Innovation Council, 2008). The OPEX of a CNG station takes into account maintenance, operation, energy consumption and labour. This cost can be calculated using equation 5 (Johnson, 2010):

$$OPEX = -2.225E10^{-7} * x^2 + 0.1257x + 7{,}014.3 \qquad \text{(Eq. 5)}$$

where x is the size of the CNG station in DGE/month (Diesel Gallon Equivalent per month).This equation assumes an electricity price of $US 0.10/kWh with a capacity charge of $US 12kW/month.

3.3. Results and discussion

3.3.1. Biomethane potential of FW

A SMY of 355 LCH_4/kgVS can be obtained from a digester with a food waste feedstock as described in section 2.1 ($C_{21.5}H_{34.4}O_{12.6}N$) with 70% VS destruction (Box 3.1).

$C_aH_bO_cN_d + \left(\frac{4a-b-2c+3d}{4}\right)H_2O \rightarrow \left(\frac{4a+b-2c-3d}{8}\right) CH_4 + \left(\frac{4a-b+2c+3d}{8}\right)CO_2 + dNH_3$
a=22, b=34, c=13, d=1

$C_{21.5}H_{34.4}O_{12.6}N + 7.35\ H_2O \rightarrow 11.5\ CH_4 + 10\ CO_2 + NH_3$

509.4 + 132.2 ⟶ 184.6 + 440 + 17
641.6 ⟶ 641.6
270 kgVS + 70.1 kg Water ⟶ 97.8 kg CH_4 + 233.2 kg CO_2 + 9 kg NH_3 [270 kg VS/t FW]

189 kgVDS_{dest} + 49.1 kg Water ⟶ 68.5 kg CH_4 + 163.2 kg CO_2 + 6.3 kg NH_3 [70% VS destruction]

Density of CH_4 = 16/ (22.412 m_n^3/kg) = 0.714 kg/ m_n^3

68.5 kg CH_4/0.714 kg/ m_n^3 = 96 m_n^3 CH_4

96 m_n^3 CH_4/ 270 kgVS = 0.355 m_n^3 CH_4/ kgVS = 355 LCH_4/ kgVS

Box 3.1. Theoretical methane yield using Buswell equation

This is lower than the yields reported by Browne and Zhang (Browne and Murphy, 2013; Zhang et al., 2012) and similar to the yields reported by Banks (Banks et al., 2008) as per Table 3.2.

Table 3.2. Biomethane potential from FW in literature

Author	Country	Substrate	SMY (m^3 CH_4/tVS)
Browne et al (2013)	Ireland	ssFW[a]	467-529
Zhang et al (2012)	UK	ssFW	445-456
Banks et al (2008)	UK	ssFW	370

[a] Source segregated food waste

Therefore, the yields used in this calculation can be seen as conservative. If all the FW generated in a year were digested, it could generate 1,178 Mm^3CH_4 with a total energy content of 42.32PJ (Table 3.3), this equates to 6.95% of the natural gas consumption in the industry sector in Mexico in 2015 and 6.48% of the energy content in diesel used in the same year in transport.

Table 3.3. Biomethane production from FW in Mexico

Municipal solid waste Mt/year	FW Mt/year	VS [a] Mt/year	SMY m^3 CH_4/tVS	Biogas Mm^3	Methane Mm^3	Energy PJ [b]
41	12.3	3.32	355	2,182.59	1,178.60	42.32

[a] 27% VS in FW
[b] 35.9 MJ/m^3 CH_4

This resource could fuel 25,026 urban buses, assuming that an average urban bus has an energy consumption of 1,691 GJ/year (Murphy, 2005). A total of 246 FW biogas plants with a capacity of 50,000 t/year would be necessary to produce 42.32 PJ in a year.

3.3.2. Simplified GHG savings

A simplified assessment of GHG emission reductions based on the use of biomethane as a transport fuel is presented in this analysis. This calculation does not take into account fugitive emissions related to the AD process. A more detailed analysis would require a life cycle assessment of biomethane production, which falls outside the scope of this work. Methane emissions from landfilled FW are calculated using the IPCC default method (Box 3.2), which gives the amount of methane released in a given year.

FW CH_4 emissions (Gg/year) = [(FW_T ● FW_D ● L_0) – R] ● (1-OX)

Where:
Gg= Gigagrams

FW_T= 12,300 (Total FW generated in Gg/year)

FW_D= Fraction disposed at designated sites (90%)

L_0= Methane generation potential = [MCF ● DOC ● DOC_F ● F ● 16/12] (CH_4/FW)

MCF= 0.832 (Methane correction factor)

DOC= 0.15 (Fraction of degradable organic carbon)

DOC_F= 0.77 (Fraction DOC dissimilated without including lignin)

F= 0.53 (Fraction by volume of CH_4 in Landfill gas)

R= 1 Gg/year (Recovered CH_4)

OX= 0 (Default oxidation factor for developing countries)

Thus;
L_0 = [0.832 ● 0.15 ● 0.77 ● 0.53 ● 16/12] = 0.0679 Gg CH_4/Gg FW

FW CH_4 emissions (Gg/year) = [(12,300 ● 90% ● 0.0679) – 1] ● (1-0)

FW CH_4 emissions (Gg/year) = 750.73

CO_2e emissions (Gg/year) = 15,765.54 (assuming CH_4 is 21 times more potent than CO_2)

Box 3.2. Greenhouse gases emissions from landfilled food waste (Adapted from (IPCC, 2000; SEMARNAT, 2013b))

Emissions for the direct combustion of diesel are given at 86.64 gCO_2/MJ (Ludwiczek and Neeft, 2012). CO_2 emissions from the biomethane plant are given at 96.97 kgCO_2/t FW (Jin et al., 2015), taking into account feedstock pretreatment and the AD process. Upgrading emissions are calculated at 145.67 gCO_2/m^3 of raw biogas treated, assuming an electricity consumption of 0.25 kWh/m^3 of biogas and an emission factor of 582.7 gCO_2/kWh (SEMARNAT, 2013b; Urban et al., 2008). By digesting 100% of the recovered FW generated in the country and displacing the diesel consumption of 42.32 PJ with biomethane, 17.91 MtCO_2e can be prevented from being released

to atmosphere annually (Box 3). Anaerobic digestion of FW for biomethane generation and its further use as a transport fuel, could achieve 6.06% of the 2050 GHG emissions target (Box 3.3).

Emissions reductions by displacing 42.32 PJ of diesel
= 3.66 $MtCO_2$ /year assuming 86.64 gCO_2/MJ for combustion of diesel

Emissions reductions by digesting 12.3 Mt of FW
= 15.76 $MtCO_2$/year (from Box 2)

Emssions produced by AD of 12.3 Mt of FW
= 1.19 $MtCO_2$/year assuming 96.97 $kgCO_2$/t FW

Emissions produced by the upgrading of 2,182.59 Mm^3 of biogas (from Table 3.3)
= 0.32 $MtCO_2$/year assuming 145.67 gCO_2/m^3 of raw biogas **GHG savings**

(3.66 $MtCO_2$/year + 15.76 $MtCO_2$/year) – (1.19 $MtCO_2$/year + 0.32 $MtCO_2$/year)

= 17.91 $MtCO_2$/year

Emissions target for Mexico
50% of 590.94 $MtCO_2e$ (baseline year 2000) = 295.47 $MtCO_2e$

(17.91 $MtCO_2$/year) / (295.47 MtCO2e) x (100%)= 6.06 %

Box 3.3. GHG emissions reductions

3.3.3. Scenario 1: Co-digestion of food waste and sewage sludge

A total of 15,492 tVS/year of FW (157.2 t FW/day) from the city of Merida is available in scenario 1. At a VS ratio of 80:20 (FW:sewage sludge), this requires 3,873 tVS/year of sewage sludge . In Table 3.4, a breakdown of the total resource in terms of energy content and an example case of a ca 60,000 t/a digestion plant is illustrated for scenario 1. This scenario can produce a total of 6.4 Mm^3 of CH_4 per year equivalent to an energy content of 230,113 GJ and could fuel 136 urban buses per year or supply the thermal energy needs of 11,079 houses (20.77GJ/house/year) (SENER/IEA, 2011). The average biogas yield is 95.8 m^3 /t of wet weight, which is similar to the yields of full scale OFMSW digesters reported in the literature (Joshua.

Rapport et al., 2008).Two centralised biomethane plants of approximately 60,000 t/year could potentially be constructed in this scenario. This plant size falls within the range of centralised anaerobic digestion plants in Denmark which varies from 20,000 t/year to 80,000 t/year (Browne et al., 2011; Singh et al., 2010). It is assumed that transportation of FW and sewage sludge is carried out by the municipality and the wastewater treatment operators respectively as this is the common practice in the region. This avoids the need of purchasing loading trucks, reducing CAPEX and OPEX substantially.

3.3.4. Scenario 2: Co-digestion of food waste and pig slurry

As per scenario 1, 15,492 tVS/year of FW is available in scenario 2 and for pig slurry, 3,873 tVS/year is required to match the 80:20 VS ratio. To facilitate transportation, it is assumed that pig slurry is dried to a DS content of 8%. A total of 6.68 Mm^3 CH_4 is produced per year with an energy content of 240,124 GJ in this scenario (Table 3.4). The resource could fuel 142 urban buses or provide the thermal energy needs of 11,561 houses. Two biogas plants with a capacity of approximately 60,000 t/year are proposed to be built. The average biogas yield is 102.5 m^3/t of wet weight. This figure is slightly higher than that of scenario 1 and similar to full-scale OFMSW digesters reported in the literature. Unlike scenario 1, transportation of pig slurry from the farms to the biogas plants is to be effected by the biogas plant operators. The purchasing of a hauling truck is considered in scenario 2.

Table 3.4. Feedstock and energy production for Scenarios 1 and 2

		Scenario 1		Scenario 2	
Food waste		Total production	ca. 60,000 t/year plant	Total production	ca. 60,000 t/year plant
VS	t/year	15,492	7,746	15,492	7,746
DS [a]		17,213.4	8,606.7	17,213.4	8,606.7
Wet weight		57,378	28,689	57,378	28,689
Sewage sludge [b]					
VS	t/year	3,873	1,936.5		
DS [a]		4,997.4	2,498.7		
Wet weight		62,467.9	31,233.9		
Pig slurry [b]					
VS	t/year			3,873	1,936.5
DS [a]				4,723.1	2,361.5
Wet weight				59,039.8	29,519.9
Total feedstock (wet weight)	t/year	119,845.9	59,922.9	116,417.8	58,208.9
Total DS		22,210.8	11,105.4	21,936.5	10,968.2
Biomethane production					
Biogas from FW	m^3/year	10,184,595	5,092,297.5	10,184,595	5,092,297.5
Biogas from sewage sludge		1,300,226.4	650,113.2		
Biogas from pig slurry				1,748,552.3	874,276.1
Total biogas		11,484,821.4	5,742,410.7	11,933,147.3	5,966,573.6
CH_4 from FW [c]		5,499,681.3	2,749,840.6	5,499,681.3	2,749,840.6
CH_4 from sewage sludge [c]		910,158.5	455,079.2		
CH_4 from pig slurry [c]				1,189,015.6	594,507.8
Total CH_4		6,409,839.8	3,204,919.9	6,688,696.9	3,344,348.4
Total Energy [d]	GJ/year	230,113.2	115,056.6	240,124.2	120,062.1

[a] Food waste 30%DS 27%VS ; Sewage sludge 8%DS 6.2%VS ; Pig slurry 8%DS 6.5%VS
[b] Calculated assuming a VS ratio of FW/pig slurry: sewage sludge of 80:20 respectively
[c] Food waste SMY 355 m^3CH_4/tVS; sewage sludge SMY 235 m^3CH_4/tVS; pig slurry SMY 307 m^3CH_4/tVS
[d] 35.9 MJ/m^3 Biomethane

3.3.5. Scenario 1 economic analysis

The costs given in this analysis are for a plant of ca. 60,000 t/year as described in Table 3.4. The initial investment of a FW and sewage sludge biogas plant is calculated at $US 349.71/tDS (see Appendix A, table A.3 for calculation). The costs in Appendix A were taken from a grass and

slurry digester under the assumption that the CAPEX of a plant is directly related to the dry solids content of the feedstock as described in section 2.4.1. A plant for scenario 1 will have a cost of $US 3,883,676 ($US 64.81/t/a) (Table 3.5). This is higher than the cost of an agricultural digester in the region ($US 20/t/a of feedstock) (Gutierrez et al., 2016), which clearly indicates the higher technical specifications of a digester treating FW, however, this cost is much lower than that reported by Browne et al (2011) for an OFMSW digester ($US 308/t/a). This is due to different specifications; higher costs in Ireland and the need of pasteurization of OFMSW before the AD process. The cost of the land for the biogas plant is not taken into account in this analysis as it is assumed that the developers of the project already own the site. The cost of the upgrading facility is calculated using equation 3, giving a total cost of $US 1,625,700 (680 m^3/hr). The size of the filling station was calculated by taking into account the annual energy production of the biomethane plant of 115,056 GJ (Table 3.4) equating to 9,588 GJ/month. This gives an energy content in GGE of 78,784.3/month, taking 1 GGE at 0.1217 GJ. The cost of the filling station is calculated using equation 4 giving a total cost of $US 3,404,228. The station is proposed to be built beside the biogas plant. The total CAPEX of scenario 1 is then calculated at $US 8,913,605 (Table 3.5). The operational cost of the biogas plant is charged at $US 51.78/tDS (see Appendix A, Table A.2). This cost includes for electricity consumption, maintenance and wages. This is approximately a third of the OPEX of a European digester (Browne et al., 2011). A cost of $US c 4/$m^3$ of biogas for the operation of the upgrading plant is assumed (Gutierrez et al., 2016)(Urban et al., 2008). This cost includes for maintenance, energy consumption and operation. The operational cost for a CNG station of 9,588 GJ/month (70,604 DGE/month, taking 1 DGE at 0.1358 GJ) is calculated using equation 5. This gives an operational cost of $US 14,780 per month equating to $US 177,361 per year. The OPEX of this scenario is given at $US 982,096 per year

(Table 3.5). FW transport is done by the municipality. For this, a gate fee of $US 12.13 ($ 224.5 MXN) per ton of waste is paid to the waste management company, which in this scenario is assumed to run the biogas plant. Sewage sludge transport is to be undertaken by the WWTP operators. The same gate fee as for FW is to be applied. This gives a revenue of $US 726,865/year. The income of the sale of biomethane is calculated at $US 1,587,781/year, taking the sale price of a GJ of compressed natural gas at $US 13.8 (Gas Natural Fenosa, 2017). In this calculation it is assumed that prices of natural gas are stable throughout the life span of the biogas plant (20 years). Within these conditions the NPV is $US 2,431,154 which means that the project is economically feasible (Appendix A, table A.4). The LCOE of this scenario is given at $US 11.32/GJ for the NPV to be zero (Appendix A, table A.4), which is lower than the one reported by O'Shea et al of 59.4 €/MWh (18.15 $US/GJ) for a food waste digester in Ireland (O'Shea et al., 2016). This is due to lower CAPEX and OPEX in the region. On the other hand the figure reported in this study is higher than the one reported by the British Columbia innovation council of $US 8/GJ, however, higher gate fees of between $US 15 to $US 22 per ton of waste were used in that study (B C Innovation Council, 2008). In this scenario, nearly 33% of the revenues come from gate fees which clearly indicates that this source of income is key on the feasibility of the project.

3.3.6. Scenario 2 economic analysis

The same methodology applied for scenario 1 is used in this analysis for scenario 2. The cost of a biogas plant of ca. 60,000 t/year is calculated at $US 3,835,722 ($US 349.71/tDS). The price of a loading truck in the region is given at $US 135,500 (Gutierrez et al., 2016), increasing the initial investment to $US 3,971,222.

Table 3.5. Economic analysis

Annual costs ($US/year)	Scenario 1	Scenario 2
CAPEX ($US)		
Biogas plant	3,883,676.20	3,971,222.27
Upgrading	1,625,700.00	1,641,700.00
CNG station	3,404,228.93	3,540,580.74
Total	8,913,605.13	9,153,503.01
Revenues ($US/year)		
Biomethane sales	1,587,781.42	1,656,857.11
Gate fee	726,865.89	347,997.57
Total	2,314,647.32	2,004,854.68
OPEX ($US/year)		
Biogas plant	575,038.61	567,938.29
Upgrading	229,696.43	238,662.95
Transport		10,977.82
CNG station	177,361.16	180,811.10
Total	982,096.21	998,390.16

As in the first scenario, the cost of the land for the biogas plant is not taken into account. The costs of an upgrading facility of 700 m^3 biogas/hr is calculated at $US 1,641,700. Similar to scenario 1 the size of the filling station was calculated taking into account the annual energy production of the biomethane plant of 120,062.11 GJ (Table 3.4), this is equal to 10,002.17 GJ/month (82,211 GGE/month taking 1 GGE at 0.1217 GJ). The cost of the filling station is calculated using equation 4, giving a cost of $US 3,540,581. The CAPEX is then calculated at $US 9,153,503 (Table 3.5). The operational cost of the biogas plant is charged at $US 51.78/tDS (see Appendix A, table A.3). This cost includes for electricity consumption, maintenance and wages. To allocate the biogas plant and minimise transportation routes a p-median solution was used. Biogas plants are to be built beside the marked farms (Figure 3.2), as the introduction of non-innocuous material to the farm grounds is not permitted due to food safety. The total distance is given at 58.7 km. Assuming that 50% of the traveled route the truck is fully loaded (41.5 t) and

the other half is empty (11.5 t), a total of 19.9 litres of diesel are needed per day (Table 3.6). Transport annual costs are given at $US 10,977/year and include for diesel consumption and maintenance.

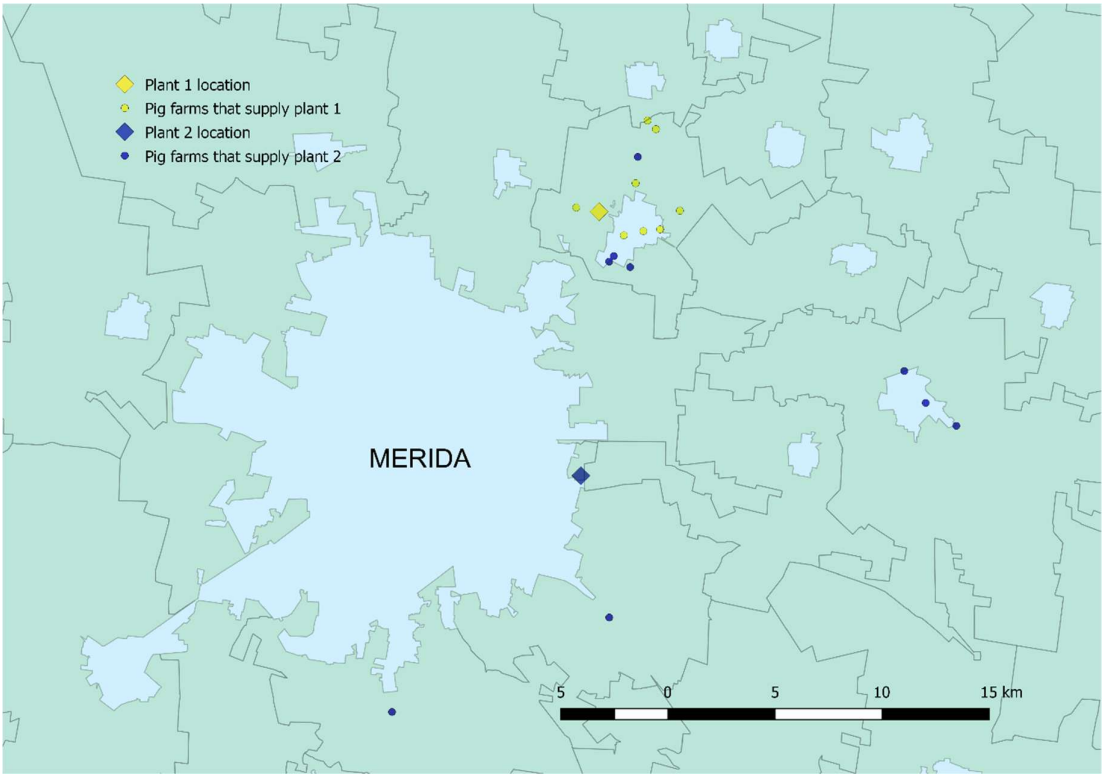


Figure 3.2. Farms and AD plant locations

Table 3.6. Transportation costs (adapted from Gutierrez et al., 2016)

km/day	Empty truck MJ/day [a]	Full MJ/day [b]	Total MJ/day	Litres of diesel/day [c]	Diesel cost $US/day [d]	Maintenance $US/day [e]	Cost $US/year
58.7	168.7	609	777.7	19.9	17.7	12.3	10,977.8

[a] Truck empty weight is 11.5 tons; total travelled distance is 29.35 km; energy intensity is 0.5 MJ/t/km
[b] Truck full weight is 41.5 tons; total travelled distance is 29.35 km; energy intensity is 0.5 MJ/t/km
[c] 39MJ/L
[d] $US 0.89/L
[e] $US 0.21 /km

A cost of $US c 4/$m^3$ of biogas for the operation of the upgrading plant is assumed (Smith and Gonzalez, 2014; Urban et al., 2008). Operation costs for a CNG filling station of 10,002.1 GJ/month (73,675.8 DGE/month taking 1 DGE at 0.1358 GJ) are calculated using equation 5. This gives an operational cost of $US 15,067.6 per month, equating to $US 180,811/year (Table 3.5). The OPEX of the plant is given at $US 998,390 (Table 3.5). Biomethane sales are calculated at $US 1,656,857/year, taking the price of a GJ of natural gas at $US 13.8 (Gas Natural Fenosa, 2017). In these conditions the NPV is $US -584,903 which means that the project is not economically feasible (Appendix A, table A.4). The LCOE is calculated at $US 14.38/GJ at a NPV of zero (Appendix A, table A.4). This price is lower than the cost of a GJ of diesel ($US 27.61) but higher than the cost of a GJ of CNG (Gas Natural Fenosa, 2017). If the same gate fee for FW is applied to pig slurry the NPV increases to $US 2,463,606 indicating the importance of this source of revenue. The increase in CAPEX and OPEX also affects directly the economic feasibility of the project.

3.3.7. The role of subsidies in biomethane projects

It has been suggested by some authors that in order to make biogas projects economically viable, subsidies are necessary (Gebrezgabher et al., 2010). This is especially significant for plants where the feedstock has no gate fee, such as animal slurries (scenario 2). These subventions can be given in several ways including single capital grants and feed-in tariffs (Cucchiella and D'Adamo, 2016). In this calculation, three different subsidies equivalent to 5%, 10% and 15% of the cost of a GJ of diesel ($US 27.61/GJ) were applied to the biomethane sale price and analysed. This is shown in Table 3.7.

Table 3.7. NPV including subsidies

	Scenario 1				Scenario 2			
	No subsidy	5% subsidy	10% subsidy	15% subsidy	No subsidy	5% subsidy	10% subsidy	15% subsidy
OPEX ($US/year)	982,096				998,390			
Revenues ($US/year)	2,314,647	2,473,425	2,632,203	2,790,981	2,004,854	2,170,540	2,336.226	2,501,911
NPV	2,431,154	3,782,921	5,134,689	6,486,457	-584,903	825,672	2,236,248	3,646,824

For scenario 1, NPV increases by $US 4.05 million for the 15% subsidy taking the no subsidy condition as a base line. By applying a 5% subsidy ($US 1.38/GJ) on scenario 2, the NPV becomes positive. Greater cash flows are achieved by increasing the level of subsidies to 10% and 15%, making scenario 2 economically feasible. As an example, these subsidies are significantly smaller as compared to the Biofuel Obligation Certificate (BOC) scheme used in Ireland for transport biofuels ($US 11.6/GJ to $US 32.4/GJ) (O'Shea et al., 2016). However, currently there is no scheme that allows energy producers to benefit from incentives for the production of renewable energy and transport biofuels in Mexico. An alternative solution would be to apply a scheme similar to the CEC in the electricity sector. In scenario 1, a subsidy of 15% the cost of a GJ of diesel ($US 4.14/GJ) is equivalent to 32% of the cost of a CEC in 2016 ($US 12.61/GJ), thus illustrating the benefit a similar economic instrument would have for the production of renewable fuels.

3.4. Conclusions

Mexico's strategy for reducing its greenhouse gas emissions is mainly focused on the production of renewable electricity, however the transition from diesel to less pollutant fuels in transport has been encouraged by government officials. Natural gas has been proposed as a diesel substitute, which can pave the way for a biomethane industry. Biomethane as a transport fuel from FW in Mexico has a significant theoretical potential (42.32 PJ/year). The use of biomethane as a transport fuel can help reduce Mexico's GHG emissions targets by 6.06% (17.91 $MtCO_2$). An urban biogas plant of a capacity of approximately 60,000 t/year co-digesting FW and sewage sludge (scenario 1) can have a positive NPV. When pig slurry is co-digested with FW (scenario 2) higher methane yields are achieved, however the NPV of this scenario is negative. The lack of gate fees for the handling and final disposal of pig slurry directly impacts the economic feasibility of scenario 2. For a Mexican city, the co-digestion of FW and sewage sludge is currently preferable from an economic standpoint. When subsidies are applied, the positive cash flow increases making scenario 2 economically feasible. Subsidies and gate fees are essential for the development of a biomethane industry in the country. Economic instruments such as the CEC in the electricity market can help in this regard.

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4. Can slurry biogas systems be cost effective without subsidy in Mexico?

Can slurry biogas systems be cost effective without subsidy in Mexico?

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Abstract

Biogas from pig slurry in Mexico has potential to produce 21PJ per year, equivalent to 3.5% of natural gas consumption in 2013. In this paper, three different scenarios are analysed: mono-digestion of pig slurry in a finisher farm (scenario 1); co-digestion of pig slurry and elephant grass in a finisher farm in situ (scenario 2) and co-digestion of pig slurry and elephant grass in centralised biogas plants (scenario 3). The digesters proposed are anaerobic high density polyurethane (HDPE) covered lagoons. HDPE centralised plants can be 5 times cheaper than European biogas plants. The economics of utilisation of biogas for electricity generation and as biomethane (a natural gas substitute) were investigated. Economic evaluations for on-site slurry digestion (Scenario 1) and on-site co-digestion of elephant grass and pig slurry (Scenario 2) showed potential for profitability with tariffs less than \$US 0.12/kWh$_e$. For centralised systems (Scenario 3) tariffs of \$US 0.161/kWh$_e$ to \$US 0.195/kWh$_e$ are required. Slurry transportation, energy use and harvest and ensiling account for 65% of the operational costs in centralised plants (Scenario 3). Biomethane production could compete with natural gas if a subsidy of 4.5 c/L diesel (1m^3 of biomethane) equivalent was available.

Keywords: Pig slurry; Elephant grass; Tropial Digesters; Biogas; Biofuel; Economic evaluation.

4.1. Introduction

4.1.1. Sources of energy in Mexico

Energy is a key factor for economic development. Mexico is the 10th largest oil producer in the world (BP, 2014) and the energy market is dominated by fossil fuels. In 2013, 88% of primary energy production (7,945 PJ) was derived from hydrocarbons; final energy consumption was 5,132 PJ (SENER, 2013a). Transport was the principal energy consuming sector with 44.1% (2,262 PJ) of final energy consumption in that year, followed by the industrial sector with 31.4% of the share (1,613 PJ). The demand for NG in the country is growing with the rise in the electricity and industry sectors. NG is progressively replacing oil as a source of fuel in power generation; the demand for NG increased 31 % in the 2002-2012 period (SENER, 2012). The use of NG in the transport sector is still developing, with approximately 4500 natural gas vehicles (NGVs) in operation in 2013. It is expected that the NGV fleet will grow to 255,500 vehicles by 2028 (SENER, 2014a) . According to SENER (Ministry of Energy) in 2013, in Mexico, 7% (636 PJ) of primary energy production was renewable (SENER, 2013a). The source of these energies was diverse (Figure 4.1), however, 59.6% (379 PJ) was obtained from the combustion of wood and sugarcane bagasse (SENER, 2013a). Wood remains the main source of renewable energy in Mexico and it is extensively used for heating and cooking purposes, especially in rural areas. The federal government has published a new law for the use and production of renewable energy in Mexico (LAERFTE), which states that 35 % of the electricity generated in the country by 2024 must come from a non-fossil fuel source and/or employ CO_2 sequestration (DOF, 2013). In a recent projection made by SENER, it is expected that by 2028 biogas and sugarcane bagasse will have a share of 4.8% of renewable electricity, equating to 4.7 TWh_e (SENER, 2014b).

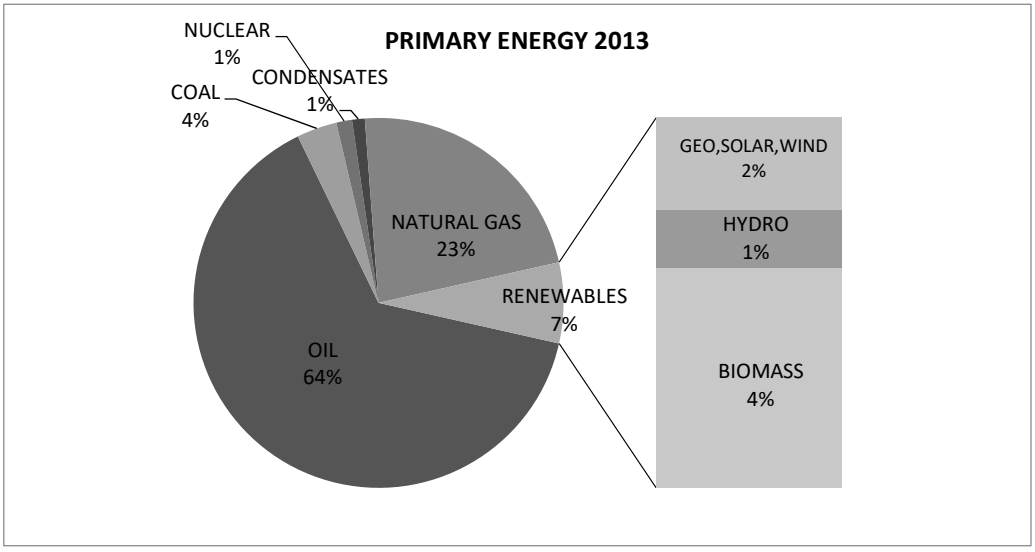


Figure 4.1. Primary Energy Production in Mexico for 2013 (SENER, 2013a)

4.1.2. Biogas and pig slurry treatment systems in Mexico

Biogas can be used as a substitute for natural gas once it is upgraded to biomethane. Biogas that has been upgraded to 95-97% methane content and has been scrubbed to remove water vapor, hydrogen sulfide, oxygen, ammonia, siloxanes, hydrocarbons and nitrogen is termed biomethane (Ryckebosch et al., 2011). Biomethane can be used: as a source of heat distributed via the natural gas grid; as a source of vehicle fuel; and in electric power stations. Compressed Natural Gas (CNG) is used extensively as a transport fuel in countries such as India. Landfill gas may be upgraded to biomethane for use as a transport fuel but there are difficulties as the gas is quite contaminated (Czyrnek-Delêtre et al.,2015). However, biogas from crops and slurries are more easily upgraded to biomethane, which has been used as a transport fuel (and a natural gas substitute) in countries like Germany, Sweden and Finland since the 20th century. Methane, which is the major component of biogas, can be used in Natural Gas Vehicles (NGVs) (Lampinen, 2014).

More pork is eaten in the world than any other meat. It is expected that in 30-50 years, meat consumption will double (McGlone, 2013). In Mexico, pig farming has increasing from 14 million pigs produced in 2000 to 15 million in 2010 (Garzón and Buelna, 2014). Pig farming activities produce large quantities of manure, often producing the waste equivalent of a small city (NRDC, 2013). The quantity and composition of manure vary depending on the feed, the age of the pigs and the type of farm. Manure production increases as pigs grow from feeders to finishers. Pig manure is made up of urine and faecal material (Ouellet-Plamondon et al., 2010). Typically, between 85-90% of dry solids (DS) are volatile solids (VS) (Hamilton et al., 2003). The most common way to treat slurries in Mexico is in open anoxic lagoons (Méndez et al., 2009), however, this trend has changed recently with the introduction of more high specification biodigesters (anaerobic covered lagoons which employ impermeable liners and a membrane cover to prevent escape of gas). The use of anaerobic digestion in the treatment of pig slurry prevents volatile organic compound emissions, controls odours and mineralizes nutrients (Bonmati and Flotats, 2002). The biogas produced, if it is upgraded to biomethane can replace NG (Thamsiriroj et al., 2011). The first large scale anaerobic covered lagoons built in Mexico were promoted by the clean development mechanism (CDM) functioning under the Kyoto protocol (Eaton, 2010; SAGARPA, 2010a).The main purpose of these systems was for the sale of carbon credits; the biogas generated was combusted in industrial flare stacks. According to REEMBIO (Mexican Bioenergy Network), there were 966 anaerobic digesters treating cattle and pig slurries in 2012 (Weber et al., 2012). Manure generated in farms is flushed through slatted floors to a collecting pit. In farms where there are no slatted floors, manure is sent to canals using hoses and then sent to a collecting pit. Slurries are subsequently pumped to an anaerobic covered lagoon.

Of late in Mexico there is a realisation of the potential to produce electricity from biogas. The first biodigesters to treat pig manure were built without agitation systems leading to low efficiencies and low biogas yield (SAGARPA, 2010a). A recent report showed that 47% of these digesters are not well designed while 61% of the digesters analysed had a biogas production lower than 80% of the value expected (SAGARPA, 2010a); low electricity generation efficiencies between 14% to 18% were also found during site visits. In a European Context electrical efficiencies between 30 and 40% would be normal (Murphy et al., 2011). Operational problems included: lack of removal of solids in the digester leading to blockages in pipes and pumps; persistent problems in gas blowers; operational problems in H_2S filters; short circuits in generators; equipment maintenance; and mixing system failures.

4.1.3. Potential for co-digestion of pig slurry with grass

Co-digestion of pig slurry and crops (residual or energy crops) can increase methane yields (Wall et al., 2013). Grass silage has a high VS content and is considered to be a good feedstock for AD, since it can decrease ammonia inhibition; maintain a suitable pH for methanogens and provide a better carbon/nitrogen ratio (Xie et al., 2011). Grasses are composed of lignin, cellulose and hemi-cellulose. Lignin is not easily degradable during AD (Montgomery and Bochmann, 2014; Seppälä et al., 2009). Methane yields between 253 m^3CH_4/tVS to 400 m^3CH_4/tVS can be expected in mono-digestion of grass (Seppälä et al., 2009; Wall et al., 2013). Due to the lack of trace nutrients in grass, biological failure may occur in long term mono-digestion (Thamsiriroj et al., 2012). Several types of grasses are used in Mexico as forage for grazing animals. Elephant grass and Napier grass have been used as a forage grass in recent years due to good DS yields and low

fertilization and water requirements (Améndola et al., 2005). In pig farms, the effluent of digesters is commonly used to irrigate grass, which can be later cut and sold to cattle farmers.

4.1.4. Benefits of centralised biogas plants

Centralised biogas facilities treat mixtures of animal manure, biodegradable feedstocks such as waste from the food industry, sewage sludge and the organic fraction of municipal solid waste (Hjort-Gregersen, 1999). Centralised plants have several advantages over farm scale plants. Improved technology can be used in centralised biodigesters; larger plants can benefit from economies of scale and farmers can delegate plant responsibilities to external operators (Raven and Gregersen, 2007), these external operators will have experience of other developments and employ the necessary skill sets based on previous knowledge. As of 2010 there were 23 centralised biogas plants in operation in Denmark with a total installed capacity of 50-600 m^3 manure per day. There were also 60 farm plants with a capacity of 5-50 m^3 per day (Lybæk et al., 2010). Alternatively, in Asia, most biodigesters are small scale and many are household plants. Latin America is developing a biogas industry supported by favourable policy frameworks (Seadi et al., 2008), however the use of centralised plants in both regions is not well documented. At present there are no centralised biogas plants in Mexico.

4.1.5. Requirement for cost effective digestion in tropical and less developed countries

Many European countries employ numerous digestion systems of high specification and associated high cost (Murphy et al., 2011). Subsidies are required to allow developers of biogas

plants to remain in production. These subsidies can be in the range of 15 to 25 US c/kWh$_e$. This is not feasible in tropical countries which are not wealthy. There is a need for simple cheaper technologies for treatment of wastes (such as slurries), that allow for clean water free from eutrophication associated with slurry run-off to water courses, and that provide sustainable decentralized renewable energy to large populations.

4.1.6. Aims and objectives

The aim of this study is to assess the potential for anaerobic digestion of pig slurry in Mexico and associated production and use of biogas. To the knowledge of the authors no paper has been published in peer review scientific press that analyses the potential of the biogas industry in Mexico; furthermore, there are few papers on cost effective digestion in tropical countries that are able to run free from tariffs. The objectives are to:

- Estimate the biogas potential of pig slurry in Mexico;
- Analyse systems which co-digest pig slurry and elephant grass;
- Assess potential for substitution of natural gas in Mexico;
- Assess the feasibility of centralised biogas plants.

4.2. Methodology

4.2.1. Biogas yield from pig slurry

To calculate the biogas potential from swine manure in Mexico relevant data from SAGARPA (Ministry of Agriculture, Livestock, Rural Development, Fisheries and Food) was used along with

a literature review. Several studies have been undertaken to assess the biogas yield potential of swine manure. The methane content in biogas from pig manure ranges between 63% to 71% (Eaton, 2009). Vedrenne et al. give a methane yield of 244 to 343 LCH_4/kgVS (Vedrenne et al., 2008). Other authors give similar methane yields (Appendix B, table B.1). An average of 307 m^3CH_4 /tVS is proposed taking into account methane yields reported in the literature. The quantity of manure excreted by animal is calculated using the ASABE (American Society of Agricultural and Biological Engineers) standards (Appendix B, table B.2) (ASABE, 2005). In order to give an estimation of the swine biomethane potential (SBP), it is assumed that the biomethane yield is proportional to the VS content in this waste (Rios and Kaltschmitt, 2013). Given this, the following formula is proposed:

$$SBP = TW \times VS_{per}W \times B_0 \times AF \times HV \quad \text{(Eq. 6)}$$

Where:

TW represents the total weight per year of swine production (live weight) according to the swine official census of 2012;

$VS_{per}W$ is the content of VS in manure per ton of animal weight per year, (calculated as 1.7 tVS/t of animal live weight per year from Appendix B, table B.2);

B_0 is the methane yield ratio per ton of VS, for this purpose, an average of 307 m^3CH_4/tVS was considered;

AF is the percentage of pigs kept in barns (only slurry produced within barns can be collected and used as a feedstock for large scale biodigesters) (Rios and Kaltschmitt, 2013); in this analysis, 70% of pigs are assumed to be kept in feedlots (Hernández et al., 2008);

HV is the lower heating value of methane (35.9 MJ/m^3).

Thus Equation 6 may be simplified as $SBP = TW * 13.115$ GJ

4.2.2. Scenario 1: Biogas generation using manure from a finisher farm

Finisher farms are the last stage in swine production. Pigs of weight between 25 to 120 kg (60 to 164 days old) are kept on these farms for 105 days on average. According to the ASABE, a finisher pig of 70 kg produces 0.375 kgVS/day (ASABE, 2005), this means 5.36 kgVS/day per 1000 kg of live weight. During site visits it was found that farmers register water consumption using flow meters, however, there were no wastewater (WW) discharge meter devices on these farms, therefore, the quantity of waste generated is unknown. The amount of WW produced will depend on how much water is used on the farm; how organic matter is handled (flushing and manual shoveling); the temperature and the feeding regime (Pérez, 2001). On a technified farm, flushing tanks are used twice per day on average, and each pen row has an independent flush tank, this means that there are two flush tanks for each barn ranging from 1000 litres to 2000 litres (Chastain and Henry, 2003). To calculate the volume of WW produced on a farm, data from Drucker et al was used (Drucker et al., 2003). Drucker et al calculated the volume of waste generated on farms in the Mexican state of Yucatan, taking into account animal weight and size of the farm; 16 litres per unit of animal population (UAP) for a big size farm is utilised. An UAP is equal to 100 kg of live weight. The system mass-energy flow is described in figure 4.2.

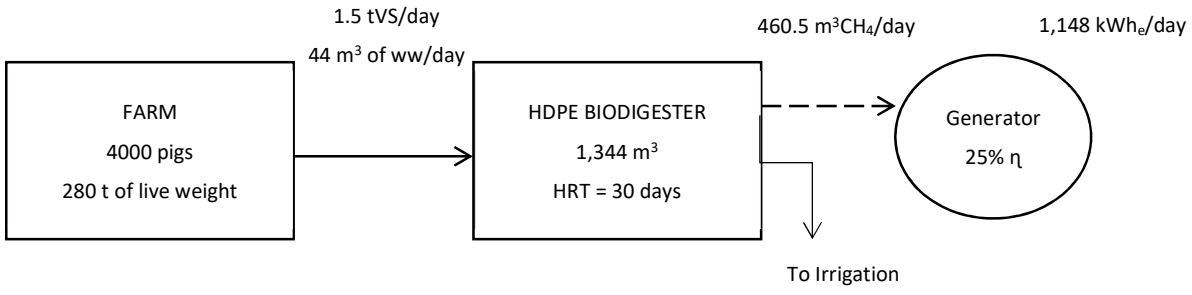


Figure 4.2. Scenario 1 mass-energy flow

According to SEMARNAT a hydraulic retention time of 30 days is necessary to achieve 60% VS destruction efficiency (SAGARPA, 2010b), taking this into account, a digester of 1,344 m^3 is proposed. The digester is a high density polyethylene (HDPE) anaerobic covered lagoon with a 7 kW_e submersible stirrer (0.005 kW_e per m^3 of volume) (Appels et al., 2008).

4.2.3. Scenario 2: Pig slurry and grass co-digestion on site

Elephant grass (pennisetum purpureum) has been introduced in the Mexican state of Yucatan in recent years as a fodder crop (Ramos Trejo et al., 2013). Yields of between 18 tDS/ha/year (without fertilizer) and 29 tDS/ha/year (using manure as fertilizer) have been reported (Binh and Nung, 1995). Elephant grass has a DS content of 20.2%, a VS content of 90.2% of DS, a C:N ratio of 26.6, a nitrogen content of 16.7 g/kg DS, a biogas yield of 579 m^3/tVS and a methane yield of 339 m^3CH_4/tVS (Frederiks, 2012).

This yield is higher than that of pig slurry on a wet basis. Elephant grass is ensiled prior to its addition into the digester. According to a literature review, grass silage bulk density varies with moisture content. A typical DS content in elephant grass silage is 36% (Ajayi, 2011).The density of grass silage is 0.684 t/m^3 calculated using Equation 7 (Curry and Pillay, 2012):

$$D = 1 - e^{\left(\frac{-0.3}{b-0.1}\right)} \qquad b \geq 0.15 \quad \text{(Eq. 7)}$$

Where D is the density of the material and b is the propotion of dry solids (expressed as a decimal). The same farm as scenario 1 is considered in this case. The digester has two submersible stirrers. As proposed by Wall et al (Wall et al., 2013)and Xie et al(Xie et al., 2011), a proportion of 1:1 of

VS of each substrate is used. To avoid high transportation costs and simplify logistics, grass is planted within the farmland. According to Xie et al, a HRT of 41 days for co-digestion plants treating a 1:1 mixture of grass and pig slurry is suggested to achieve 80% of maximum biogas generation (Xie et al., 2011). Taking this into account a digester of 2,330 m^3 is proposed. Two submersible stirrers of 6 kWe each are used to mix the reactor. The mass -energy flow of scenario 2 is shown in figure 3.

4.2.4. Scenario 3: Biogas generation in a centralised farm treating pig manure and grass

Centralised biogas plants of size between 20,000 t to 120,000 t per year have been reported in the literature (Browne et al., 2011; Hjort-Gregersen, 1999). In this case, three co-digestion plants with a capacity of 50,000 t/year are considered, similar to those proposed by Browne et al (Browne et al., 2011). Transportation of slurry is an important part of the process. Most Danish centralised plants are located in areas of high manure production, reducing distances and transport operating costs (Hjort-Gregersen, 1999). Mendez et al, analysed the amount of pig manure generated in the Mexican state of Yucatan, finding the most intensive manure area situated in the north part of the state, with a total daily manure production of 3,885 t (1.42 Mt/year) (Méndez et al., 2009). The municipality of Conkal has the largest density of manure per km^2 (2,539 kg manure/km^2/day) (Méndez et al., 2009). There are 19 pig farms in a radius of approximately 13 km, ranging from small to big farms with a total population of 24,746 pigs and a live weight of 1,546,203 kg (Appendix B, table B.3). The centralised biogas plants are proposed near the farm sites in order to reduce transportation costs. Some authors have suggested a maximum of 25 km distance

(Polifacio and Murphy, 2007). To calculate the number and capacity of the manure transport vehicles, relevant data from Drucker et al and Mendez et al was used (Drucker et al., 2003; Méndez et al., 2009). HDPE anaerobic covered lagoons with mechanical agitation is again considered. The mass-energy flow of scenario 3 is shown in figure 4.3

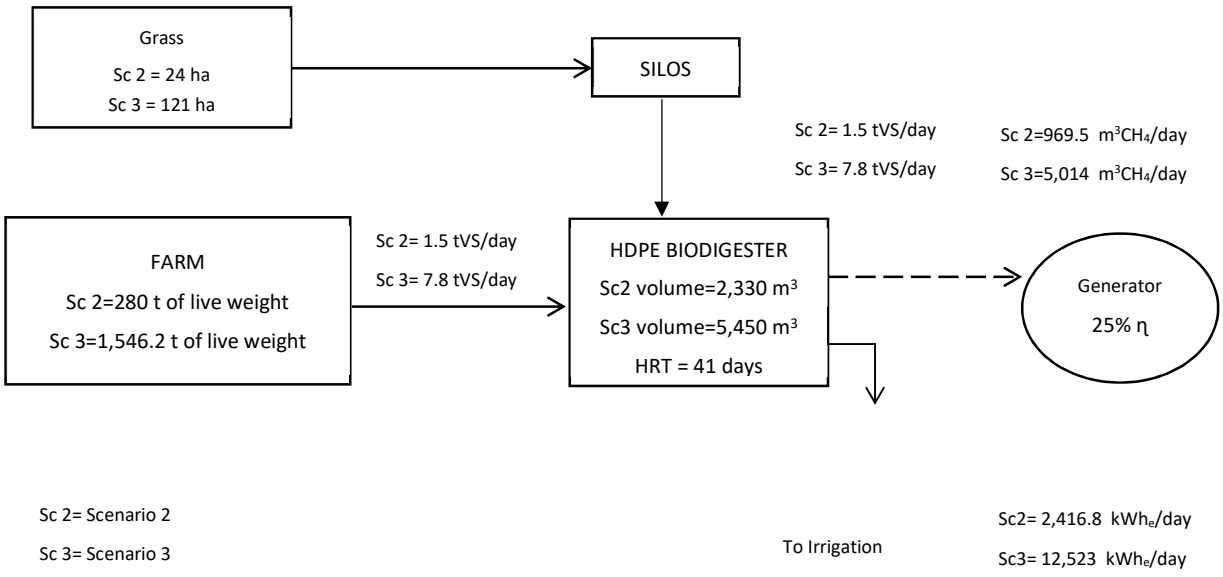


Figure 4.3. Scenario 2 and 3 mass-energy flow

4.2.5. Cost analysis

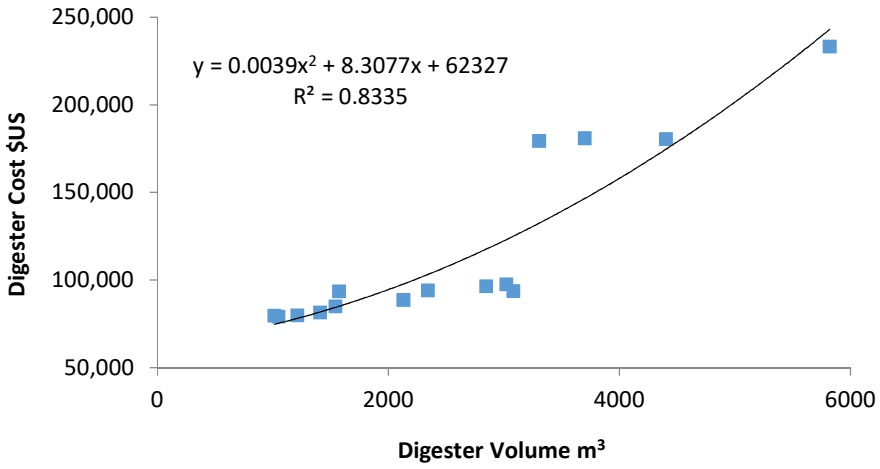
The costs and revenues used in these calculations were taken from the available literature and from discussions with industry, government departments and farmers in Mexico. The capital costs (CAPEX) of a biogas plant treating pig slurry will vary depending on the type of reactor and size and materials used in its construction and mixing system. For scenario 1 and 2 it is considered that a storage lagoon already exists in the site. Most of the biodigesters built in Mexico are anaerobic covered lagoons without agitation (SAGARPA, 2010a). According to a study carried out by the International Renewable Research Institute (IRRI) (Eaton, 2010), the capital cost of an anaerobic

digester for a medium to big size farm in Mexico is in the range of 40 to 78 $US/m^3 of digester volume (Figure 4.2 (a)). Capital costs included for civil works, electrical and mechanical installations. Currently there are no biogas upgrading facilities in Mexico or in Latin America; data from Urban et al (Urban et al., 2008) was used to calculate capital costs and operational costs of such systems (Figure 4.2 (b)).

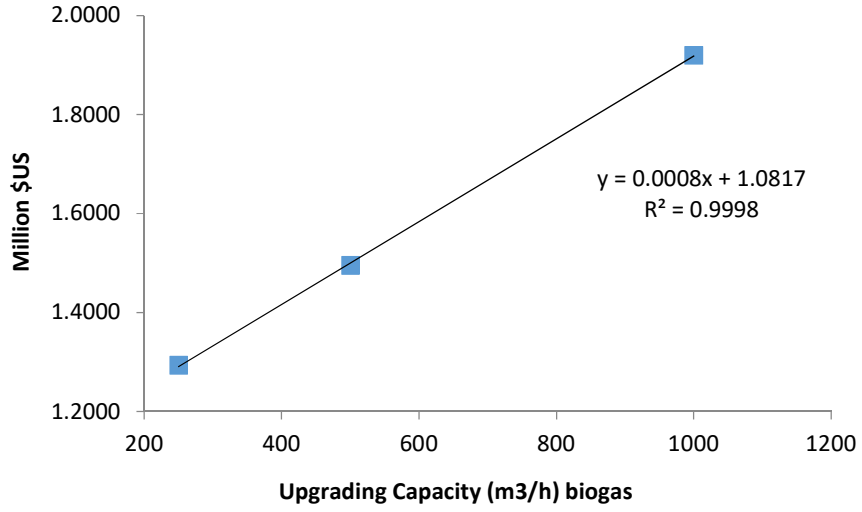
4.3. Results and discussion

4.3.1. Biogas potential from pig slurry

According to equation 6, Mexico with ca. 1.6M t of pigs in live weight (CNSPP, 2014) has a potential to generate an annual methane production from swine manure of approximately 21PJ (1.6Mt * 13.115GJ/t). This is equivalent to 584.6 Mm^3 CH_4 or 3.5 % of the natural gas consumption in the industry sector in 2013 (593 PJ) or 1.5 % of gas used in power generation in the same year (1,355 PJ). After upgraded to biomethane this gas can supply the thermal energy needs (cooking and heating) of 1,010,592 houses (20.77 GJ/house/year) (SENER/IEA, 2011). If used to generate electricity at 55% efficiency in combined cycle gas turbine (CCGT), 3207 GWh_e (11.5 PJ) can be produced. This is equal to the electricity consumption of 1,571,963 houses (2.04 MWh_e/house/year) (SENER/IEA, 2011) and 1.4% of the electricity consumption in 2013 (SENER, 2013a).



(a) HDPE biodigester capital costs (Eaton, 2010)



(b) Biogas ugrading facility capital costs (Urban et al., 2008)

Figure 4.4. Capital costs of HDPE digester and up-grading facilities

4.3.2. Scenario 1: Anaerobic digestion of slurry on site

Scenario 1 is based on a 4000 pig farm working 365 days per year with the "all in all out" system. Pigs are on average 70 kg each (mid cycle) generating 280 t live weight of pigs. From section 4.2.2, 5.36 kgVS/day manure is produced per ton of live weight; this means that 547.8 tVS is produced per year. The WW flow equates to 16 litres per day per UAP (per 100 kg live weight);

thus the WW flow is 44.8 m^3/day or 16,352 m^3/year. The concentration of the WW is 33.5 kgVS/m^3 (547.8 tVS/16,352 m^3). According to SEMARNAT a hydraulic retention time of 30 days is necessary to achieve 60% VS destruction efficiency (SAGARPA, 2010b), taking this into account, a digester of 1,344 m^3 is proposed. A total of 168,174 m^3CH_4/year can be produced at a specific methane yield of 307 m^3/tVS generating an energy content of 6,037 GJ/year. In this specific case, biogas is proposed to be utilised to produce electricity, given that the total biomethane production (19m^3CH_4/h) is too low to make an upgrading plant economically feasible (Warren, 2012). Assuming a conservative 25% efficiency in power generation, 419 MWh_e per year can be produced by a power generator of 55 kW_e operating 90% of the time. A submersible stirrer of 7 kW_e is used to mix the reactor (0.005 kW_e per m^3 of volume) (Appels et al., 2008). The farm of scenario 1 can provide the electricity demand of 205 houses. Pig farming is a high energy consumption activity. Approximately 0.107 kWh_e/day/100kg of live weight is used in a standard farm without ventilation. This requires 300 kWh_e/day, equating to 110 MWh_e/year.

4.3.3. Scenario 2: Pig slurry and grass co-digestion on site

A DS yield of 26.1 t/ha/year of elephant grass can be produced if land is fertilized at a rate of 300 kgN/ha/year (Ramos Trejo et al., 2013). A dry solids content of 20.2% yields a total harvest of 129 t/ha/year. Allowing for 90.2% VS yields 23.5 tVS/ha/year (Ajayi, 2011; Binh and Nung, 1995) . A significant amount of nitrogen can be obtained using the effluent of the digester in an irrigation system. A typical nitrogen content of digestate from a digester of a finisher farm is 1.67 kgN/m^3 (Garzón and Buelna, 2014). The nitrogen content of 1 m^3 of grass is 2.3 kg (calculated from Frederiks (Frederiks, 2012)). At 1:1 concentration, the digestate of this scenario has a

nitrogen content of 1.98 kgN/m^3. Thus 151 m^3 of digestate/ha/year (15 L/m^2) are required to satisfy the nitrogen requirement of elephant grass. The farm from scenario 1 produces 547.8 tVS/year, therefore, the same amount of VS of grass is needed.,If it is considered that 23.5 tVS/ha/year of elephant grass is produced, this means that 23.3 ha of land is necessary to produce 547.8 tVS/year. Co-digestion of these feedstocks on-site can produce 353,770 m^3 of methane with an energy content of 12,703 GJ (Table 4.1).

Table 4.1. Energy from grass and pig slurry co-digestion on site

Feedstock	t wet weight	tVS	Biogas m^3	m^3CH_4	Energy GJ
Pig slurry	16,352	547.8	258,730	168,175	6,037
Grass	3,005	547.8	317,256	185,704	6,666
Total	19,357	1,095.6	575,986	353,879	12,703

882 MWh_e can be generated at 25% efficiency with a power generator of 110 kW_e with a capacity factor of 90%. Electricity can be uploaded to the grid and sold to a third party. This energy can provide the electricity needs of 432 houses per year. If biogas is upgraded to biomethane, it could fulfil the thermal energy demand of 610 houses. For this purpose a biogas upgrading facility of 65 Nm^3/h is necessary, however, as reported by Warren (Warren, 2012), an upgrading plant smaller than 150 Nm^3/h is not economically feasible.

4.3.4. Scenario 3: Biogas generation in a centralised farm treating pig manure and grass

A total of 115,304 m^3 of slurry (see Appendix B and Table B.2) and 15,568 t of grass can be expected in this scenario. Three co-digestion plants of approximately 50,000 t/year can be built in the proposed area with a total methane generation of 1.83 Mm^3CH_4 per year. Scenario 3 can

generate 4,571 MWh_e per year at 25% electrical efficiency. This is equal to the electricity consumption of 2,240 houses per year. If upgraded to biomethane, it can provide the thermal energy consumption of 3,149 houses. A flow of 340 Nm^3/h of biogas is produced by the three plants, which is more than sufficient to allow for cost efficient biogas upgrading (Warren, 2012). In order to avoid high capital costs, the three digesters share the same biomethane upgrading plant. Approximately 121 ha of grass are necessary to produce 2,838 tVS for the three digesters. As in Scenario 2, 151 m^3/ha/year of digestate is necessary to reach the nitrogen fertilization rate. Pig slurry transport can be effected by three vehicles with a capacity of 30 tons. The biogas plant developer is responsible for all the investment and operational costs and slurry transportation. Farmers do not need to invest in biogas facilities. In this scenario, farmers need to have a slurry storage tank where transport vehicles could extract the waste. The farm collecting pit can be used for this purpose.

Table 4.2. Energy from grass and pig slurry co-digestion in centralised plants

	Feedstock	t wet weight per year	tVS per year	m^3CH_4	Energy GJ	Electricity MWh_e
Plant 1	Pig slurry	39,016	787	241,713	8,678	603
	Grass	4,317	787	266,908	9,582	665
	Total	43,334	1,575	508,622	18,260	1,268
Plant 2	Pig slurry	39,550	1,119	343,544	12,333	856
	Grass	6,136	1,119	379,355	13,619	946
	Total	45,686	2,238	722,899	25,952	1,802
Plant 3	Pig slurry	36,738	932	286,046	10,269	713
	Grass	5,115	932	315,860	11,339	787
	Total	41,852	1,863	601,905	21,608	1,501

4.3.5. Cost analysis of Scenario 1

The cost of the HDPE digester can be calculated using the equation in Figure 4.2. (a), generating a cost of $US 80,537. This cost is similar to the one reported by FIRCO in a case study of a farm in the state of Yucatan, where the CAPEX of a 3,336 m^3 HDPE digester was calculated at $US 151,890 excluding power generation (Aviña, 2013). An anaerobic covered lagoon of 3,642 m^3 in New Zealand has a cost of $US 112,132, including for a power generator of 43 kW_e (Craggs and Heubeck, 2008). This value is significantly lower than the average Mexican covered lagoon. From conversations with industry the cost of a power generator is priced at $US 1,100/$kW_e$ including installation and synchronization panel ($US 60,500 for 55 kW_e generator). A submersible stirrer is priced at $US 1042/$kW_e$ including for installation, lifting device and guide rail ($US 7294 for 7 kW_e). The CAPEX of the plant is thus calculated at $US 148,331 ($US 9/t of feedstock). Browne et al reported a cost of $US 117/t (110 €/t) of feedstock for a European biogas plant (Browne et al., 2011). This suggests a HDPE anaerobic covered pond for pig slurry is 12 times cheaper than an average European biodigester. This difference can be due to the use of a simple technology and cheap materials. In its basic principle, an anaerobic covered lagoon is a waterproof wastewater pond with a cover on top to collect the biogas generated. Relatively high temperatures in tropical regions allow the reactor to work within a mesophilic range without the need of heating systems; decreasing initial investments and energy consumption.

Biodigester maintenance cost is suggested at 3% of the biodigester cost per year ($US 2,416/year) (Eaton, 2010) and generator maintenance is priced at $US 60/$kW_e$/year equating to $US 3,300/year. Stirrer maintenance is given at $US 125/$kW_e$ ($US 875/year). Electricity consumption is calculated at $US 21,372/year excluding VAT (see Appendix B, Table B.3). The OPEX for this

scenario is $US 27,963. The electricity is uploaded to the national grid and sold to a third party. For this, the National Electricity Company charges a rate of grid use of $US .0081/$kWh_e$ transmitted (CRE, 2015). The price of a kWh_e is variable and it is dependable on the cost of fuels used in its production (SENER, 2013b). A net present value (NPV) analysis is given in order to evaluate the economic feasibility of the project (equation 8).

$$NPV = -CAPEX + \sum_{t=0}^{N} \frac{R_t}{(1+i)^t} \qquad \text{(Eq. 8)}$$

Where,

NPV is the net present value, t is the period of time (years), N is the number of years considering the life span of the power generator (15), i is the discount rate (7.5%) (Herrera, 2008; Romero, 2011) and R_t is the positive cash flow, (revenues-OPEX).

To break even (see Appendix B, Table B.4) electricity has to be sold at $US 0.115/$kWh_e$. FIRCO can give grants of 50% of the digester cost and up to 50% of the cost of the power generator (FIRCO, 2015). Applying these grants, the CAPEX of the project decreases to $US 77,813 allowing a breakeven price of electricity of $US 0.096/$kWh_e$.

4.3.6. Cost analysis of Scenario 2

The digester has a volume of 2,330 m^3, which gives a cost of $US 102,856. This digester has two submersible stirrers of 6 kW_e each, giving a total of $US 12,504. For this scenario it is considered that a storage lagoon already exists on site. This scenario requires a 110 kW_e generator, which has a total cost of $US 121,000. The cost of an irrigation system is priced at $US 1,358/ha. For this facility 24 ha will be required to be irrigated costing $US 32,592. According to Smyth et al

(Smyth et al., 2010) a silo of 25 m long, 2.1 m height and 10 m wide has a capacity of 360 t of fresh silage. A total of 9 silos are required, giving a total cost in the region of $US 84,305. Land preparation and grass planting have a cost of $US 25,200 ($US 1050/ha) (Guerrero and Enriquez, 2011). The CAPEX of the plant is priced at $US 378,457 ($US 20/t). The plant operational costs are $US 63,558 per year, including electricity consumption and maintenance (Table 4.3 and Appendix B, Table B.5).

Table 4.3. Operation and maintenance costs of scenario 2

Biodigester	Mixers	Generator	Irrigation	Harvest and Ensiling	Energy	Total
3,086	1,500	6,600	2,520	16,656	33,196	63,558

Prices in $US per year
Biodigester maintenance is 3% of biodigester cost
Mixer maintenance is priced at $US 125 /$kW_e$
Generator maintenance is priced at $US 60 /$kW_e$
Irrigation system maintenance and operation is priced at $US 105 /ha/year
Harvest and ensiling is priced at $US 694/ha/year

As for scenario 1, electricity is uploaded to the national grid and sold to a third party. To break even, electricity has to be sold at $US 0.129/$kWh_e$.

FIRCO can provide grants for energy crops of up to 30% the total cost of the project; taking this into account and applying the grants for biodigester construction and power generator, the CAPEX decreases to $US 258,969 (Appendix B.Table B.4). With this new figure, electricity has to be sold at $US 0.114/$kWh_e$ to break even.

4.3.7. Cost analysis of Scenario 3

4.3.7.1 Electricity sector

Table 4.4 outlines the CAPEX of the 3 biogas plants using the same unit costs from scenario 2. The cost of a HDPE storage lagoon and a collecting pit are priced at $US 20.5/$m^3$ included for civil works and membrane installation. The collecting pit has a submersible chopper pump of 3.7 kW_e, which is priced at $US 1,419 per kW_e and includes for installation and lifting device. For this scenario an electric substation is considered. The cost of a substation in the region is given at $US 160 per KVA, including transformer, accessories and installation. The CAPEX of the 3 plants are priced at $US 812,072 ($US 19/t of feedstock); $US 997,438 ($US 22/t of feedstock) and $US 884,136 ($US 21/t of feedstock) respectively (Table 4.4). The initial investment is heavily influenced by the power generator size, which accounts for approximately 30% of the CAPEX.

Transportation costs, energy consumption and harvest and ensiling account for approximately 65% of the operational costs (Table 4.5). The same operation and maintenance costs from scenario 2 were used.

To calculate the operational costs, it was necessary to analyse the transportation routes; wages, truck maintenance and fuel consumption. Plant 1 route has a total of 175.5 km (total traveled distance). Energy intensity in transport for loading trucks is given at 0.5 MJ/t/km (SENER/IEA, 2011), the empty truck weight is 11.5 tons. Assuming that 50% of the traveled distance the truck is fully loaded (41.5 t) and the other 50% is empty (11.5 t), a total of 60 L of diesel is needed (diesel energy content taken at 39 MJ/L), giving as a result a daily expenditure of $US 55, taking the diesel price at $US 0.92/L (“Mexico Diesel Prices, liter,” 2015).The same methodology was

used for Plant 2 (150.5 km) and plant 3 (120 km). Transport costs are influenced not only by the farm distance to the plant, but also by the size of the farm. Small farms generate less slurry as compared to big farms. This means that more small farms are needed to reach the feedstock rate proposed, increasing distance and fuel consumption. Truck maintenance is given at $US 0.21/km and includes for spare parts, regular services and tyre change. The entire vehicle fleet is changed after 450,000 km as recommended by local transport companies. As well as in scenario 2, electricity is uploaded and sold to a third party. It is important to highlight that unlike scenario 1 and scenario 2, FIRCO does not give grants for biogas plant developers (FIRCO, 2015).

For plant 1 electricity has to be sold at $US 0.195/kWh$_e$ to break even. Plant 2 and plant 3 require an electricity price of $US 0.161/kWh$_e$ and $US 0.169/kWh$_e$ respectively (see Appendix B, Table B.4). Smyth et al analysed the economic feasibility for a grass biogas plant with electric generation on site and found that an electricity price of between € 0.196/kWh$_e$ to € 0.256/kWh$_e$ ($US 0.2078/kWh$_e$ to $US 0.2714/kWh$_e$) was necessary to break even (Smyth et al., 2010). These figures are higher than the ones reported in this study; reflecting the higher specification employed. If electricity is sold to the commercial, industrial or agricultural sectors, profits can be obtained.

Table 4.4. Biogas plants CAPEX

	Plant 1		Plant 2		Plant 3	
Digester	Volume m^3	$US	Volume m^3	$US	Volume m^3	$US
	5,092	205,751	5,450	223,444	4,967	199,809
Generator	kW_e	$US	kW_e	$US	kW_e	$US
	160	176,000	230	253,000	190	209,000
Land	ha	$US	ha	$US	ha	$US
	34	35,700	48	50,400	40	42,000
Silo	Unit	$US	Unit	$US	Unit	$US
	12	111,072	17	155,685	14	137,840
Mixer	kW_e	$US	kW_e	$US	kW_e	$US
	26	27,092	28	29,176	25	26,050
Lagoon	Volume m^3	$US	Volume m^3	$US	Volume m^3	$US
	2,546	52,193	2,725.00	55,863	2,483	50,902
Irrigation system	ha	$US	ha	$US	ha	$US
	34	46,172	48	65,184	40	54,320
Collecting pit	Volume m^3	$US	Volume m^3	$US	Volume m^3	$US
	224	4,592	253	5,187	230	4,715
Electric substation	KVA	$US	KVA	$US	KVA	$US
	112.5	18,000	150	24,000	150	24,000
Truck	Unit	$US	Unit	$US	Unit	$US
	1	135,500	1	135,500	1	135,500
CAPEX	812,072		997,438		884,136	

4.3.7.2. Biomethane production

The cost of an upgrading biogas plant is calculated using the equation in Figure 4.4 (b). In this case an upgrading capacity of approximately 350 m^3/h is needed, giving a total cost of $US 1,361,700. The CAPEX of the 3 plants is recalculated excluding the costs of power generators and

including the upgrading plant. The electricity consumption of the upgrading plant is 0.25 kWh_e/m^3 (Urban et al., 2008).

Table 4.5. Operational costs

	Plant 1	Plant 2	Plant 3
Digester	6,173	6,703	5,994
Generator	9,600	13,800	11,400
Mixers	3,250	3,500	3,125
Irrigation	3,570	5,040	4,200
Pit and pump	960	960	960
Lagoon	460	460	460
Substation	400	400	400
Harvest and Ensiling	23,596	33,312	27,760
Transport	33,474	28,706	22,698
Energy*	35,253	41,204	36,222
Wages**	19,515	19,515	19,515
Total	136,250	153,600	132,734

*Tariff OM ($US 0.0847/$kWh_e$ and a monthly charge of $US 12.24 KW_e of demand)
**3 Workers for each plant and a wage of $US 17.8 per day

Given this, an electric substation of 150 KVA is necessary with a cost of $US 24,000 yielding a CAPEX of $US 2,079,647. Maintenance is given at 2% ($US 27,234) of the upgrading plant per year and operation is priced at $US 7,120/year (Urban et al., 2008). Substation maintenance is $US 400/year and electricity consumption is $US 77,775/year with a maximum demand of 87.5 kW_e (see Appendix B, Table B.3). This gives an OPEX of $US 500,687 per year.

To break even NG has to be sold at $US 11.60/GJ. The price of a GJ of NG in Mexico is $US 10.4 on average (including distribution) ("Gas Natural: Precio Gas Natural Usuarios Finales," 2015), this means that a subsidy of $US 1.2/GJ is necessary to break even. This is equivalent to 4.5 c per m_n^3 of biomethane or per litre of diesel equivalent when considered as a transport

biofuel. This would in the EU be seen as a small grant. For example in Ireland the biofuel obligation certificate (BOC) system would award a minimum of 30 c/L of biomethane from slurry if used as a transport biofuel (Ahern et al., 2015)

4.4. Conclusions

Biogas from pig slurry has significant potential in Mexico (21 PJ). The use of HDPE anaerobic covered lagoons can significantly reduce capital costs as compared to European digesters. When elephant grass is used as a co-substrate, a significant increase in methane production is reached. For scenario 1 (mono digestion of pig slurry) a minimum tariff of \$US 0.115/kWh_e and \$US 0.096/kWh_e (with and without grants) respectively is necessary to break even. For scenario 2 (co-digestion of pig slurry and grass) a minimum tariff of \$US 0.129/kWh_e and \$US 0.114/kWh_e (with and without grants respectively) is necessary to break even. There is significant potential for viable commercial operation. Centralised biogas plants (scenario 3) appear less profitable. Biomethane production cannot compare with the price of natural gas; if used as a transport biofuel, a minimum subsidy of 4.5 c/L diesel equivalent (1m^3 of biomethane) would be required.

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5. Anaerobic co-digestion of *Ulva lactuca*, dairy slurry and grass silage for the production of gaseous biofuel in coastal regions

Anaerobic co-digestion of *Ulva lactuca*, dairy slurry and grass silage for the production of gaseous biofuel in coastal regions

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Abstract

Seaweed is a promising feedstock for anaerobic digestion since greater biogas yields per hectare can be achieved as compared to land-based energy crops. *Ulva lactuca* is a green seaweed that accumulates in shallow bays as a result of eutrophication, however it can also provide a potential biomass resource. Fresh cast samples of *U.lactuca,* collected in summer months, were mono-digested and co-digested with grass silage and dairy slurry in batch biomethane potential (BMP) tests and subsequently in a continuous digestion process. Three feedstock mixes containing 70% grass silage, fresh *U.lactuca* at a ratio of 5%, 15% and 25% and dairy slurry at 25%, 15% 5% (based on volatile solids) were tested. From the BMP tests, a mix containing 25% *U.lactuca*, 5% dairy slurry and 70% grass silage gave the highest specific methane yields (SMYs). A bespoke pilot scale continuously stirred tank reactor (CSTR) was designed and built specifically for testing continuous digestion, where, at an organic loading rate (OLR) of 2kgVS/m^3/d, SMYs of 89% the BMP value were achieved. When the OLR was increased to 3kgVS/m^3/d, the SMYs decreased in continuous digestion. Levels of FOS:TAC (ratio of volatile organic acids to total inorganic carbon) and total volatile fatty acids were shown to increase, affecting the process stability.

Keywords: *Ulva lactuca* ; Grass silage; Anaerobic co-digestion; Biomethane

5.1. Introduction

5.1.1. Transition to third generation biofuels

Biofuels account for approximately 4% of the world's road transport fuel and this figure is set to increase to 4.3% by 2020 (IEA, 2015). First generation liquid biofuels such as bioethanol and biodiesel are the main renewable fuels consumed in the transport sector (Sawin et al., 2016). However, controversy has arisen regarding the use of first-generation biofuels, given the limited greenhouse gas emissions savings they can achieve (Aro, 2016). Furthermore, the use of food-based crops for biofuel production can have a negative impact on food security, increasing the price and availability of the crops (Tenenbaum, 2008). Thus, the European Commission has encouraged the transition from first-generation biofuels to biofuels derived from ligno-cellulosic biomass (second generation biofuels) and advanced biofuel sources such as macro- and micro-algae (third generation biofuels) (Boutesteijn et al., 2017). Macro-algae (seaweed) are an ideal feedstock for the production of third generation biofuels due to their high production rate and low lignin content; furthermore, there is no land requirement in its production (Alam et al., 2015). The target in the recast Renewable Energy Directive is for 3.6% of energy in transport to be met with advanced biofuels by 2030 (EC, 2017), indicating the low technology readinesss level in this sector.

The production of gaseous biofuel in the form of biomethane can be preferable to first generation liquid biofuels due to the relatively low energy input required in the anaerobic digestion process and the availability of waste feedstocks (Allen et al., 2014). The use of macro-algae to produce gaseous biofuels has been tested in biomethane potential (BMP) tests and continuous anaerobic digestion laboratory experiments. Seaweed species such as *Laminaria digitata* and *Ascophyllum*

nodosum have recorded specific methane yields (SMYs) of 218 LCH_4/kg of volatile solids (VS) and 166 LCH_4/kg VS respectively (Allen et al., 2015). As a feedstock for digestion, seaweed can sometimes have difficulties in relation to high polyphenol concentrations (brown seaweeds) and low C:N ratios (green algae) (Allen et al., 2015; Tabassum et al., 2016).

5.1.2. Anaerobic digestion of *Ulva lactuca*

Ulva lactuca is a green macro-algae that can be found in several coastal regions and climates. It can be considered a potential energy source due to its high growth rates and high content of carbohydrates (Bruhn et al., 2011). In recent years, eutrophication has led to a proliferation of *Ulva* species accumulating in countries such as Ireland, Japan and France, causing environmental problems in coastal regions with long shallow bays. *U.lactuca* tends to wash to the shores where it is left to decompose (Allen et al., 2013; Briand and Morand, 1997; Bruhn et al., 2011). In Ireland, these so-called "green tides" have occurred primarily in the east and south coasts where eutrophication, due to intensive farming of land and nutrient fertiliser application (N, P and K), is more common (Allen et al., 2013). It has previously been proposed that *U.lactuca* biomass could be used as animal feed or as a fertilizer, however, the content of toxic metals in the *U.lactuca* species may complicate such applications (Charlier et al., 2008). Previous literature has also suggested that anaerobic digestion of *U.lactuca* is preferable to landfilling and composting (Briand and Morand, 1997), whereby with a low lignin content, it can be easily degraded to produce biogas (Allen et al., 2013). Despite recent interest in the use of algae (macro and micro) for the production of biomethane, research on *U.lactuca* as a biogas feedstock is limited. Most of the available literature regarding the digestion of *U.lactuca* consists of batch scale laboratory

experiments. The use of continuous pilot scale reactors for the digestion of *U.lactuca* has not been explored in previous scientific literature.

Table 5.1. Biomethane yields from mono-digestion of *U.lactuca*

Author	Country	Pre-treatment type	Reactor type	SMY $LCH_4/kgVS$
Costa et al	Portugal	Dried and ground	Batch	196
Bruhn et al	Denmark	Fresh and cut (2 cm)	Batch	174
		Fresh and macerated	Batch	271
Briand and Morand	France	Fresh no pre-treatment	Batch	110
Allen et al	Ireland	Fresh and cut (1 cm)	Batch	205
		Dried and ground	Batch	226

SMYs ranging from 110 $LCH_4/kgVS$ to 271 $LCH_4/kgVS$ have been reported in batch studies for *U.lactuca* (Table 5.1). The SMY is often dependent on the type of pre-treatment applied. Higher yields may be achieved by macerating *U.lactuca* prior to digestion (Bruhn et al., 2011). However, digestion of *U.lactuca* can be problematic due to its low C:N ratio. To overcome this, co-digestion with other substrates such as animal slurries has been suggested to increase the C:N ratio (Allen et al., 2014; Peu et al., 2011). Peu et al. analysed the continuous digestion of pig slurry and *U.lactuca* (48%:52% wet weight respectively) in a continuously stirred tank reactor (CSTR), with 3.5 L working volume, at an organic loading rate (OLR) of 1.6 $kgVS/m^3/d$ achieving a SMY of 148 L/kgVS (Peu et al., 2011). Allen et al. co-digested fresh and dried *U.lactuca* with dairy slurry in CSTRs, with 4 L working volume, varying the OLR from 0.5 to 2.5 $kgVS/m^3/d$, testing three different mixes based on the VS content; the optimum mix was 25% fresh *U.lactuca* and 75% dairy slurry (Allen et al., 2014). Co-digestion of dried *U.lactuca* species and sewage sludge in batch reactors has also been analysed achieving a maximum SMY of 296 L/kg VS (15%

U.lactuca:85% sludge based on DS) (Costa et al., 2012). Table 5.2 illustrates the SMYs attained from previous studies that co-digested *U.lactuca.*

Table 5.2. Biomethane yields from co-digestion of *U.lactuca*

Author	Country	*U.lactuca* pre-treatment type and portion of feedstock	Co-substrate and portion of feedstock	Dry Solids content of feedstock %	Volatile Solids content of feedstock %	Reactor type	OLR kgVS/m^3/d	SMY LCH_4/kgVS
Peu et al	France	Fresh macerated 48% wet weight	Pig slurry 52% wet weight	12.2	6	CSTR	1.6	196
Costa et al	Portugal	Fresh macerated 15% DS	Sewage sludge 85% of Dry Solids	4.7	3.1	Batch	--	296
Allen et al	Ireland	Fresh macerated 25%VS	Dairy slurry 75% of Volatile Solids	6.4	4.3	CSTR	2	178

5.1.3. Feedstock availability in Irish coastal regions

The majority of agricultural land in Ireland is under grass with ca. 3.5 million hectares of grassland (Eurostat, 2018). The resource of grass in Ireland is set to grow as Teagasc (the Agriculture and Food Development Authority), aim to increase the production of grass on farms to at least 10t dry solids (DS) per hectare per annum under the Grass10 initiative (Teagasc, 2017). Grass produced in excess of livestock requirements could potentially be used to produce biogas, which can be further upgraded to biomethane, a renewable gas with similar properties to that of natural gas. Previous studies have indicated that an excess of 1.7 million tons of grass DS grass may be available per year (McEniry and O'Kiely, 2013), equating to 155,000 ha of grassland (Wall et al., 2013). The biomethane potential for grass is high at ca. 400 LCH_4/kgVS, however, long term mono-digestion of grass can diminish SMYs due to a lack of essential trace elements

(Thamsiriroj et al., 2012). To overcome this co-digestion with dairy slurry has been proposed (Wall et al., 2014). Dairy slurry is an abundant resource in Ireland with approximately 18,000 dairy farms in Ireland (O'Connor and Kean, 2014). The current management practice for slurry is application to land as a fertiliser without any treatment (Wall et al., 2013). However, slurry from farms could also be used as a co-substrate for anaerobic digestion with return of digestate to land as a biofertiliser. The biomethane potential of dairy slurry ranges from 136 to 239 LCH_4/kgVS (Allen et al., 2014; Wall et al., 2013). When co-digested with grass at 80% grass:20% slurry (based on VS), yields of 345 LCH_4/kgVS can be achieved (Wall et al., 2013).

In coastal regions where eutrophication is evident, *U. lactuca* can be an added biomass stream for digestion that leads to reduced pollution and improved waste management. To illustrate the concept, the village of Timoleague in West Cork was taken as a model for this study. Timoleague is a coastal town located in one of the most intensive dairy farming areas of Ireland with approximately 25,000 dairy cows in the surrounding region (O'Connor and Kean, 2014). Each year large quantities of *U.lactuca* (in the order of 10,000 wet tons) are stranded in the estuaries of Timoleague, having an adverse impact in the surrounding environment as previously detailed by Allen et al. (2013).

5.1.4. Reactor design for the co-digestion of *Ulva lactuca*, grass silage and dairy slurry

Reactor configuration and design is typically dependent on the feedstock employed. CSTRs are commonly used for digesting feedstocks with a DS content of 2- 12% such as animal slurries, sewage sludge and energy crops. A crucial element in this type of reactor is the agitation systems that can employ vertical or horizontal paddle stirrers or biogas recirculation (Murphy and

Thamsiriroj, 2013). Different species of macro-algae have been digested in lab scale CSTRs obtaining good results in terms of SMYs and reactor operation (Allen et al., 2014; Peu et al., 2011; Tabassum et al., 2016). Grass silage is commonly digested in CSTRs however the high solid and fibrous content in grass silage can be problematic for mixing systems as it tends to accumulate between interlocking parts. To ensure a homogenous mix, systems of greater capacity are needed, increasing the amount of energy required (Thamsiriroj and Murphy, 2010). Thamsiriroj and Murphy (2010) operated a 600 L two stage pilot scale CSTR for 101 days using only grass silage as substrate. The study found that the highly fibrous material and high solids content of grass made it difficult to mix. During the commissioning period grass floated, forming an indigestible layer that was not possible to break using horizontal mixing paddles. Pumping is also an important part of the system. For high solids substrates such as food waste and the organic fraction of municipal solid waste (OFMSW), heavy duty solid pumps are commonly used at an industrial scale (Joshua. Rapport et al., 2008). For lab scale pilot reactors, peristaltic pumps are used given that they can supply slower flows as compared to industrial pumps (EPA, 2008). A good reactor design for high solid content substrates must take into account all the above to facilitate efficient operation of the system.

5.1.5. Aims and objectives

The aim of this experiment is to assess the feasibility of co-digesting *U.lactuca* with grass silage and dairy slurry in a pilot scale CSTR under mesophilic conditions. To the knowledge of the authors, no previous literature studies have analysed the continuous co-digestion of *U.lactuca*, grass silage and dairy slurry in a pilot scale reactor. The specific objectives are to:

- Analyse the optimal percentage of *U.lactuca* that can be digested with grass silage and dairy slurry.
- Design, build, commission and operate a bespoke pilot scale reactor to digest the selected feedstocks.
- Assess continuous digestion with increasing OLR of the optimal feedstock mix in the bespoke pilot scale reactor.
- Assess the biomethane potential of a coastal rural biogas plant in Ireland

5.2. Materials and methods

5.2.1. Materials and inoculum

Samples of cast *U.lactuca* were collected in Timoleague Cork in June 2013, August 2016 and September 2017. The seaweed was manually cleaned from impurities such as stones and other non-biodegradable materials and macerated to a particle size of approximately 5 mm using a heavy duty mincer. Samples were then frozen and stored at minus 20°C. The DS of the samples varied from 155 g/kg to 179 g/kg (average 164.3 g/kg with a standard deviation of 10.5 g/kg). The VS content was on average 82.3 g/kg and the C:N ratio ranged between 7.2 and 11.2.

Dairy slurry was collected from two different dairy farms in County Cork, Ireland in April 2016 and May 2017. The slurry (faeces and urine) produced on the farms was collected through slatted floors when the livestock were indoors. Once collected, slurry samples were frozen by storage at -20°C. The DS of the slurry varied from 58.4 g/kg to 78.8 g/kg (average of 68.6 g/kg with a standard deviation of 9.8g/kg). The VS content was on average 48.6 g/kg and the C:N ratio ranged

from 15.8 to 18.6.

The grass silage used was an early cut of perennial ryegrass (*Lolium perenne*). The DS and VS of the grass silage was 349 g/kg and 316 g/kg respectively. The C:N ratio was recorded at 29.8. Grass silage was macerated to a 5 mm particle size using a heavy duty mincer and stored at -20°C prior to use. Samples were fully defrosted before being fed to the digester.

The inoculum for both batch and continuous trials was taken from a pilot scale reactor fed with dairy slurry. Before its use, the inoculum was sieved through a 1 mm sieve and degassed for two weeks at 38°C.

5.2.2. Analytical methods

DS and VS were analysed and calculated using standard methods (APHA, 2011). The pH was measured weekly using a Jenway 3510 pH metre. A Titronic Universal automatic titrator was used to determine FOS:TAC on a weekly basis. FOS:TAC is the ratio of volatile organic acids (FOS) to total inorganic carbon (TAC); it was measured according to the two-point titration method described by Drosg using a 0.1 N sulphuric acid solution with pH 5 and pH 4.4 as endpoints (Drosg, 2013). Values below 0.3 indicate a stable process (Drosg, 2013). Chloride concentrations and total ammoniacal nitrogen (TAN) were determined weekly using a Hach Lange DR3900 benchtop spectrophotometer and CLK 303 and LCK 311 cuvettes, respectively. Free ammonia was calculated using equation 1 which uses pH and temperature (Hansen et al., 1998).

$$NH_3 = (NH_4 - N) * \left(1 + \frac{10^{-pH}}{10^{-(0.09018 + \frac{2729.92}{T(K)})}}\right) \qquad \text{(Eq. 9)}$$

NH_3 represents the concentration of free ammonia in mg/L, NH_4-N is the total ammonium concentration and T (K) is the temperature in Kelvin.

The content of carbon (C) and nitrogen (N) was determined using an elemental analyser (CE 440 Elemental Analyser). Samples were dried at 105°C and then ground and sieved using a 6µm sieve before undertaking this analysis. Biogas composition was determined using an Agilent 6890 GC equipped with a Hayesep R packed column and a thermal conductivity detector. Samples were analysed on a weekly basis. Volatile fatty acids (VFA) were also determined with an Agilent 6890 GC, equipped with a Nukol fused silica capillary column (30 m x 0.25 mm x 0.25 µm), argon as a carrier gas and a flame ionisation detector. Samples were analysed every other week.

5.2.3. Biomethane potential test

Three different feedstock mixes were tested on a VS basis:

- 5% *U.lactuca*, 25% dairy slurry and 70% grass silage;
- 15% *U.lactuca*, 15% dairy slurry and 70% grass silage; and
- 25% *U.lactuca*, 5% dairy slurry and 70% grass silage.

Bioprocess Control automatic methane potential test systems (AMPTS II) were used to undertake the BMP tests. The AMPTS consisted of 15 glass bottles of 650 ml total volume that were continuously stirred. A heated water bath kept a constant temperature in the bottles of 38°C (mesophilic conditions). The bottles were initially flushed with nitrogen to create anaerobic conditions. Each mix was assessed in triplicate with a VS ratio of inoculum to substrate of 2:1. The total test period was 30 days. The biogas generated in digestion passed through a 3 M sodium hydroxide solution that removed carbon dioxide, resulting in only the methane volume being

recorded via water displacement. All data was recorded and downloaded once the test was complete.

5.2.4. Pilot scale reactor design and set up

A 50-litre pilot scale CSTR was designed and built specifically for continuous digestion.

5.2.4.1. Control system

The CSTR was equipped with pH probes that continuously monitored pH readings. Gas flow meters were installed at the top of the tanks and connected to a programmable logic controller (PLC). Temperature was measured using two thermocouples.

5.2.4.2. Mixing system

The CSTR consisted of two stainless steel tanks that were agitated using vertical paddles specifically designed to break any layers of floating grass which formed undesirable scum layers on the liquid surface (Figure 5.1). The mixer was installed in the middle of the tank and comprised of an AC electric motor attached to a gearbox. The speed of the stirrer was controlled by a variable frequency drive which could be adjusted by the user. The motor shaft was submerged in the reactor's content, avoiding any overpressure that could damage the seal creating any subsequent leaks.



Figure 5.1. Vertical paddles in reactor

5.2.4.3. Heating system

The tank was positioned on top of a water vessel which was heated by electric heating elements. The heating elements were controlled by the PLC via a thermocouple that measured the internal temperature of the reactor to meet the target temperature. The temperature could be set manually by the user.

5.2.4.4. Feeding system

A reception hopper installed at the top of the reactor received the substrate (Figure 5.2). The hopper was attached to an electric grinder. The suction of a peristaltic pump was connected to the grinder and the discharge to the CSTR. The speed of the pump was controlled by the PLC, allowing for a slow or fast feeding. An extra feeding port with a valve was placed at the top of the tank. In order to feed through the port a plastic plunger was necessary to push down the feedstock to the bottom of the tank.



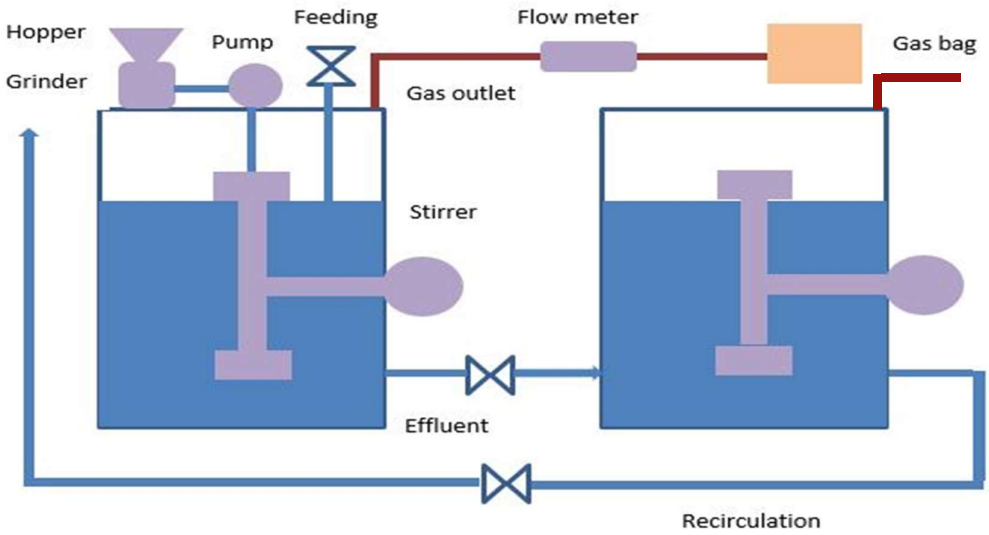


Figure 5.2. Photo and schematic of continuous pilot scale reactor

2.5. Experimental setup and reactor operation

The pilot reactor was continuously fed with the mix that reached the highest SMY as indicated in the BMP tests. An initial OLR of 2 kgVS/m^3/d was chosen as suggested by Allen et al. (Allen et al., 2014). The mix was run for two hydraulic retention times (HRTs). An increase in OLR to 3

kgVS/m^3/d was tested for a further two HRTs. The SMYs indicated from the BMP results for the mix was used as a target for the continuous digestion trials. The reactor was fed daily. To prevent stirrer malfunction, digestate from the second tank was sieved and recirculated to keep the solids content at 10%.

5.3. Results and discussion

5.3.1. Biomethane potential tests

The SMYs produced from the BMP tests are shown in Figure 5.3. Cellulose yielded 350 LCH_4/kgVS indicating the viability of the inoculum. Grass had the highest methane yield with 387 LCH_4/kgVS which is similar to the yields reported in the literature (Nizami and Murphy, 2011; Wall et al., 2013). The C:N ratio of grass silage was 29.89 which falls within the optimum range for anaerobic digestion (Allen et al., 2015). *U.lactuca* reached 191.5 LCH_4/kgVS, which is comparable with the studies shown in Table 5.1. Slurry gave a methane yield of 115 LCH_4/kgVS. These results are similar to the yields reported in the literature for dairy slurry (Allen et al., 2014; Wall et al., 2013). The mix containing 25% *U.lactuca*, 5% dairy slurry and 70% grass silage gave the highest numerical yield amongst the different mixes tested, producing a SMY of 325 LCH_4/kgVS. From the BMP results it was evident that as the content of *U.lactuca* in the mix increased, the numerical value of the SMY also increased, but with very little difference. The three design mixes generated between 313 (5% *Ulva*) and 325 LCH_4/kgVS (15% *Ulva*). A one-way ANOVA analysis was run to confirm that the SMY of the three mixes were statistically different at the p-value of <0.05, [$F(2,6)=6.80$, $p=0.028$]. There was a significant variation on methane yields as the amount of *Ulva lactuca* was increased from 5% to 25%. When comparing

the results with the predicted pro-rata yields, the mixes produced up to 6% more methane than expected (Table 5.3). This could be due to a synergistic effect as a result of combining the three substrates, typically associated with a more optimal C:N ratio as would be achieved by combining *U.lactuca* with grass and slurry (Allen et al., 2014; Wall et al., 2013). Again however, there was very little difference. Work by Allen et al. (2014) suggested that long term digestion is not feasible with more than 25% *U.lactuca* and suggested that this was due to salt accumulation, levels of sulphur in the U.lactuca and the level of nitrogen associated with the seaweeed.

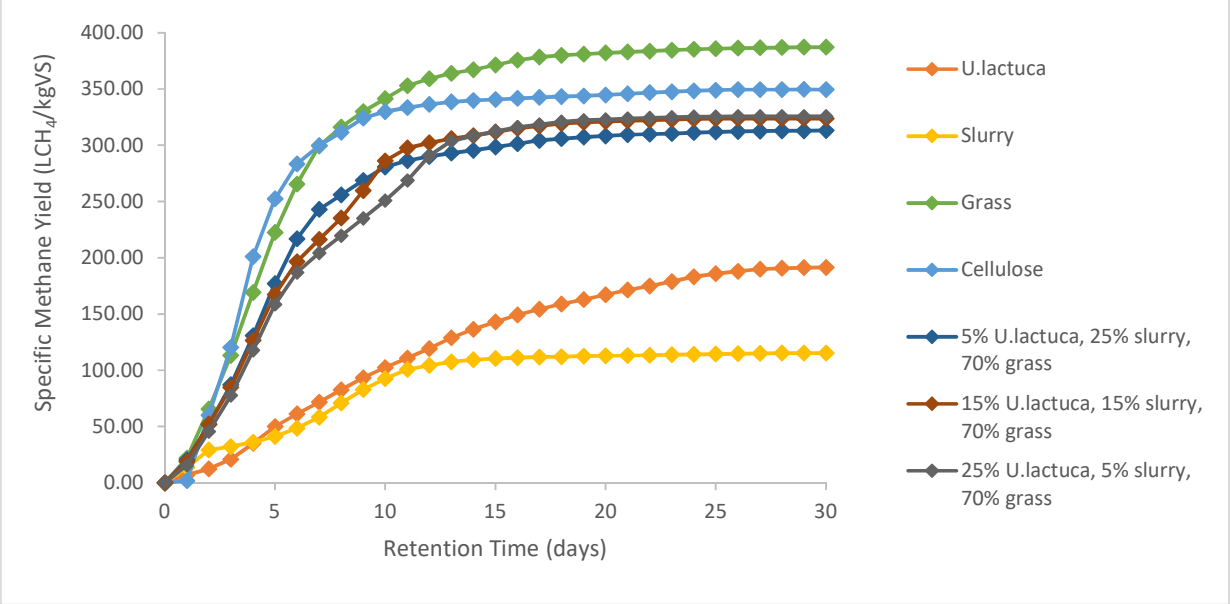


Figure 5.3. BMP results for feedstock mixes

5.3.2. Commissioning phase of the pilot scale digester

The bespoke pilot scale digester was designed by the research team and fabricated locally (see figure 5.2). On completion the digestion tanks were filled with inoculum, heated to 38°C and

mixed. Subsequently, in a commissioning period, the reactor was fed with dairy slurry at an OLR of 1 kgVS/m^3/d. During this period, several problems were experienced including:

- Blockages and leaks in the feeding system
- Inconsistencies in biogas flow and pH readings.
- Problems in regulating the set temperature.

5.3.2.1. Feeding system.

To avoid further leaks, silicone hoses and clamps were changed and readjusted. The pump case was cleaned and all the pieces were tightly re-installed. In order to reduce blockages, the grinder was thoroughly cleaned and the blades were tightly screwed. Feeding was resumed after these changes; however, when grass silage was added to the mix blockages occurred again. The grinder's electric brake was constantly tripping due to current overloads caused by the clogged grass. The same problem was experienced by the pump having to reset and clean the pump each time the digester was fed. With the issues described above, a change in the method of reactor feeding was undertaken, using the feeding port provided for this purpose.

5.3.2.2. Control system

From the first week of the commissioning period the biogas flow readings were inconsistent. The device did not register flow even when there was a constant biogas production. The communication between the meter device and the PLC was checked and no issues were found, narrowing the problem down to a faulty flow meter. To overcome this, a wet gas tipping bucket working under the water displacement principle was used. This type of flow meters is widely used in laboratory experiments (Allen et al., 2014; Tabassum et al., 2016; Wall et al., 2013), given its

simplicity and reliability in reading biogas flows. The pH probes readings were unstable and below 5. To determine the accuracy of the instrument, a sample from the reactor was taken and analysed with a Jenway pH meter, finding the pH values between 7 to 7.5. The probes were not re-calibrated as this parameter could be easily read with and external pH meter. From this point, samples from the digester were taken weekly to measure pH.

5.3.2.3 Heating system

The set temperature (38°C) fluctuated during the first weeks of the commissioning period, reaching temperatures of up to 50°C. To tackle this issue, a complete inspection of the heating system was carried out, finding the problem on the contacts of a relay. The relay was changed solving the malfunction of the system.

5.3.2.4 Mixing system

The vertical paddles of the mixing system functioned well, breaking any accumulation in the surface layer and preventing grass from floating. This could be examined through an inspection glass placed at the top of the reactor.

5.3.3. Continuous digestion of *U.lactuca*, grass silage and slurry

A mix containing 25% *U.lactuca*, 5% dairy slurry and 70% grass silage was chosen to be further analysed in a continuous digestion system as it recorded the highest SMYs among the three mixes tested in the BMP (Table 5.3). In the continuous pilot scale digester, SMYs reached 89% of the BMP value with an average of 288 LCH_4/kgVS. This efficiency is similar to the that reported by

Allen et al. for a lab scale CSTR digesting *U.lactuca* and dairy slurry and lower as compared to the efficiencies reported by Wall et al. for a lab scale CSTR digesting grass silage and dairy slurry (Allen et al., 2014; Wall et al., 2013). The SMY was relatively constant throughout the experiment (Figure 5.4) and an average methane concentration of 55.3% was recorded in the biogas (standard deviation of 1.2%). FOS:TAC levels were low, between 0.16 and 0.25, allowing for stable digestion (Figure 5.4). The maximum total VFA concentration recorded was 399.5 mg/L.

After two HRTs the OLR was increased to 3 kgVS/m^3/d for the same feedstock mix (25% *U.lactuca*, 5% dairy slurry and 70% grass silage). The SMYs reduced steeply from 253 LCH_4/kgVS (week 1) to 143 LCH_4/kgVS on the final week, equating to 44% of the BMP value (Figure 5.4). FOS:TAC and total VFA levels rose to 0.41 and 3,146 mg/L respectively. The pH of the reactor remained between 7.32 to 7.66. Methane concentrations in the biogas decreased from 53.4% to 47.88%. The results presented suggest that the reactor performed optimally at a lower OLR of 2kgVS/m^3/d.

Table 5.3. Specific methane yields assessment

Feedstock	BMP result LCH_4/kgVS	Predicted yield LCH_4/kgVS[a]	Continuous SMY LCH_4/kgVS	Biodegradability index[b]
Grass	387	-	-	-
Slurry	115	-	-	-
U.lactuca	191	-	-	-
5% *U.lactuca*, 25% slurry 70% grass	313	309	-	-
15% *U.lactuca*, 15% slurry 70% grass	324	317	-	-
25% *U.lactuca*, 5% slurry 70% grass	325	325	288	0.89

[a] Calculated in proportion to the BMP results on a pro-rata basis
[b] SMY in continuous digestion / BMP results

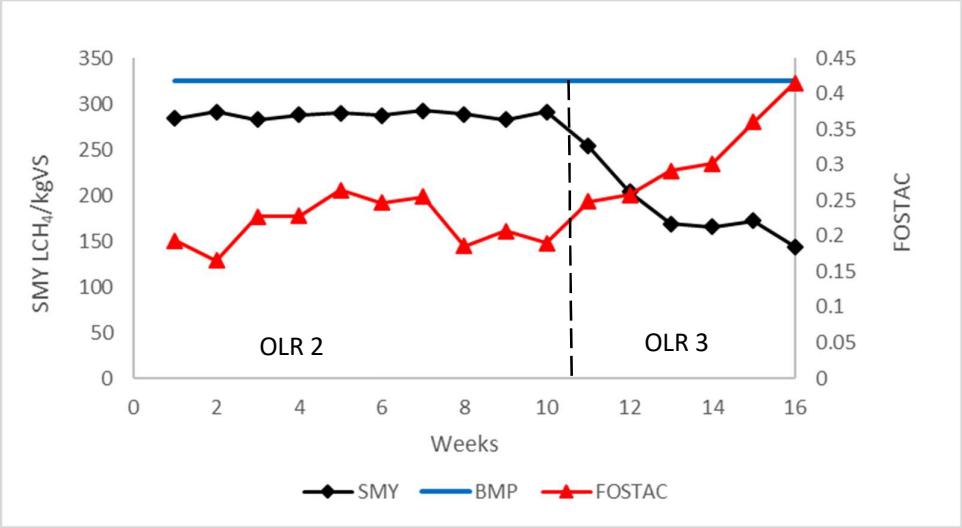


Figure 5.4. Specific methane yields (SMY) and FOSTAC from pilot scale reactor

5.3.4. Operational parameters

5.3.4.1. TAN

High ammonia concentration has been identified as an inhibitor of anaerobic digestion. Concentrations greater than 5,000 mg/L are deemed to hinder the anaerobic process (Murphy and Thamsiriroj, 2013). Inhibitory levels of free ammonia (NH_3) depend on the substrate and operating conditions applied (Moestedt et al., 2016). Concentrations of 337 mg/L to 1,000 mg/L have been described as detrimental to the AD process (Moestedt et al., 2016; Yenigün and Demirel, 2013). A low C:N ratio in the feedstock can reduce organic conversion, leading to a build up of ammonium in the reactor (Allen et al., 2014; Lehtomäki et al., 2007). For an OLR of 2kgVS/m^3/d TAN levels reached a maximum concentration of 2,106 mg/L but decreased by the end of the second HRT to 1,336 mg/L. pH levels of 7.5 to 7.7 were recorded. At an OLR of 3, levels of TAN remained steady with an average concentration of 2,002 mg/L and a maximum concentration of 2,189 mg/L, similar to that of the lower OLR.

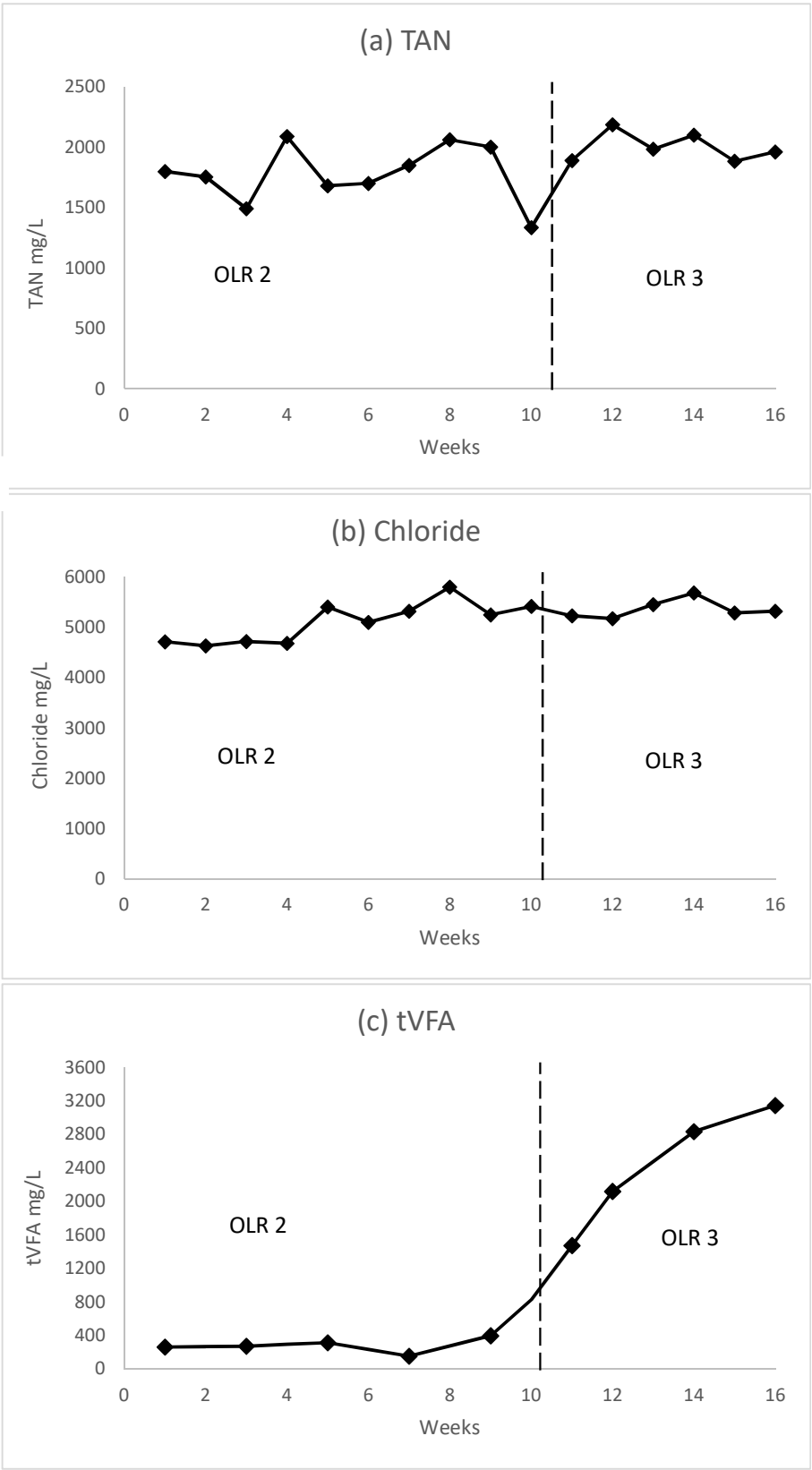


Figure 5.5 (a) total ammoniacal nitrogen content (TAN), (b) chloride content, (c) total volatile fatty acid content (tVFA).

Levels of TAN remained throughout below 2,500 mg/L (Figure 5.5 (a)). These values are similar to the ones reported by Allen et al of 2,168 mg/L for a CSTR digesting *U.lactuca* and dairy slurry at a VS ratio of 25%:75% respectively (Allen et al., 2014). These low TAN levels could be explained by the optimum C:N ratio of the mixes and the relatively low OLRs employed. Levels of free ammonia remained below the recommended safety thresholds at 305 mg/L. From the results obtained it was concluded that TAN concentrations did not affect methane production.

5.3.4.2. Chloride

High levels of chloride can potentially impede anaerobic digestion by increasing the osmotic pressure and causing dehydration of bacterial cells (Herrmann et al., 2016; Ward et al., 2014). Values of chloride in excess of 9,600 mg/L have been identified as an inhibitor for the digestion process (Herrmann et al., 2016). Despite this, Tabassum et al., recorded concentrations of up to 14,000 mg/L in an acclimitised process over a significant period of time without affecting methane production (Tabassum et al., 2016). At an OLR of 2kgVS/m^3/d chloride levels remained stable reaching a maximum concentration of 5,419 mg/L on week 8 (figure 5.5 (b)). At an OLR of 3kgVS/m^3/d, chloride levels remained stable for the two HRTs reaching a maximum concentration of 5,689 mg/L, slightly higher than the values reported for an OLR of 2kgVS/m^3/d. In this trial, a minor increase in chloride concentrations was evident over the lifetime of the experiment (Figure 5.5 (b)). This was expected as the quantity of *U.lactuca* increased with the increasing OLR. The results presented are in accordance with the ones reported by Allen et al. and Tabassum et al. who recorded an increase in chloride as the content of seaweed and OLR increased (Allen et al., 2014; Tabassum et al., 2016) In this study levels of chloride remained

below 5,900 mg/L, and no direct correlation between chloride concentrations and methane yields was illustrated.

5.3.4.3. Volatile fatty acids and FOS:TAC

An accumulation of VFAs can directly affect methanogenic archaea, reducing methane production (Herrmann et al., 2016). A concentration of 1,000 mg/L has been identified as the maximum upper level in which a stable process is achievable, however, in some reactors concentrations of up to 4,000 mg/L have been reached without affecting the process (Drosg, 2013). This wide range depends heavily on the feedstock employed. Energy crops such as maize and grass silage with high DS tend to have lower levels of VFAs as compared to wastes where the substrate digestion occurs rapidly (Drosg, 2013). Total VFA concentrations for reactors digesting grass silage and dairy slurry at a VS ratio of 80%:20% respectively, have been shown to operate well below the recommended limits even at high OLRs of 4 kgVS/m^3/d (Wall et al., 2014). Higher VFA concentrations, in the range of 1,720 mg/L to 1,955 mg/L, were reported by Allen et al. for a reactor digesting dairy slurry and *U.lactuca* at a VS ratio of 75%:25% respectively (Allen et al., 2014). In this study, total VFA production at an OLR of 2 kgVS/m^3/d remained below 400 mg/L (Figure 5.5 (c)), behaving similarly to the reactor described by Wall et al. (2013). Acetic and propionic acid were the major contributors to the total VFA production during continuous digestion. Some smaller quantities of iso-butyric acid were also found. With the increase in OLR to 3 kgVS/m^3/d the total VFAs started to rise, reaching a maximum value of 3,146 mg/L in the last week of the experiment. This value is higher than the suggested upper limit. As the VFAs rose the SMY slowly decreased, however, the reactor continued producing methane. A similar trend was found by Allen et al. when increasing the OLR from 2 to 2.5 kgVS/m^3/d, methane production

started declining nevertheless. However VFA concentrations in that study were stable, below 2,000 mg/L, and thus the reduction in methane production was linked to a deficiency in trace elements (Allen et al., 2014).

FOS:TAC levels were low and steady at an OLR of 2 kgVS/m^3/d, remaining below the 0.3 threshold, which has been identified as the safety limit. An excess of VFA production and its further accumulation may lead to a higher FOS:TAC value, which indicates process instability. When the OLR was increased the FOS:TAC level increased to 0.41; this was in the last week of the experiment. FOS:TAC and VFA levels showed that the process was somewhat impaired by the increase in OLR having a negative impact on methane generation.

5.3.5. Biomethane resource case study

Timoleague is a coastal town located in west Cork, Ireland, where ca. 10,000 wet tonnes of *U.lactuca* is produced annually through eutrophication. From the results of this analysis, a biogas plant of approximately 23,000 t/year is required to co-digest the *U.lactuca* generated in the area with grass silage and dairy slurry, utilising the optimal mix as identified in this study. Algae can be stored in silos to provide a year-round supply. This digester would have a methane production of approximately 1.19 Mm^3/year as calculated in Table 5.4. This equates to a resource of 42.7 TJ/year, taking the energy in biomethane at 35.9 MJ/m^3. Such a resource could replace the direct fuel consumption of up to 897 houses (assuming the energy in fuel consumed per dwelling at 47.6 GJ/year) or fuel the equivalent to 1,587 diesel cars (average diesel car consumption 750 L/year; energy content in diesel 35.9 MJ/L) (Allen et al., 2013, SEAI, 2017).

Table 5.4. Energy production of a biogas plant in Timoleague

Mix	Feedstock	DS/year	VS/year	Wet weight/year	SMY[d]	CH_4m^3/year	Energy TJ/year[e]
25% *U.lactuca*, 5% slurry, 70% grass	Grass[a]	3,247	2,940	9,304	288	1,190,592.00	42.7
	Dairy slurry[b]	197.6	144	3,349			
	U.lactuca[c]	1,790	1,050	10,000			
Total				22,653			

[a] DS 34.9%, VS 31.6%
[b] DS 5.9%, VS 4.3%
[c] DS 17.9%, VS 10.5%
[d] Mix C Continuous SMY 288 LCH_4/kgVS
[e] 35.9 MJ/m^3

5.4. Conclusions

The optimal feedstock mix for grass silage, *U.lactuca*, and dairy slurry for digestion, as identified through this analysis, is 70%, 25% and 5%, respectively, on a volatile solids basis. This feedstock mix reached 89% of the SMY evaluated through a BMP test in a continuous pilot scale trial operating at an OLR of 2 $kgVS/m^3/d$. When the OLR was increased to 3 $kgVS/m^3/d$, levels of FOS:TAC and total VFAs began to increase having an adverse effect on the methane yields attainable. An OLR of 2 $kgVS/m^3/d$ is proposed as optimal. A different feeding configuration is needed for this specific feedstock since frequent blockages occurred. Manually feeding through a port proved to be a reliable solution, however, in an industrial scale this may not be feasible. Vertical paddles helped mixing the reactor's content avoiding the formation of scum layers. The biomethane resource identified in Timoleague, where the *U.lactuca* was sourced was significant, having the potential of producing 42.7 TJ per year.

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6. Assessment of continuous fermentative hydrogen and methane co-production using macro- and micro-algae with increasing organic loading rate

Assessment of continuous fermentative hydrogen and methane co-production using macro- and micro-algae with increasing organic loading rate

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Abstract

A two-stage continuous fermentative hydrogen and methane co-production using macro-algae (*Laminaria digitata*) and micro-algae (*Arthrospira platensis*) at a C/N ratio of 20 was established. The hydraulic retention time (HRT) of first-stage H_2 reactor was 4 days. The highest specific hydrogen yield of 55.3 L/kg volatile solids (VS) was obtained at an organic loading rate (OLR) of 6 kgVS/m^3/d. In the second-stage CH_4 reactor at a short HRT of 12 days, a specific methane yield of 245 L/kgVS was achieved at a corresponding OLR of 2 kgVS/m^3/d. At these loading rates, the two-stage continuous system offered process stability and effected an energy yield of 9.4 kJ/gVS, equivalent to 77.7% of that in an idealised batch system. However, further increases in OLR led to reduced hydrogen and methane yields in both reactors. The process was compared to a one-stage anaerobic co-digestion of algal mixtures at an HRT of 16 days. A remarkably high saline level of 13.3 g/L was recorded and volatile fatty acid accumulation were encountered in the one-stage CH_4 reactor. The two-stage system offered better performances in both energy return and process stability. The gross energy potential of the advanced gaseous biofuels from this algal mixture may reach 213 GJ/ha/yr.

Keywords: Macro-algae; micro-algae; two-stage co-fermentation; hydrogen; methane

6.1. Introduction

In recent years there is an increased interest in producing advanced biofuels from alternative feedstocks. The need to improve energy yields and allay sustainability concerns including land use change of first and second generation biofuels have led to research of algae (both macro and micro) as viable substrates for the production of advanced biofuels. Algal biofuels can overcome the food-or-fuel debate associated with first generation biofuels (Maity et al., 2014a; Murphy JD, 2011) and do not face the complex conversion processes required for second generation biofuel production (Bhutto et al., 2017; Zheng et al., 2014). Aquatic algae possess several advantages over terrestrial plants. Firstly, both macro-algae and micro-algae have higher growth rates and biomass productivities as compared to agricultural crops (Dismukes et al., 2008; Ghosh et al., 2016; Tabassum et al., 2017). Secondly, the cultivation of algae may not require arable lands or fresh water. A win-win situation can be achieved through coupling algae production with wastewater treatment (Gurung et al., 2012; Maity et al., 2014b; Wall et al., 2017). Thirdly, algae may provide continuous biomass supply throughout the year with optimised cultivation such as CO_2 supplementation using flue gas for micro-algae (Jacob et al., 2015; Zhao et al., 2015) and efficient preservation such as ensiling for macro-algae (Herrmann et al., 2015).

Production of liquid biofuels (such as biodiesel and bioethanol) using algae biomass has been extensively explored (Sirajunnisa and Surendhiran, 2016; Williams and Laurens, 2010). However, the parasitic energy demand for the generation of liquid biofuels from raw feedstocks exceeds that in the conversion from substrates to gaseous biofuels such as biohydrogen and biomethane (Allen et al., 2015; Power and Murphy, 2009; Stephenson et al., 2010), leading to comparatively lower overall energy efficiencies. Besides, gaseous biofuels offer more utilisation options, including:

compression for vehicles fuels; injection into the existing natural gas grids for use as renewable heat in industry such as breweries(Huang et al., 2017); on site electricity generation using internal combustion engines (Sun et al., 2015); or increased efficiency through use of biomethane from the gas grid at combined cycle gas turbines.

Biological hydrogen production through dark hydrogen fermentation of algae biomass shows advantages over conventional energy-intensive hydrogen-producing methods such as steam methane reforming (Dou et al., 2017) due to mild reaction conditions and renewability of the produced hydrogen (Xia et al., 2016a). However, limited energy conversion restricts its application. An alternative gaseous product biomethane generated through biological anaerobic digestion of algae biomass with better energy output has been analysed in previous studies (Dębowski et al., 2013; Sirajunnisa and Surendhiran, 2016; Ward et al., 2014). Nevertheless, some major bottlenecks still restrict the application of this process. The abundant recalcitrant organics such as polyphenols in macro-algae (Tabassum et al., 2017) and triglycerides in micro-algae are not readily digested by the microbes and thereby decrease the biodegradability of biomass (Ward et al., 2014). In addition, the rigid cell wall structures of algae act as barriers between the intracellular biodegradable contents and anaerobic microbes, hence hindering the degradation and methanogenesis of algae biomass in anaerobic digestion process (Dębowski et al., 2013). To tackle this problem, a two-stage process combining hydrogen fermentation and anaerobic digestion can serve as a promising solution. The two-stage set-up separates the process phases and optimises the operational conditions for each. In the first stage of hydrogen fermentation, the anaerobic fermentative bacteria (AFB) favour the pH condition of 5-6 where they can efficiently degrade the large-molecular-weight organics such as carbohydrates and proteins into gaseous hydrogen, carbon dioxide, and liquid soluble metabolic products (such as volatile fatty acids

(VFAs), alcohols, and lactic acid) in a short retention time (2-4 days) (Xia et al., 2016a). Subsequently, the liquid fermentation effluents rich in small-molecular-weight VFAs and alcohols can be readily utilised by the methanogenic organisms in the second stage of anaerobic digestion. Therefore, compared with one-stage anaerobic digestion, the two-stage process presents better energy yields with improved biogas production and significantly shortens the overall retention time with concurrent increase in organic loading rates (OLRs). Yang et al.(Yang, Z.; Guo, R.; Xu, X.; Fan, X.; Luo, 2011) used lipid-extracted residues of microalgae *Scenedesmus* for two-stage batch fermentative hydrogen and methane co-production and obtained a 22% increase in methane yield and a 27% increase in energy efficiency in contrast to that in one-stage anaerobic digestion. Massanet-Nicolau et al. (Massanet-Nicolau et al., 2015) investigated the two-stage continuous fermentative hydrogen and methane co-production of pelletized grass, which exhibited an overall energy yield of 11.74 kJ/g volatile solids (VS) with an increase of 13.4% compared with one-stage anaerobic digestion. Process stability was maintained whilst the hydraulic retention time (HRT) was greatly shortened from 20 days in the one-stage to 12 days in the two-stage process (Massanet-Nicolau et al., 2015).

Apart from relatively limited biodegradability of algae compared with some first generation feedstocks (Tabassum et al., 2017), the intrinsic compositional unbalance of certain algae biomass (in particular micro-algae biomass) can impair the anaerobic digestion process (Herrmann et al., 2016). Proteins occupy a large portion of organics in micro-algae, leading to a low C/N ratio in the biomass. The excessive nitrogen is released in the form of ammonia during the degradation of proteins, resulting in severe decrease in the microbial activities of methanogenic microbes (Chen et al., 2008) . By contrast, some species of macro-algae, such as brown seaweeds *Laminaria digitata* and *Saccharina latissima*, contain rich carbohydrates and have a high C/N ratio when

harvested at optimum times (Tabassum et al., 2017). This can in certain cases lead to limited nitrogen supply for the basic metabolisms of AFB in hydrogen fermentation and the methanogens in anaerobic digestion (Xia et al., 2016b). The optimum C/N ratio was suggested to be 20-30 for algal feedstocks (Dou et al., 2017; Montingelli et al., 2015). Thus, adjusting the C/N ratio by mixing nitrogen-rich micro-algae and carbon-rich macro-algae as co-substrates offers an excellent strategy to improve the process performances of both hydrogen fermentation and anaerobic digestion. Xia et al. (Xia et al., 2016b) mixed micro-algae *Arthrospira platensis* and macro-algae *L. digitata* for batch fermentative hydrogen production and achieved an optimal H_2 yield of 85 L/kgVS at a C/N ratio of 26.2. A study on the continuous one-stage anaerobic digestion of mixed *S. platensis* and *L. digitata* at a C/N ratio of 25 was conducted and the highest specific methane yield (SMY) of 273.9 L/kgVS was recorded at an OLR of 3 kgVS/m^3/d and an HRT of 28 days (Herrmann et al., 2016).

The authors previously conducted a two-stage batch fermentative hydrogen and methane co-production using co-substrates of macro-algae (*L. digitata*) and micro-algae (*Chlorella pyrenoidosa* and *Nannochloropsis oceanica*) (Ding et al., 2016b). The micro-algae biomass supplied nitrogen to balance the C/N ratio of the algal mixtures. Co-fermentation facilitated the hydrolysis and acidogenesis of the algal co-substrates and further boosted the energy conversion in anaerobic digestion. Although the batch co-fermentation provided some innovative findings, these experimental configurations have significant limitations. Batch systems allow sufficient guaranteed retention times, efficient mixing and anaerobic conditions; they also allow an optimum inoculum to substrate VS ratio of 2:1 which minimises inhibitory effects such as accumulation of volatile fatty acids and ammonia. Batch assays have limited replicability compared to likely industrial applications. In the majority of commercial industrial applications, the loading of the

reactor is continuous. As such it is necessary to undertake continuous laboratory experiments to assess the impact of higher OLRs and shorter HRTs for a prosperous and stable fermentation process. Economics dictate the need for high processing capability and biofuel outputs for minimum size of reactor system. Therefore, continuous two-stage laboratory co-fermentation is essential to address long term optimised operational conditions. Nevertheless, to date, long term continuous two-stage co-fermentation of micro- and macro-algae biomass remains uninvestigated in literature. This paper will address this knowledge gap in the state of the art through the following objectives:

- Assess co-generation of hydrogen and methane using the mixture of macro-algae (*L. digitata*) and micro-algae (*S. platensis*) at the optimal C/N ratio of 20 with increasing OLRs.
- Evaluate the effects of different OLRs and HRTs on the specific hydrogen yields (SHYs), the acidification yields in first-stage dark hydrogen fermentation and the SMYs in second-stage anaerobic digestion.
- Compare the performances of two-stage and one-stage systems on the overall energy conversion and process stability.
- Estimate the gross energy potential of this advanced gaseous biofuel system.

6.2. Materials and methods

6.2.1. Algal biomass and inoculum

The macro-algae *L. digitata* was naturally grown in the open sea and collected in September in West Cork, Ireland. The harvested *L. digitata* was washed with tap water to remove attached sands and other impurities, and then cut to small particles (4-5 mm) by a mincer (Buffalo Heavy Duty Mincer CD400). The micro-algae powder of *S. platensis* was purchased from Bluegreen Life Foundation Inc. (Lewes, DE, USA). Both macro- and micro-algal samples were cryopreserved at -20 °C before the experiment.

The hydrogen inoculum used in biohydrogen potential (BHP) test and continuous hydrogen reactor was taken from the anaerobic sludge of an Irish farm digester. The original sludge was heated at 100 °C in an autoclave (Sanyo MLS-3780, Japan) for 30 min to inactivate methanogens and subsequently acclimatized 3 times (3 days each time) using a modified culture medium to activate the spore-forming hydrogenogenic bacteria. The compositions of the modified medium were detailed in our previous study (Appendix C, table C.1) (Ding et al., 2016b).

The inoculum used in the biomethane potential (BMP) test and continuous digestion reactors was obtained from the digestate of an existing laboratory scale seaweed anaerobic digester. The methane inoculum was degassed at a temperature of 37 °C for 7 days before the experiment.

6.2.2 Biohydrogen and biomethane potential tests

The two-stage batch BHP and BMP tests on the mixture of *L. digitata* and *S. platensis* were conducted in triplicate in an AMPTS II system (Bioprocess Control, Sweden).

In the BHP test, 3 g VS of the algal substrate were added to each glass bottle and then the liquor volume was adjusted to 270 mL using distilled water. Subsequently, 30 mL of hydrogen inoculum was added into each bottle to make the total working volume 300 mL. The VS portions of the two algal biomass in each bottle were calculated to effect a C/N ratio of 20: 2.82 gVS of *L. digitata* mixed with 0.18 gVS of *S. platensis*. The initial pH was adjusted to 6 ± 0.05 with 1 M NaOH and 1 M HCl solutions. All bottles were sealed with rubber stoppers and purged with N_2 for 5 min to maintain anaerobic conditions, and then placed in a water bath at a temperature of 37 °C for 4 days. Stirrers which were set to switch between on and off for 60 s periods with a mixing speed of 60 rpm were applied to the bottles. Carbon dioxide in the produced gas was absorbed by 80 mL of 3 M NaOH solution and then the hydrogen gas flow was recorded by a gas tipping device based on water displacement. The recorded hydrogen gas volumes were automatically normalised to standard temperature and pressure (STP) and zero moisture content by the AMPST II system. After the BHP test, the effluent in each bottle was analysed and then prepared for subsequent BMP test. The pH values of effluents were adjusted to 8 ± 0.05 with 1 M NaOH and then inoculumted with methane inoculum at the inoculum to substrate VS ratio of 2:1. The total working volume of each bottle was 400 mL and the BMP test ran for 26 days so that the two-stage batch BHP and BMP tests duration reached 30 days. All the other BMP test settings were the same as those in the BHP test. A control group with just blank inoculum (no substrates) was established and all the hydrogen and methane volumes produced from experimental groups were corrected for the ones produced from control group.

6.2.3. Set-up and operation of continuous reactors

Four lab-scale (5 L) continuously stirred tank reactors (CSTR), which comprised of one H_2 reactor and three CH_4 reactors, were used for the continuous fermentation trials as shown in Figure 6.1. The H_2 reactor and CH_4 reactors A and B comprised the two-stage fermentation systems. The CH_4 reactor C acted as a one-stage fermentation system as a comparison to the two-stage system. The working volumes of H_2 reactor and CH_4 reactors were 3 L and 4 L, respectively. The temperature of the reactors was maintained at 37 ± 1 °C using a temperature controller unit. The volume of the produced biogas from each reactor was measured using a wet tip gas meter which was connected to an automated data acquisition system. The reactor configuration has been detailed in previous studies (Appendix C, table C.2.) (Herrmann et al., 2016; Voelklein et al., 2016).

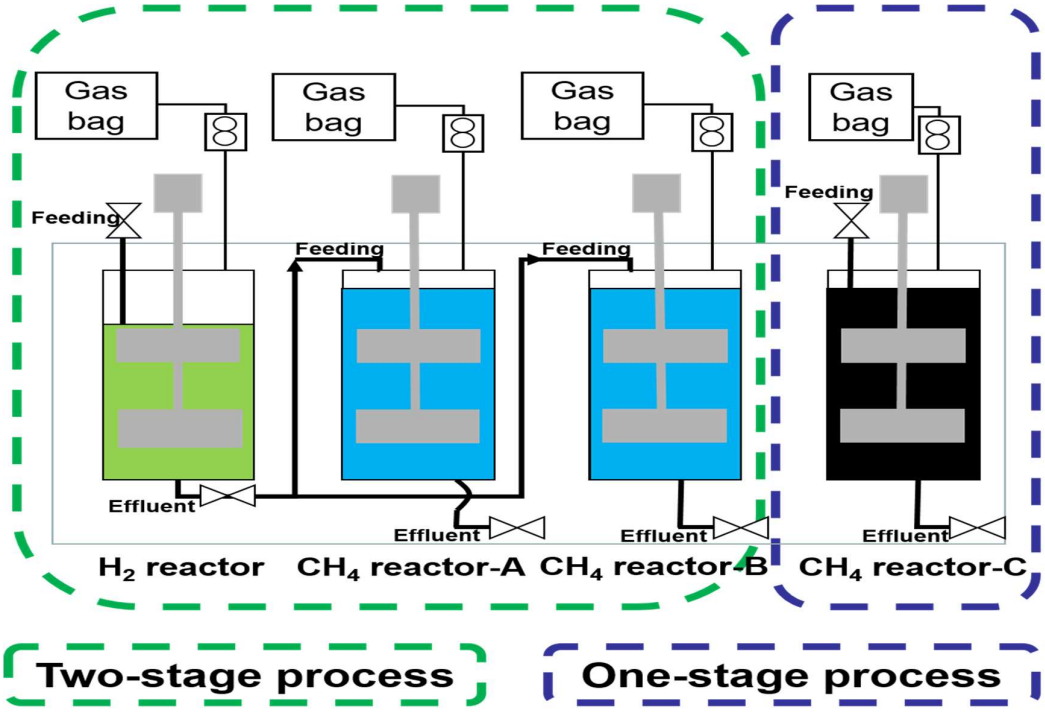


Figure 6.1 Schematic of continuous fermentation system

The HRT of the H_2 reactor was set to 4 days. The HRTs of CH_4 reactors A and B were set to 12 days and 24 days, respectively. The HRT of the one-stage CH_4 reactor C was set to 16 days to match the overall HRT of the first two-stage system comprising of the H_2 reactor and the CH_4 reactor A. In a similar fashion, the overall HRT of the second two-stage system comprising of the H_2 reactor and CH_4 reactor B was set to 28 days to match the one in a previous study that investigated the one-stage co-digestion of *L. digitata* and *S. platensis* for methane production (Herrmann et al., 2016).

The OLR of the H_2 reactor was increased from 3 to 12 kgVS/m^3/d with an increment of 3kgVS/m^3/d each time. This was achieved by diluting the algal biomass with a calculated volume of water to keep the HRT unchanged. Every time after feeding, the pH value in H_2 reactor was adjusted to ca. 5.5 using 1 M NaOH solution to ensure the pH did not drop to a level to inhibit hydrogen-producing microbes. The effluent from the H_2 reactor was divided into three parts: the first one as the feedstock for CH_4 reactor A, the second one as the feedstock for CH_4 reactor B, and the third one for analyses. The OLR of CH_4 reactor A ranged from 1 to 4 kgVS/m^3/d with an increment of 1 kgVS/m^3/d each time, whilst that of CH_4 reactor B increased from 0.5 to 2 kgVS/m^3/d with an increment of 0.5 kgVS/m^3/d each time. The OLR of the CH_4 reactor C (in the single stage system) started from 1 kgVS/m^3/d with an increment of 1 kgVS/m^3/d until reactor failure was observed. Each OLR of each reactor was maintained constant for 48 days, which equates to two HRTs of CH_4 reactor C, which had the longest retention time.

6.2.4. Analytical methods

Dry solids (DS) and VS contents of *L. digitata*, *S. platensis*, and inoculum were determined using

Standard Methods 2540 G (AMERICAN PUBLIC HEALTH ASSOCIATION (APHA), 2017). The pH value was measured using a pH meter (Jenway 3510, UK). The ratio of VFAs to total alkalinity (FOS/TAC) was determined based on a two points titration method using 0.1 N H_2SO_4 with end points of pH 5.0 and pH 4.4 (Drosg, 2013) .Carbon, hydrogen, and nitrogen contents were determined by an elemental analyser (Exeter Analytical CE 440, UK) and oxygen was calculated as the remaining content of VS. Soluble chemical oxygen demand (sCOD) and total ammoniacal nitrogen (TAN) were measured using Hach Lange cuvette tests (LCK 914 and LCK 303, respectively) and evaluated on a DR3900 Hach Lange Spectrophotometer. Salinity of effluents was determined on a VWR hand held C0310 monitor (VWR international, USA).

The composition of biogas (H_2, CO_2, O_2, N_2, and CH_4) produced in CSTR reactors was determined using a gas chromatograph (GC, Hewlett Packard HP6890, USA) equipped with a Hayesep R packed column and a thermal conductivity detector. The compositions of VFAs in the effluents were determined using a GC (Hewlett Packard HP6890, USA) equipped with a Nukol fused silica capillary column and a flame ionisation detector (Voelklein et al., 2016).

6.2.5 Calculations

The energy values of *L. digitata* and *S. platensis* were calculated using the weight percentages of C, H, N, and O on the basis of the modified Dulong Formula as shown in Eq. (10) (Nizami et al., 2009):

Energy value of algal biomass (kJ/kg) = 337 C+ 1419 (H-0.125 O)+ 23.26 N (Eq. 10)

The energy conversion efficiency (ECE) was calculated based on Equation 11 (Xia et al., 2013).

$$ECE = \frac{\text{Energy value of } H_2 + \text{Energy value of } CH_4}{\text{Original energy value of algal biomass}} \times 100\% \quad \text{(Eq. 11)}$$

The total chemical oxygen demand (tCOD) of algal biomass was calculated based on the element compositions using Equation 1:

$$C_aH_bO_cN_d + (a + \frac{b}{4} - \frac{c}{2} - \frac{3}{4}d)O_2 \rightarrow aCO_2 + \frac{b-3d}{2}H_2O + dNH_3 \quad \text{(Eq. 1)}$$

The acidification yield in the H_2 reactor is defined as the percentage of the COD from VFAs to sCOD as shown in Equation 13 (Voelklein et al., 2016):

$$\text{Acidification yield} = \frac{COD_{VFAs}}{sCOD_{increase}} \times 100\% \quad \text{(Eq. 13)}$$

The theoretical calculation of biomethane yield was based on the Buswell equation as shown in Equation 14 (Voelklein et al., 2016):

$$C_aH_bO_cN_d + (a - \frac{b}{4} - \frac{c}{2} + \frac{3}{4}d)H_2O \rightarrow (\frac{a}{2} + \frac{b}{8} - \frac{c}{4} - \frac{3}{8}d)CH_4 + (\frac{a}{2} - \frac{b}{8} + \frac{c}{4} + \frac{3}{8}d)CO_2 + dNH_3 \quad \text{(Eq. 14)}$$

6.3. Results and discussion

6.3.1 Characteristics of algal biomass

Table 6.1 presents the characteristics of *L. digitata* and *S. platensis* biomass. The macro-algae *L. digitata* was harvested from natural environments in shallow coastal waters, resulting in a lower VS/TS ratio as compare to the artificially cultivated micro-algae *S. platensis* which avoided the significant salt accumulation from seawater. The harvest timing of September coincided with the peak carbohydrate accumulation in *L. digitata* biomass (Tabassum et al., 2017), leading to a high C/N ratio of 26.47. By contrast, the rich proteins in *S. platensis* contributed to the high nitrogen content.

Table 6.1. Characteristics of algal biomass

Parameter	*Laminaria digitata*	*Arthrospira platensis*	Mixed *Laminaria digitata* and *Arthrospira platensis*
Proximate analysis			
Moisture (wt%)	81.87	6.40	81.16
DS (wt%)	18.13	93.60	18.84
VS (wt%)	13.31	86.77	14.01
VS/DS (%)	73.44	92.70	74.34
Ultimate analysis			
C (DS%)	36.08	49.27	36.70
H (DS%)	4.67	6.58	4.76
O (DS%)	31.32	25.48	1.84
N (DS%)	1.36	11.38	31.05
C/N ratio	26.47	4.33	20.00
Biological analysis			
Proteins (DS%)	7.32[a]	71.13[a]	10.32
Lipids (DS%)	0.92[b]	5.00[c]	1.11
Carbohydrates (DS%)	65.20[d]	16.57[d]	62.91
Energy value (kJ/gVS)	18.1	23.4	18.4
tCOD (gCOD/gVS)	1.36	1.50	1.37
Theoretical biomethane yield (L/kgVS)	476.3	525.2	479.2

a The contents of proteins are calculated by multiplying the nitrogen contents by a factor of 5.38 for brown seaweeds (Lourenco et al) and 6.25 for microalgae (Richmond et al).
b The lipid content of *Laminaria sp.* is suggested to be 0.92% of the dry weight by Sánchez-Machado et al..
c The lipid content of *Arthrospira sp.* is suggested to be 5% of the dry weight by Dismukes et al..
d It is assumed that the sum of proteins, lipids, carbohydrates equates to the VS of algal biomass.

This also provided the possibility of mixing the two algal substrates at an appropriate C/N ratio of 20. Moreover, *S. platensis* biomass exhibited higher energy content and theoretical biomethane potential on the basis of elemental composition, despite potential antagonistic effects of recalcitrant organic components on the biodegradability (Herrmann et al., 2016). *L. digitata* biomass is rich in carbohydrates, which generate 20 times higher hydrogen-producing potential than proteins and lipids (Lay et al., 2003) and as such serve as the major components utilised by the AFB for biohydrogen production. *S. platensis* are rich in proteins and can supply essential nitrogen sources for the anaerobes in both H_2 and CH_4 reactors to maintain effective metabolism (Xia et al., 2016b). The lipid content is relatively low in both algal species and is not readily utilised by the AFB for hydrogen production. These lipids however can be slowly degraded and further converted to biomethane in the second-stage anaerobic digestion with a longer retention

time (Xia et al., 2016a).

6.3.2. Batch biohydrogen and biomethane potential tests

After the sequential 4-day BHP and 26-day BMP tests using the mixed *L. digitata* and *S. platensis* biomass, a BHP yield of 94.6 mL H_2/gVS and a BMP yield of 309.3 L CH_4/gVS were recorded (Figure 6.2). The BHP yield exceeds the result (60.5 L H_2/kgVS) obtained in a previous study using algal mixture of *L. digitata* and *S. platensis* at a C/N ratio of 16.5 (Xia et al., 2016b) , indicating the C/N ratio of 20 is preferred during the batch hydrogen fermentation of this specific algal mixture. Moreover, the BHP yield is close to the findings (94.5-97 H_2 L/kgVS) of our previous study on batch hydrogen co-fermentation of macro-algae (*L. digitata*) and micro-algae (*Chlorella pyrenoidosa* and *Nannochloropsis oceanica*).

After hydrogen fermentation, the VFA compositions in the hydrogenogenic effluent were as follows: 0.64 g/L of acetic acid, 0.02 g/L of propionic acid, 0.02 g/L of isobutyric acid, 0.97 g/L of butyric acid, 0.03 g/L of isovaleric acid, and 0.01 g/L of valeric acid. The acetic and butyric acids accounted for 95.1% of the total VFAs, indicating that the predominant metabolic pathways of the AFB during hydrogen fermentation were acetic and butyric routes (Xia et al., 2016a). As shown in Figure 6.2b. during subsequent BMP test, the soluble VFAs that are readily utilised by methanogens contributed to the first peak of biomethane production rate at 6 days, whereas the solid remnants continued to be hydrolysed and resulted in the second peak of biomethane production rate at 12 days. The BMP yield matches that from the one-stage batch anaerobic co-digestion of *L. digitata* and *S. platensis* (311.5 L CH_4/gVS) achieved by (Herrmann et al., 2016). Although no significant enhancement of BMP yield was obtained, the two-stage batch co-

fermentation of *L. digitata* and *S. platensis* secured an overall energy yield of 12.1 kJ/gVS that is 8.5% higher than that from the one-stage biomethane production (Herrmann et al., 2016).

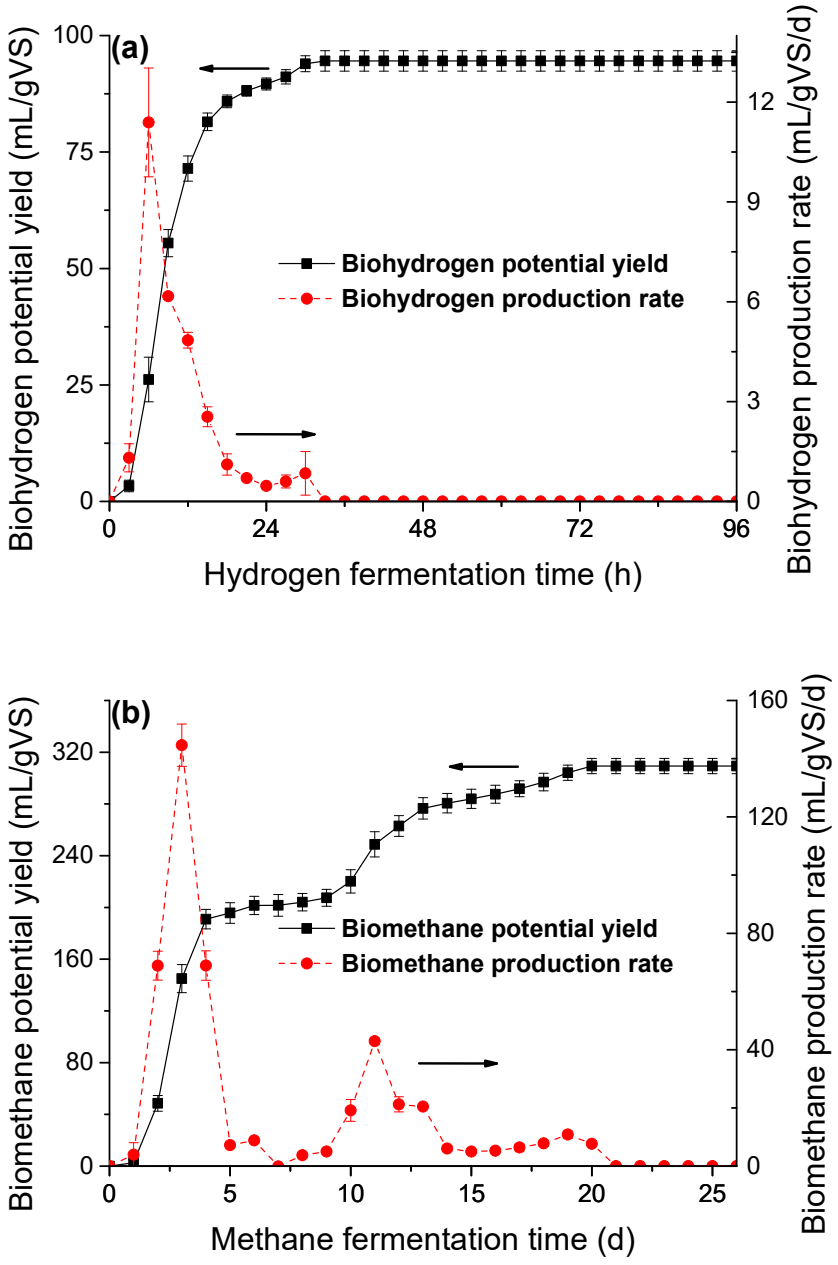


Figure 6.2 Two-stage batch biohydrogen and biomethane co-production from mixed *L. digitata* and *S. platensis* biomass at a C/N ratio of 20: (a) biohydrogen production and (b) biomethane production. Arrows indicate the peaks on the cumulative yields.

6.3.3. Continuous fermentation performances with increasing OLRs

The performance characteristics of all four reactors of the two-stage and one-stage systems over increasing OLRs are summarised in Table 6.2. The first HRT at each OLR in each reactor was deemed as the acclimatisation period for anaerobic microbes, thus the data in Table 6.2 are displayed as mean values over the post-first HRT duration of each OLR. Throughout the entire experiment, the TAN concentrations of all CH_4 reactors stayed low, indicating that no ammonia inhibition occurred.

6.3.3.1. Performance of H_2 reactor

Figure 6.3 shows the SHYs of the H_2 reactor with increasing OLRs; Figure 6.4a shows the compositions of VFAs. At the initial OLR of 3 kgVS/m^3/d, the SHYs were quite limited. However, the acidification yield reached 87.5%, indicating a large portion of mixed *L. digitata* and *S. platensis* were utilised by the AFB to maintain basic metabolisms. Thus, the low mean SHY (14.3 L/kgVS) and the high acidification yield at this low OLR indicated that the AFB in H_2 reactor were underfed to some extent. When the OLR increased from 3 to 6 kgVS/m^3/d, the SHYs drastically increased. Although the SHYs fluctuated between 40.5 and 72 L/kgVS over this OLR, an average of 55.3 L/kgVS was achieved, which equates to 58.5% of the BHP yield in the batch trial. As the sCOD of 14.2 g/L at this OLR (6 kgVS/m^3/d) was over 2-fold of that (7 g/L) at the initial OLR (3 kgVS/m^3/d), it could be assumed that the hydrolysis of mixed algal substrates was even a little bit more efficient. The tVFA also increased to 5,254 mg/L, corresponding to an acidification yield of 63%. Similarly, the salinity increased by 55.6%, illustrating that this OLR provided excessive biomass supply for the basic metabolisms of AFB and hence more algal substrates were degraded and utilised for hydrogen production.

When the OLR was further lifted from 6 to 9 kgVS/m^3/d, a sharp drop in hydrogen production was recorded. The mean SHY of 20.4 L/kgVS was 63.1% lower than that at the OLR of 6 kgVS/m^3/d. This result was attributed to the accumulation of large quantities of VFAs that inhibited the hydrogen-producing pathways of AFB in the H_2 reactor. The increased loading of algal substrates resulted in sCOD and tVFA values higher by 29.6% and 26.1% in the liquid phase, respectively, whereas the remaining VS in the H_2 reactor (at 9 kgVS/m^3/d) increased by 57.5%.

As the increase in remaining VS exceeded the increase in sCOD and tVFA, it was assumed that H_2 reactor was overfed and hydrolysis and acidification of loaded algal substrates were limited to some extent. With the OLR further rising to 12 kgVS/m^3/d, the average SHY marginally declined to 19 L/kgVS. Although the sCOD slightly increased, the tVFA unexpectedly decreased a little bit, leading to a lower acidification yield as compared to that at the OLR of 9 kgVS/m^3/d. This also indicated that more algal substrates were fermented through ethanol and lactic acid producing pathways. This was probably ascribed to the enhanced fluctuations of pH values at higher OLRs. With the loading increasing, soluble acidic metabolites accumulated and hence the pH drop became more severe between each feed. The lower pH facilitated the shift of acetic and butyric routes to ethanol and lactic acid producing pathways in the H_2 reactor (Chen et al., 2015; Dębowski et al., 2013; Voelklein et al., 2016).

Table 6.2 Summary of results from two-stage and one-stage co-fermentation of *L. digitata* and *S. platensis* (mean values of post-first HRT for each OLR)

	H_2 reactor				CH_4 reactor A				CH_4 reactor B				CH_4 reactor C		
HRT (days)	4				12				24				16		
OLR (kgVS/m^3/d)	3	6	9	12	1	2	3	4	0.5	1	1.5	2	1	2	3
SHY (L/kgVS)	14.3	55.3	20.4	19.0	/	/	/	/	/	/	/	/	/	/	/
SMY (L/kgVS)	/	/	/	/	265.5	245.0	229.1	174.0	242.5	228.9	223.8	236.5	204.5	134.8	72.2
FOS/TAC	/	/	/	/	0.22	0.17	0.21	0.27	0.19	0.17	0.17	0.17	0.61	1.03	1.68
TAN (mg/L)	7	2	4	5	216	148	251	269	281	197	290	279	95	43	158
TS (g/kg)	14.3	23.8	37.9	45.3	11.8	12.8	18.9	23.9	17.3	13.3	19.4	23.5	12.2	26.3	47.6
VS (g/kg)	9.4	15.3	24.1	29.5	5.6	5.4	6.7	9.3	8.7	5.4	7.3	8.0	6.7	12.0	22.5
sCOD (g/L)	7.0	14.2	18.4	21.5	0.6	0.9	2.3	5.2	0.8	0.7	1.4	2.2	2.5	10.3	21.7
tVFA (mg/L)	3776	5254	6626	6587	354	349	877	1365	243	287	279	551	1287	6593	5982
COD_{VFAs} (g/L)	6.2	8.9	11.4	11.5	0.6	0.6	1.4	2.1	0.4	0.5	0.5	0.9	2.1	8.9	10.5
Acidification yield (%)	87.5	63.0	62.2	53.5	/	/	/	/	/	/	/	/	/	/	/
Salinity (g/kg)	3.6	5.6	6.5	5.8	4.6	6.4	8.1	5.6	6.6	6.5	8.1	7.7	4.5	9.7	13.3
Energy yield (kJ/gVS)	0.2	0.6	0.2	0.2	9.5	8.8	8.2	6.2	8.7	8.2	8.0	8.5	7.3	4.8	2.6
ECE (%)	0.8	3.3	1.2	1.1	51.7	47.7	44.6	33.9	47.2	44.6	43.6	46.0	39.8	26.2	14.1

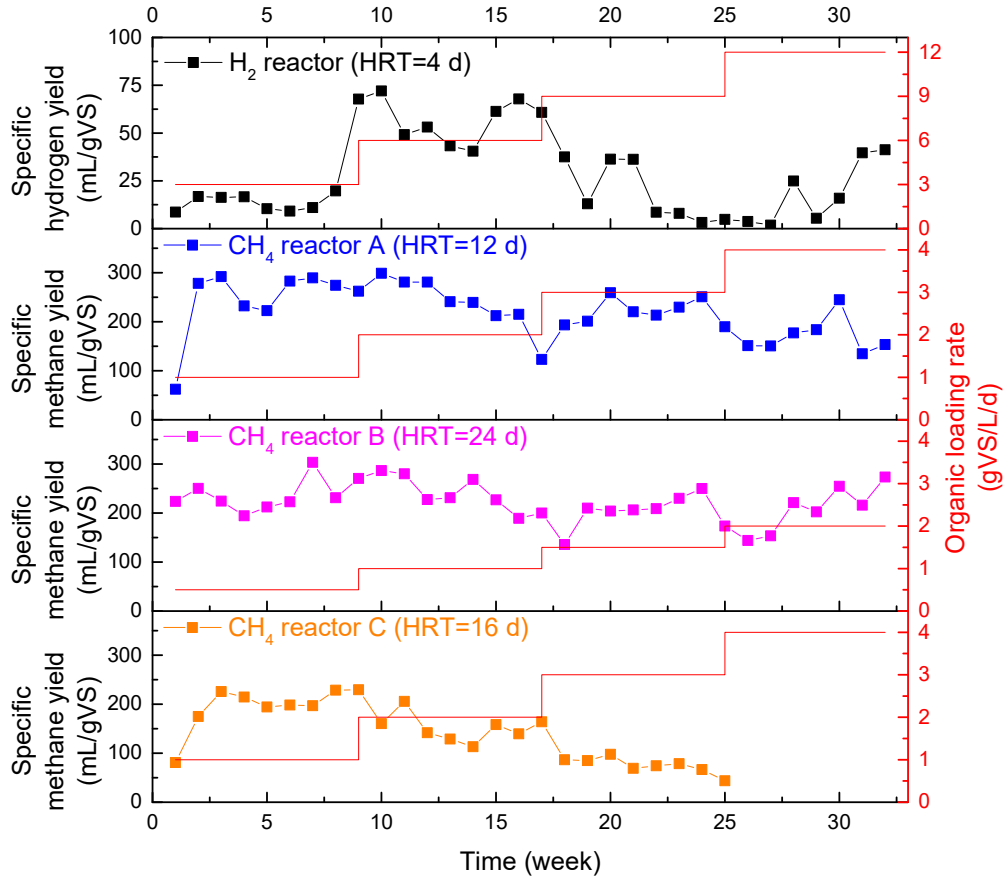


Figure 6.3. Specific hydrogen yields of H_2 reactor and specific methane yields of CH_4 reactors A, B, and C with increasing organic loading rates in continuous two-stage and one-stage systems

These results suggested that the optimum OLR for continuous biohydrogen production through co-fermentation of macro-algae *L. digitata* and micro-algae *S. platensis* was 6 kgVS/m^3/d in the H_2 reactor. The insufficient biomass supply at lower OLR failed to provide essential feedstock for the AFB to produce hydrogen, whereas the overfeeding of algae at higher OLRs resulted in the accumulation of VFAs which in turn suppressed the hydrogen-producing metabolisms.

6.3.3.2. Performance of CH_4 reactors A and B

The SMYs of CH_4 reactors A and B of the two-stage system and the variation trends of tVFA and

FOS/TAC values over increasing OLRs are illustrated in Figure 6.3 and Figure 6.5, respectively. At the initial OLR of 1.0 kgVS/m^3/d, CH_4 reactor A performed best with an average SMY of 265.5 L/kgVS which accounted for 85.8% of the BMP value in the batch trial. The sCOD and tVFA were low at 0.6 g/L and 354 mg/L, respectively, indicating that most of the soluble metabolites produced via first-stage dark hydrogen fermentation were utilised by the microbes in CH_4 reactor A. The FOS/TAC value was low (0.22) as well. When the OLR increased to 2 kgVS/m^3/d, the average SMY slightly decreased to 245 L/kgVS, signifying 79.2% of the BMP yield.

The low FOS/TAC value of 0.17 ensured the process stability of second-stage anaerobic digestion. Under the conditions of higher sCOD and tVFA inputs from effluents of the H_2 reactor, the sCOD and tVFA values of CH_4 reactor A remained almost as low as those at the previous OLR of 1 kgVS/m^3/d, resulting in even higher sCOD and tVFA destruction efficiencies (93.7% and 93.3%, respectively). The continuous increase of OLR from 2 to 3 kgVS/m^3/d further led to a 9.4% drop in SMY. Although the FOS/TAC value remained within a suitable range, both the VFAs and sCOD increased. The average tVFA value of 877 mg/L was not high, however, the variation trend shown in Figure 6.5 implied that the accumulation of VFAs was in progress. Especially as shown in Figure 6.4b, the content of propionic acid in CH_4 reactor A significantly increased at 3 kgVS/m^3/d as compared to the lower loading rates.

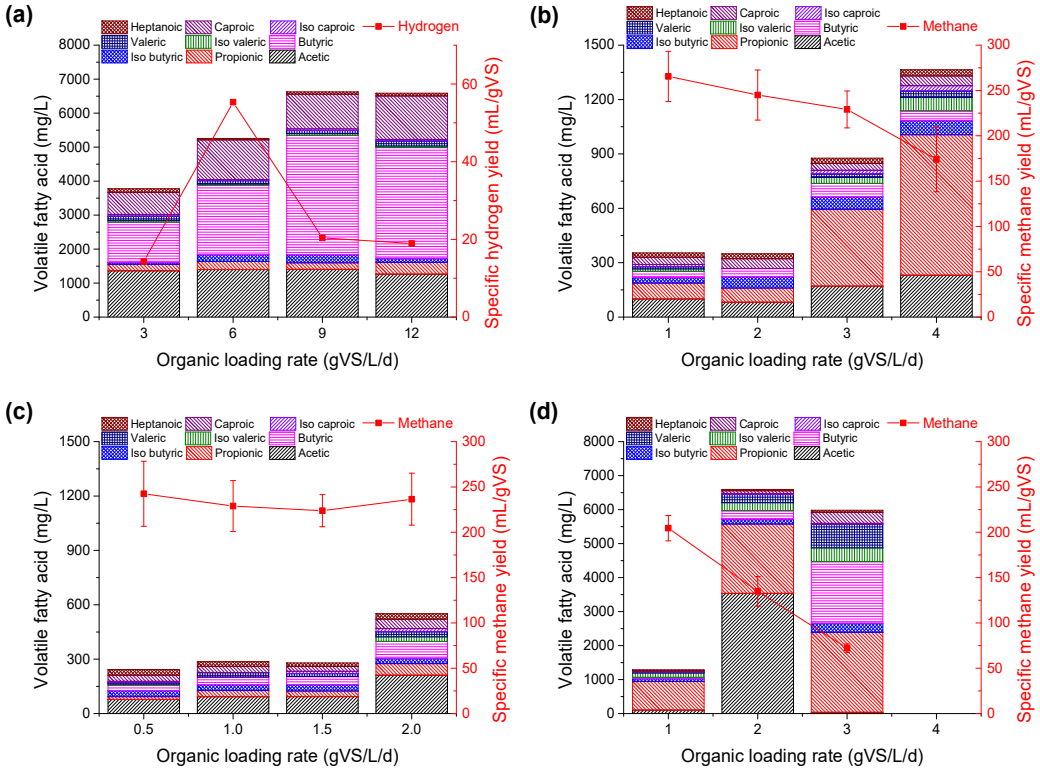


Figure 6.4 Compositions of VFAs with increasing organic loading rates in (a) H_2 reactor, (b) CH_4 reactor A of two-stage system, (c) CH_4 reactor B of two-stage system, and (d) CH_4 reactor C of one-stage system

The accumulation of propionic acid in the digester is always deemed as an indicator of impending anaerobic digestion failure (Gallert and Winter, 2008; Pullammanappallil et al., 2001). At the maximum OLR of 4 kgVS/m^3/d, a notable reduction in SMY was recorded: the SMY of 174 L/kgVS was lower than that at 3 kgVS/m^3/d by 24.1% and only equivalent to 65.5% of the highest one obtained at 1 kgVS/m^3/d. The sCOD and tVFA further accumulated in CH_4 reactor A. The average FOS/TAC value increased to 0.27 and the variation trend shown in Figure 6.4 suggested that the FOS/TAC of CH_4 reactor A was rising towards the threshold value. Figure 6.4b shows that the propionic acid concentration further increased to 775 mg/L and almost all the iso-acids were higher, illustrating that the process instability of CH_4 reactor A caused by the overloading of mixed algal biomass was in progress (Gallert and Winter, 2008). The struggling of CH_4 reactor A at higher OLRs could be associated with the inability of the microbial community

to acclimatise to such a high loading in a short HRT of 12 days. This may have resulted in washout of microbial community.

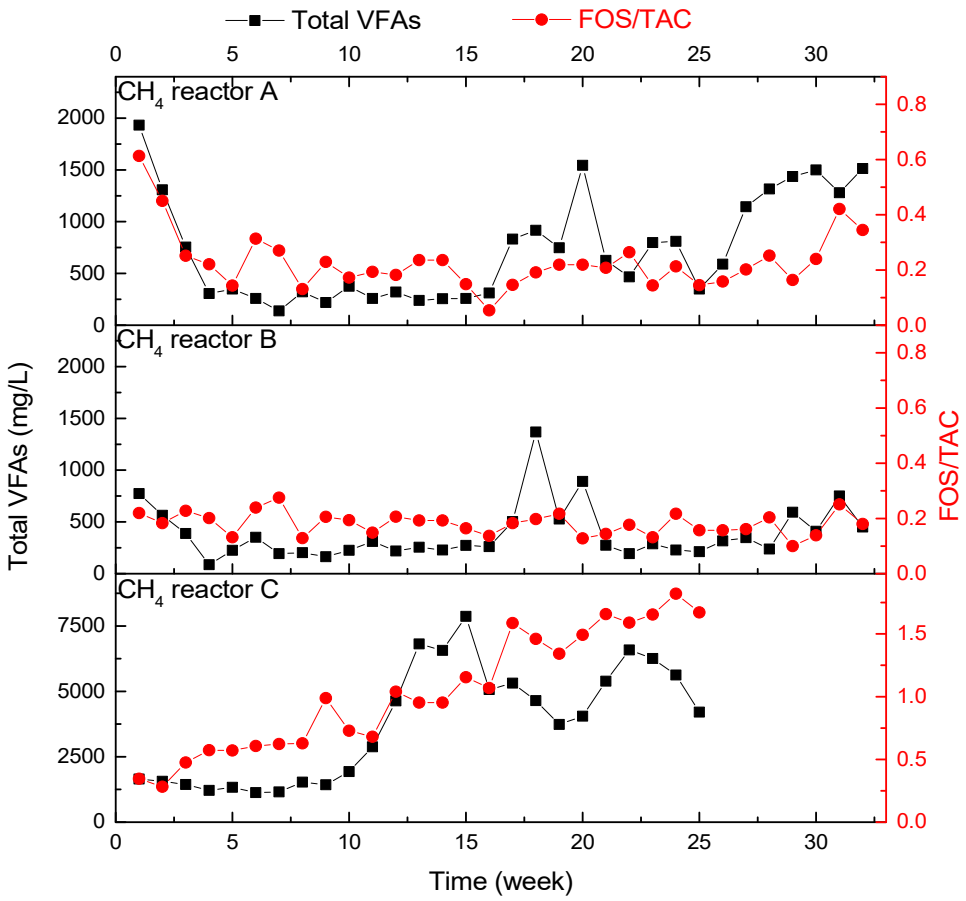


Figure 6.5 Concentrations of total VFAs and FOS/TAC values in CH_4 reactors A, B, and C during continuous anaerobic digestion

Since CH_4 reactors A and B shared the same feedstock origin (effluent from H_2 reactor), the 2-fold HRT of CH_4 reactor B led to lower OLRs which equates to half of those of CH_4 reactor A. The FOS/TAC values remained low (0.17-0.19) throughout the entire continuous experiments, indicating that a more stable second-stage anaerobic digestion process was ensured by the longer HRT and lower OLRs of CH_4 reactor B as compared to CH_4 reactor A. Although the SMYs were marginally lower than the highest one obtained in CH_4 reactor A, the average values in CH_4

reactor B were less affected by the increasing OLR from 0.5 to 2 kgVS/m^3/d and remained within a reasonable range of 223.8-242.5 L/kgVS signifying 72.4-78.4% of the BMP value and 46.7-50.6% of the theoretical methane yield. The sCOD and tVFA stayed low over increasing OLRs, leading to the high sCOD (88.6-95.1%) and tVFA (92.2-95.6%) destruction efficiencies. However, the highest average sCOD (2.2 g/L) and tVFA (551 mg/L) recorded at the maximum OLR of 2 kgVS/m^3/d were both higher than those in CH_4 reactor A at the same OLR. This was caused by the feedstock sourced from the effluent of the H_2 reactor at various OLRs. At an OLR of 2 kgVS/m^3/d, the feedstock loaded into CH_4 reactor B was obtained from the effluent of the H_2 reactor at an OLR of 12 kgVS/m^3/d, whilst the one loaded into CH_4 reactor B was originated from the effluent of the H_2 reactor at an OLR of 6 kgVS/m^3/d. The sCOD and tVFA values of the former was markedly higher than the latter, resulting in a comparatively more severe impact on the second-stage anaerobic digestion process. Nonetheless, Figure 6.4c reveals that no accumulation of propionic acid or iso-acids in CH_4 reactor B were observed at an OLR of 2gVS/m^3/d, demonstrating that no inhibition of methanogens or anaerobic digestion process failure was evident.

Overall, considering SMY, treating capacity, and process stability, an OLR of 2 kgVS/m^3/d was shown to be optimal for CH_4 reactor A at a fixed HRT of 12 days.

6.3.3.3 Performance of CH_4 reactor C

The SMYs of CH_4 reactor C of the one-stage system are shown in Figure 6.3. With the OLR increasing from 1 to 3 kgVS/m^3/d, the average SMYs gradually decreased from 204.5 to 72.2 L/kgVS. As shown in Figure 6.5, the VFAs accumulated and the FOS/TAC values rose along with the increasing OLR, indicating that the buffer capacity in the CH_4 reactor C was strongly negatively correlated with OLR in this one-stage system. At the initial OLR of 1 kgVS/m^3/d, the tVFA already reached 1287 mg/L and the VFA composition in Figure 6.4d revealed that propionic acid accounted for 65.6% of the tVFA. This phenomenon of propionic acid accumulation was

similar to that obtained in the CH_4 reactor A at the maximum OLR of 4 kgVS/m^3/d, signifying that the process instability of one-stage anaerobic co-digestion was triggered. When the OLR rose to 2 kgVS/m^3/d, a remarkable surge in VFAs was noted: the tVFA concentration of 6,593 mg/L was even close to that in the H_2 reactor at 9 kgVS/m^3/d. It was assumed that the methanogens in CH_4 reactor C suffered severe inhibition under such acidic condition. When the OLR further increased to 3 kgVS/m^3/d, the sCOD increased by 110.7%, whereas the tVFA slightly decreased instead, indicating that the acidification process was impaired even though the hydrolysis was efficient. In addition, the enhancements of propionic, butyric, and longer-chain acids and little accumulation of acetic acid were recorded in Figure 6.4d. These results suggested that the microbial community was highly affected: the activity of acetogens and methanogens were inhibited to a great extent. Furthermore, the salinity in CH_4 reactor C amounted to 13.3 g/kg, which was far higher than the highest ones obtained in CH_4 reactors B and C during the entire experiment. Although small concentrations of sodium ions (100-350 mg/L) are supposed to be essential for the maintenance of healthy metabolism of the microbes in anaerobic digesters (Murphy et al., 2015), the enhanced osmotic pressure caused by the remarkably high salinity can inhibit microbial activity and even lead to dehydration of microbes (Ward et al., 2014). Luo et al. (Luo et al., 2013) investigated the effects of saline adaptation on anaerobic digestion of sludge and observed that salinity levels higher than 8.7 g/kg impaired the methane production. On the other hand, Tabassum et al. (Tabassum et al., 2016) demonstrated acclimatisation to salinity levels of the order of 14 g/L in mono-digestion of farm cultivated S. *latissima* at an OLR of 4kgVS/m^3/d. The high salinity levels recorded here of 13.3g/kg at an OLR of 3 kgVS/m^3/d will have some inhibitory effects on the microbial consortium in CH_4 reactor C. Although the gas production did not thoroughly stop, the failure of CH_4 reactor C was inevitable.

In a previous study, (Herrmann et al., 2016) conducted continuous one-stage anaerobic co-digestion of *L. digitata* and *S. platensis* based on a C/N ratio of 25 at a long HRT of 28 days. A high OLR of 4 kgVS/m^3/d was shown to be tolerable for the CH_4 reactor and an SMY of 259.6

L/kgVS was recorded. Despite the different seed inoculum and minor variation in C/N ratios, the significant reduction in HRT (28 days as compared to 16 days here) was assumed to be the key influencing factor between these two one-stage systems. It is suggested that an HRT of 16 days did not supply sufficient time for acclimatisation and enrichment of the microbial consortium in the CH_4 reactor C and led to washout of microbes, accumulation of VFAs, and inhibition of methanogenesis.

6.3.4. Comparisons between two-stage and one-stage fermentation performances

The two-stage system comprising of the H_2 reactor and the CH_4 reactor A and the one-stage system of CH_4 reactor C shared comparable operational parameters such as overall HRT (16 days), OLR, temperature (37 ± 1 °C), and initial seed inoculum for methane production. At an OLR of 6 kgVS/m^3/d, the highest average SHY of 55.3 L/kgVS, which equates to 58.5% of the BHP yield in batch trail, was obtained in the first-stage dark hydrogen fermentation. In the second-stage anaerobic digestion, the average SMY of 245 L/kgVS equivalent to 79.2% of the BMP value was achieved in CH_4 reactor A at a corresponding OLR of 2 kgVS/m^3/d, and process stability was secured. The two-stage system effected an energy yield of 9.4 kJ/gVS and the ECE amounted to 51.0%. The energy yield of the continuous two-stage system was 22.3% lower than the batch trial. This is expected due to the disadvantages of shorter retention time (16 days in two-stage versus 30 days for batch) and the larger reactor with less efficient mixing conditions. By contrast, in the one-stage system, the CH_4 reactor C recorded its highest SMY of only 204.5 L/kgVS at the initial OLR of 1 kgVS/m^3/d. The energy yield and ECE were lower at 7.3 kJ/gVS and 39.8%, respectively. Even at this low OLR, a certain degree of VFA accumulation was observed. When the OLR rose to 3 kgVS/m^3/d, the process instability of one-stage anaerobic co-digestion of *L. digitata* and *S. platensis* became more obvious. Therefore, the two-stage system prevailed in both energy production from mixed algal feedstock and treating capacity as compared to one-stage system at a fixed HRT of 16 days. Even if the energy content in produced

hydrogen was nearly negligible, the first-stage dark hydrogen fermentation would serve as an optimised hydrolysis and acidification method pretreating the mixed algal feedstock. Similar results were reported by (Massanet-Nicolau et al., 2015; Voelklein et al., 2016) utilising grass and food waste in continuous two-stage systems. To sum up, the technical feasibility of two-stage co-fermentation of *L. digitata* and *S. platensis* biomass has been proven, and several operational parameters have been assessed via this 32-week long experimentation, thus mitigating the gaps between the fundamental innovations obtained by the small-scale batch co-fermentation and the potential commercial deployment of algal biofuel systems in future.

Although positive results on two-stage continuous hydrogen and methane co-production using mixed *L. digitata* and *S. platensis* have been achieved in this study, some issues are still noteworthy. The C/N ratio was adjusted to 20 in the mixture of macro- and micro-algae, however, the TAN levels stayed low in all four reactors throughout the entire continuous experiment, indicating that the hydrolysis or degradation of nitrogen-rich micro-algae biomass may have been somewhat limited, especially in a short HRT of 16 days. This was probably ascribed to limited degradation of untreated *S. platensis* due to its recalcitrant cell wall structures. The slow or limited utilisation of micro-algae biomass further restricted the fermentation/digestion process and also explained why the longer HRT in CH_4 reactor B and in the previous study (Herrmann et al., 2016)could ensure a more stable process. Therefore, to overcome this drawback, pretreatment of micro-algae and even macro-algae to facilitate the solubilisation and hydrolysis of feedstock is a promising option for a stable continuous fermentation/digestion process in future study.

6.3.5. Comparison between results of this study and relevant literature

To the best of our knowledge, most of the studies on biohydrogen and biomethane production from either macro- or micro-algae biomass were conducted in batch trials (Montingelli et al., 2015; Ward et al., 2014) . The data on long term continuous fermentation of algae are relatively limited. A comparison between the results of continuous fermentative gaseous biofuel production

from algal biomass and other co-substrates in this study and the state of the art in the literature is summarised in Table 6.3. Tabassum et al. (Tabassum et al., 2016) found that a mixed feedstock of 66.6% macro-algae (*L. digitata* or *S. latissima*) and 33.3% dairy slurry was optimal to obtain a maximum biomethane production efficiency during continuous anaerobic co-digestion. The energy yields (9-9.3 kJ/gVS) were close to that obtained in this study. Allen et al. (Allen et al., 2014) suggested for the green macro-algae (*Ulva lactuca*) that the optimal mixture in long term continuous digestion would be 25% macro-algae and 75% dairy slurry; this resulted in an SMY of 170 L/kgVS equivalent to 95% of the BMP value. These differences are attributed to the significant variation in biological characteristics of different macro-algal species. The green seaweed *U. lactuca* typically has a C/N ratio below 10 and as such needs to be co-digested with a carbohydrate rich co-substrate to increase the C/N ratio for better digestibility.The carbohydrate rich brown seaweeds *L. digitata* and *S. latissima* have high C/N ratios (>25) when they are *ripest* in late summer (Tabassum et al., 2016). Similarly, the protein-rich Taihu blue algae with a low C/N ratio of 6.1 resulted in an SMY of 160 L/kgVS, whereas the mixture of Taihu blue algae and carbohydrate-rich corn straw with a C/N ratio of 20 resulted in an increase in SMY of 46% (Zhong et al., 2013). Herrmann et al. (Herrmann et al., 2016) also used micro-algae *S. platensis* as a nitrogen-rich additive to macro-algae *L. digitata* for adjusting the C/N to 25. Compared with the results obtained in the one-stage reactor in this study, the longer HRT (28 days) allowed a higher OLR (4 kgVS/m^3/d) with a stable process and a higher SMY. All the above studies were conducted in a one-stage system; only one previous study investigated two-stage continuous fermentation of macro-algae *L. digitata* (Guneratnam et al., 2017). In this work the two-stage fermentation system outperformed the one-stage system with a higher energy yield in a shorter overall HRT (Guneratnam et al., 2017). This finding was consistent with the output of this study. The optimal HRT, OLR, and biofuel yields varied between the studies due to different experimental configurations, different sources of inoculum and different algal feedstocks. However, the results showed similarities in C/N ratios, and the improvement in energy return and

process stability.

Table 6.3. Comparison between the results in this study and relevant literatures on continuous fermentative gaseous biofuel production from algal biomass

Algal species	Co-substrate	Fermentation type	HRT (d)	OLR (kgVS/m^3/d)	SHY (L/kgVS)	SMY (L/kgVS)	C/N ratio	Energy yield (kJ/gVS)	Reference
Laminaria digitata	Dairy slurry	One-stage CH_4	18	4	/	261	23.4	9.3	Tabassum et al
Saccharina latissima		fermentation	13	4	/	252	15.7	9.0	
U.lactuca	Dairy slurry	One-stage CH_4	42	2	/	170	16.6	6.1	Allen et al
Taihu blue algae	/	One-stage CH_4 fermentation	10	6	/	160	6.1	5.7	Zhong et al
	Corn straw			6	/	234	20	8.4	
Laminaria digitata	*Arthrospira platensis*	One-stage CH_4	28	4	/	259.6	25	9.3	Herrmann et al
Laminaria digitata	/	One-stage CH_4	24	2	/	221	27.3	7.9	Guneratnam et al
		Two-stage $H_2 + CH_4$ fermentation	4 (H_2) + 14 (CH_4)	12 (H_2) + 3.43 (CH_4)	26	234		8.7	
Laminaria digitata	*Arthrospira platensis*	One-stage CH_4 fermentation	16	1	/	204.5	20	7.3	This study
		Two-stage $H_2 + CH_4$ fermentation	4 (H_2) + 12 (CH_4)	6 (H_2) + 2 (CH_4)	55.3	245.0		9.4	
		Two-stage $H_2 + CH_4$ fermentation	4 (H_2) + 24 (CH_4)	12 (H_2) + 2 (CH_4)	19	236.5		8.7	

6.3.6. Gross energy potential from algal mixture

In this study, the major component in the algal mixture is macro-algae *L. digitata*, which accounts for 94% of the VS. The co-substrate micro-algae may be considered as a nitrogen-rich additive. Therefore, the gross energy potential from this mixed algal feedstock is heavily associated with the *L. digitata* biomass resource. Nonetheless, the definite data on the annual yields of seaweed per hectare are not available because of a series of variations, such as algal species, locations, harvesting times, etc (Murphy et al., 2015). According to a latest report of International Energy Agency Bioenergy, the yields of *L. digitata* cultivated using advanced textiles in open sea reached 16 kg/m^2, equivalent to 160 tons wet weight per hectare per year (t wwt/ha/yr) (Laurens, 2017) Under this scenario, based on the energy yield of 9.4 kJ/gVS in the two-stage continuous co-fermentation system, the gross energy potential is calculated to be 213 GJ/ha/yr. This value is comparable with the gross energy yields of biomethane from terrestrial crops, such as maize (217 GJ/ha/yr), fodder beet (250 GJ/ha/yr), and grass (163 GJ/ha/yr) (Murphy et al., 2011). The advantages of *L. digitata* cultivation, are that as an advanced third generation biofuel there is no requirement for arable land, the fuel is outside the food-or-fuel debate, and it is an attractive process for countries with long coastlines (Murphy et al., 2015) . In addition, integrated multi-trophic aquaculture (coupling seaweed production with fish farms) captures nutrients from fish excrement enhancing seaweed growth and water quality (Tabassum et al., 2017), and leading to promotion of industrial scale advanced gaseous biofuel production from algal biomass.

6.4. Conclusions

A continuous two-stage system involving dark hydrogen fermentation and anaerobic fermentation of mixed macro-algae and micro-algae at a C/N ratio of 20 was shown to be feasible with an overall ECE of 51.0%. The short HRT (16 days) allowed an efficient fermentation process in the H_2 reactor at 6 kgVS/m^3/d and a stable digestion process in the CH_4 reactor at a corresponding

OLR of 2 kgVS/m^3/d. In contrast to the one-stage system, the first-stage dark hydrogen fermentation in the two-stage system optimised hydrolysis and acidification of algal mixtures, hence facilitating improved methane production and process stability in second-stage anaerobic digestion. The gross energy potential of 213 GJ/ha/yr makes this algal mixture comparable with terrestrial crops in gaseous biofuel production while removing any land use implications.

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7. Conclusions and recommendations

7.1. Conclusions

Biogas production in Mexico has a great energy potential and economic viability with the current technologies and feedstock available in the country. A biogas economy can be built around the use of wastes in biogas plants. The use of algae as feedstock for anaerobic digestion has a significant biogas potential depending on the species and on the anaerobic digestion technologies applied. Algae can be included into the feedstock mix alongside with wastes and ligno-cellulosic substrates specially on coastal areas helping to establish a circular economy.

The detailed conclusions of the thesis are as follows:

- Biomethane as a transport fuel from food waste in Mexico has a significant theoretical potential of approximately 42 PJ/year. The use of biomethane as a transport fuel can help reduce Mexico's GHG emissions targets by 6.06%, which signifies a reduction of 17.91 $MtCO_2$.
- An urban biogas plant of a capacity of approximately 60,000 t/year co-digesting FW and sewage sludge can be economically feasible (positive NPV). A biogas plant of approximately the same size co-digesting pig slurry and FW has negative NPV. The lack of gate fees for the handling and final disposal of pig slurry directly impacts on the economically feasibility of the plant.
- Economic instruments such as clean energy certificates and subsidies for the production of renewable fuels are needed in order to improve the economics of biogas plants for the production of biomethane as a transport fuel.
- For a Mexican city, the co-digestion of FW and sewage sludge for the production of biomethane is currently preferable from an economic standpoint.
- Anaerobic digestion from pig slurry in Mexico has the potential to produce 584 Mm^3 of methane with an energy content of 21 PJ, equating to 3.5 % of the natural gas

consumption in the industry sector in 2013 or 1.5 % of natural gas used in power generation in the same year.

- On site biogas plants for the mono-digestion of pig slurry are more profitable as compare to centralised plants that co-digest pig slurry and elephant grass when the produced biogas is used to generate electricity.
- For a rural biogas plant for the co-digestion of pig slurry and elephant grass, electricity generation is preferable to the production of biomethane.
- The biomethane resource in the Timoleague region is significant. A biogas plant of approximately 23,000 t/year for the co-digestion of *U.lactuca*, dairy slurry and grass silage can produce 1.19 Mm^3 of methane per year with an energy content of 42.7 TJ. This resource could fuel the equivalent to 1,587 diesel cars or replace the direct fuel consumption of 897 houses.
- A combination of 70% grass silage, 25% *U.lactuca*, 5% dairy slurry based on volatile solids was found to be the optimum mix for biomethane production, reaching a BMP value of 325 LCH_4/kgVS. In continuous digestion this mix reached 89% of the BMP value (288 LCH_4/kgVS).
- The digester performed its best at an OLR of 2 kgVS/m^3/d. When increased to 3 kgVS/m^3/d, the SMY dropped quite steeply reducing the overall efficiency to 44% the BMP value. This was attributed to VFA accumulation.
- The reactor's peristaltic pump and grinder provided for the feeding prove to be inefficient for this purpose. The maceration of the feedstock did not improve the pumpability of the substrate.
- The stirrer vertical paddles configuration helped mixing efficiently the content of the reactor breaking the surface and avoiding scum formation.

- A continuous single stage reactor using a mixture of 94% *laminaria digitata* and 6% *arthrospira platensis* (based on volatile solids) produced an average of 204 $mLCH_4/gVS$ at an OLR of 1 $kgVS/m^3/d$, equating to an energy production of 7.3 kJ/gVS. When the OLR was increased, the SMY started to decrease due to an accumulation of VFAs in the reactor.

- From an energy perspective a two stage reactor can achieve higher yields. A continuous two stage reactor involving hydrogen dark fermentation and methane production using the same mixture of *laminaria digitata* and *arthrospira platensis* effected an energy yield of 9.4 kJ/gVS.

- Dark hydrogen fermentation in two-stage system optimised hydrolysis of the algae mixture, facilitating the methane production and improving the process stability in the second-stage anaerobic reactor.

7.2. Recommendations

It has been demonstrated that biogas production in Mexico has a significant energy potential and could help achieving the renewable energy and greenhouse gas emissions targets. However, some further research is suggested to improve the conditions of a possible biogas industry in Mexico.

- A laboratory analysis of the characterization and seasonal variation of food waste is required. As described in this thesis, the composition of food waste varies depending on the time of the year, location and socio-economic status. Such variations have to be taken into account when estimating the energy potential of a biogas plant given that they have a direct impact on methane yields.
- A strategy regarding source segregation of food waste is necessary to effectively implement a waste management system that helps recovering waste of better quality.

- The design of an economic instrument or incentive to promote the production of biomethane is required. Such instrument should encourage the use of waste streams as a first step.
- The use different grasses other than elephant grass has to be investigated. The characteristics, yields per hectare and biomethane potential have to be analysed in order to estimate the energy potential and the system capacity to process the amount of grass produced.
- The potential use of dairy and beef slurry as a feedstock for anaerobic digestion in centralised biogas plants has to be explored. This industry is growing steadily in Mexico and alongside with pig farming it is responsible for soil, water pollution and the emission of greenhouse gases.

It was also found that the anaerobic co-digestion the green algae *Ulva lactuca* with dairy slurry and grass silage has a great potential in rural coastal areas of Ireland. However, more research is needed in order to improve the performance of the reactor and a more complete analysis of the amount of *U.lactuca* stranded throughout the year and its variation in composition is required. In order to improve the performance of the digester, the following is suggested:

- An equalization tank can help keeping solids in suspension before feeding the reactor, after this, a pump capable of handling high solids and fibrous materials is necessary to pump the substrate into the reactor. To prevent any blockage, it is suggested that the feedstock is macerated before this process.
- The monitoring of trace elements could help indicating if there is a lack of them that could affect the anaerobic process in long term digestion.

An analysis taking into account possible biogas plant locations and capital costs, distances from the substrates sources and costs associated for transport, maintenance and operation are necessary to evaluate the economic feasibility of a coastal biogas plant for the treatment of the feedstock above discussed.

A continuous two-stage system involving hydrolysis through dark fermentation and anaerobic digestion of mixed *laminaria digitata* and *arthrospira platensis* was proved to be feasible. Nevertheless, the TAN levels stayed low throughout the entire continuous experiment, indicating that the hydrolysis or degradation of nitrogen-rich micro-algae was somewhat limited. This was probably due to *Arthrospira platensis* not being prone to degradation given to its recalcitrant cell wall structures. To overcome this, pretreatment of micro-algae and even possibly macro-algae is suggested as this may facilitate the solubilisation and hydrolysis of these substrates.

Appendix A

Supplementary material chapter 3

Table A.1. Sewage sludge generation

WWTP	Technology	Projected inhabitants with service	Volume of sludge (L/d*inh)	kilograms of sludge (kg/d*inh)	Dry solids (kgDS/day)	Volatile solids	Thickened sludge (kg/day)
1	Activated sludge SBR	16,200.00	91,530.00	91,896.12	1,378.44	1,068.29	17,230.52
2	Activated sludge with extended aeration	1,879.20	8,362.44	8,395.89	83.96	65.07	1,049.49
3	Trickling filter	34,344.00	113,335.20	113,788.54	3,129.18	2,425.12	39,114.81
4	Trickling filter	27,990.00	92,367.00	92,736.47	2,550.25	1,976.45	31,878.16
5	Trickling filter	2,264.40	7,472.52	7,502.41	206.32	159.90	2,578.95
6	Septic tank	381.60	248.04	249.03	8.47	6.56	105.84
7	Septic tank	316.80	205.92	206.74	7.03	5.45	87.87
8	Activated sludge	914.40	5,166.36	5,187.03	77.81	60.30	2,204.49
9	Extended aeration	504.00	2,242.80	2,251.77	22.52	17.45	281.47
10	Extended aeration	1,800.00	8,010.00	8,042.04	80.42	62.33	1,005.26
11	Trickling filter	3,600.00	11,880.00	11,927.52	328.01	254.21	4,100.09
12	Activated sludge	5,875.20	33,194.88	33,327.66	499.91	387.43	6,248.94
13	Extended aeration	3,135.60	13,953.42	14,009.23	140.09	108.57	1,751.15
14	Extended aeration	781.20	3,476.34	3,490.25	34.90	27.05	436.28
15	Trickling filter	21,124.80	69,711.84	69,990.69	1,924.74	1,491.68	24,059.30
16	Trickling filter	13,780.80	45,476.64	45,658.55	1,255.61	973.10	15,695.13
17	Trickling filter	13,780.80	45,476.64	45,658.55	1,255.61	973.10	15,695.13
18	Trickling filter	20,282.40	66,931.92	67,199.65	1,847.99	1,432.19	23,099.88
19	Trickling filter	655.20	2,162.16	2,170.81	59.70	46.27	746.22
20	Extended aeration	2,466.00	10,973.70	11,017.59	110.18	85.39	1,377.20
21	Extended aeration	475.20	2,114.64	2,123.10	21.23	16.45	398.08
22	Activated sludge	7,707.60	43,547.94	43,722.13	655.83	508.27	8,197.90
23	Extended aeration	568.80	2,531.16	2,541.28	25.41	19.69	317.66
24	Trickling filter	14,630.40	48,280.32	48,473.44	1,333.02	1,033.09	16,662.75
25	Extended aeration	1,047.60	4,661.82	4,680.47	46.80	36.27	585.06
26	Extended aeration	1,090.80	4,854.06	4,873.48	48.73	37.77	609.18
27	Extended aeration	4,881.60	21,723.12	21,810.01	218.10	169.03	2,726.25
Total		202,478.40	759,890.88	762,930.44	17,350.27	13,446.46	218,243.03

Activated sludge sludge production 5.65 litres/inhabitant/day, DS 1.5% , 77.5% VS from DS

Extended aeration sludge production 4.45 litres/inhabitant/day, DS 1% , 77.5% VS from DS

Trickling filter sludge production 3.3 litres/inhabitant/day, DS 2.75%, 77.5% VS from DS

Septic tank sludge production 0.65 litres/inhabitant/day, DS 3.4%, 77.5% VS from DS

Table A.2. Pig slurry production

Farm	Animal population (heads)			DS per day			VS per day			Pig slurry DS per day (kg)	Pig slurry VS per day (kg)	Pig slurry L per day	Pig slurry kg per day	Pig slurry at 8% DS (kg/day)
	Sows	Growing	Finishers	Sows	Growing	Finishers	Sows	Growing	Finishers					
1	0.0	0.0	4500.0	0.0	0.0	2070.0	0.0	0.0	1687.5	2070.0	1687.5	50400.0	50601.6	25978.5
2	450.0	1500.0	1384.0	225.0	195.0	636.6	202.5	165.0	519.0	1056.6	886.5	32900.8	33032.4	13260.8
3	0.0	563.0	573.0	0.0	73.2	263.6	0.0	61.9	214.9	336.8	276.8	16501.6	16567.6	4226.5
4	0.0	1955.0	0.0	0.0	254.2	0.0	0.0	215.1	0.0	254.2	215.1	8553.1	8587.3	3189.6
5	0.0	0.0	5500.0	0.0	0.0	2530.0	0.0	0.0	2062.5	2530.0	2062.5	61600.0	61846.4	31751.5
6	41.0	80.0	120.0	20.5	10.4	55.2	18.5	8.8	45.0	86.1	72.3	6160.0	6184.6	1080.6
7	77.0	137.0	305.0	38.5	17.8	140.3	34.7	15.1	114.4	196.6	164.1	13461.9	13515.7	2467.5
8	0.0	213.0	0.0	0.0	27.7	0.0	0.0	23.4	0.0	27.7	23.4	931.9	935.6	347.5
9	60.0	150.0	150.0	30.0	19.5	69.0	27.0	16.5	56.3	118.5	99.8	8531.3	8565.4	1487.2
10	40.0	70.0	280.0	20.0	9.1	128.8	18.0	7.7	105.0	157.9	130.7	9966.3	10006.1	1981.6
11	0.0	0.0	4000.0	0.0	0.0	1840.0	0.0	0.0	1500.0	1840.0	1500.0	44800.0	44979.2	23092.0
12	0.0	0.0	100.0	0.0	0.0	46.0	0.0	0.0	37.5	46.0	37.5	2450.0	2459.8	577.3
13	30.0	0.0	0.0	15.0	0.0	0.0	13.5	0.0	0.0	15.0	13.5	2100.0	2108.4	188.3
14	15.0	30.0	75.0	7.5	3.9	34.5	6.8	3.3	28.1	45.9	38.2	3018.8	3030.8	576.0
15	190.0	0.0	0.0	95.0	0.0	0.0	85.5	0.0	0.0	95.0	85.5	13300.0	13353.2	1192.3
16	0.0	320.0	500.0	0.0	41.6	230.0	0.0	35.2	187.5	271.6	222.7	13650.0	13704.6	3408.6
17	120.0	100.0	0.0	60.0	13.0	0.0	54.0	11.0	0.0	73.0	65.0	8837.5	8872.9	916.2
18	0.0	0.0	3600.0	0.0	0.0	1656.0	0.0	0.0	1350.0	1656.0	1350.0	40320.0	40481.3	20782.8
19	1315.0	3941.0	1889.0	657.5	512.3	868.9	591.8	433.5	708.4	2038.8	1733.6	71118.8	71403.3	25586.6
Total										12915.6	10664.6	408601.9	410236.3	162091.2

Sow average weight 200 kg, Sow DS production 0.5 kg/200 kg of live weight and VS production of 0.45kg/200 kg of live weight

Growing pig average weight 12.5 kg, DS production 0.13 kg/12.5 kg of live weight and VS production of 0.11 kg/12.5 kg of live weight

Finisher pig average weight 70 kg, DS production 0.46 kg/70 kg of live weight and VS production of 0.375kg/70kg of live weight

Table A.3. Cost of a biodigester

	Farm	Animal population (heads)			DS per day (kg)			Pig slurry DS per year (t)	Grass DS (t/year)	CAPEX*	Cost* ($US/DS)	OPEX ($US/tDS/year)
		Sows	Growing	Finishers	Sows	Growing	Finishers					
	1	0	0	4000	0	0	1840.0	945.3	872.0	676572.0	372.3	56.6
	2	0	0	100	0	0	46					
	3	30	0	0	15.0	0	0					
	4	15	30	75	7.5	3.3	28.1					
	5	190	0	0	95	0	0					
	6	0	320	500	0	41.6	230					
	7	120	100	0	60	13.0	0					
	8	211	129	0	94.95	14.2	0					
Plant 1	9	0	778	0	0	101.1	0					
	10	0	0	4500	0	0	2070	1356.9	1239.5	861938.0	332.0	48.1
	11	450	1500	1384	225.0	195	636.6					
	12	0	563	573	0	73.2	263.6					
Plant 2	13	0	1955	0	0	254.2						
	14	0	0	5500	0	0	2530	1137.6	1033.2	748636.0	344.9	50.7
	15	41	80	120	20.5	10.4	55.2					
	16	77	137	305	38.5	17.8	140.3					
	17	0	213	0	0	27.7	0					
	18	60	150	150	30	19.5	69					
Plant 3	19	40	70	280	20	9.1	128.8					
											349.7	51.8

Sow average weight 200 kg, Sow TS production 0.5 kg/200 kg of live weight. VS production 0.45 kg/200 kg live weight

Growing pig average weight 12.5 kg, DS production 0.11 kg/12.5 kg of live weight. VS production 0.11kg/12.5 kg live weight

Finisher pig average weight 70 kg, DS production 0.46 kg/70 kg of live weight. VS production 0.375 kg/70 kg live weight

Without including haulling truck cost

Table A.4. Scenario 1 NPV Calculation

$$NPV = -CAPEX + \sum_{t=0}^{N} \frac{R_t}{(1+i)^t}$$

CAPEX= $US 8,913,605.13
Rt= *Income* – OPEX
Income= $US 2,314,647.32
OPEX= $US 982,096.21
Rt= $US 1,332,551.11
i= 10%
t= Time in years
N= 20
NPV = - $US 8,913,605.13 + $US 11,344,758.76
= $US 2,431,153.63

Year	$(1+i)^t$	Rt/$(1+i)^t$
1	1.10	1,211,410.10
2	1.21	1,101,281.91
3	1.33	1,001,165.37
4	1.46	910,150.34
5	1.61	827,409.40
6	1.77	752,190.36
7	1.95	683,809.42
8	2.14	621,644.93
9	2.36	565,131.75
10	2.59	513,756.14
11	2.85	467,051.03
12	3.14	424,591.85
13	3.45	385,992.59
14	3.80	350,902.35
15	4.18	319,002.14
16	4.59	290,001.95
17	5.05	263,638.13
18	5.56	239,671.03
19	6.12	217,882.75
20	6.73	198,075.23
	Total	11,344,758.76

Table A.5. Scenario 2 NPV Calculation

$$NPV = -CAPEX + \sum_{t=0}^{N} \frac{R_t}{(1+i)^t}$$

CAPEX= $US 9,153,503.01
Rt= *Income* – OPEX
Income= $US 2,004,854.68
OPEX= $US 998,390.16
Rt= $US 1,006,464.52
i= 10%
t= Time in years
N= 20
NPV = - $US 9,153,503.01 + $US 8,568,599.80
= - $US 584,903.22

Year	$(1+i)^t$	Rt/$(1+i)^t$
1	1.10	914,967.74
2	1.21	831,788.86
3	1.33	756,171.69
4	1.46	687,428.81
5	1.61	624,935.28
6	1.77	568,122.98
7	1.95	516,475.44
8	2.14	469,523.13
9	2.36	426,839.20
10	2.59	388,035.64
11	2.85	352,759.67
12	3.14	320,690.61
13	3.45	291,536.92
14	3.80	265,033.56
15	4.18	240,939.60
16	4.59	219,036.00
17	5.05	199,123.64
18	5.56	181,021.49
19	6.12	164,564.99
20	6.73	149,604.54
	Total	8,568,599.80

Table A.6. LCOE calculation scenario 1

Assuming a biomethane price of $US 11.318071/GJ ≈ $US 11.32/GJ

CAPEX= $US 8,913,605.13
Rt= *Income* – OPEX
Income= $US 2,029,084.94
OPEX= $US 982,096.21
Rt= $US 1,332,551.11
i= 10%
t= Time in years
N= 20
NPV = - $US 8,913,605.13 + $US 8,913,605.13
= $US 0

Year	$(1+i)^t$	Rt/$(1+i)^t$
1	1.10	951,807.92
2	1.21	865,279.93
3	1.33	786,618.12
4	1.46	715,107.38
5	1.61	650,097.62
6	1.77	590,997.83
7	1.95	537,270.76
8	2.14	488,427.96
9	2.36	444,025.42
10	2.59	403,659.47
11	2.85	366,963.16
12	3.14	333,602.87
13	3.45	303,275.34
14	3.80	275,704.85
15	4.18	250,640.77
16	4.59	227,855.25
17	5.05	207,141.14
18	5.56	188,310.12
19	6.12	171,191.02
20	6.73	155,628.20
		8,913,605.13

Table A.7. LCOE calculation scenario 2

Assuming a biomethane price of $US 14.372224/GJ ≈ $US 14.38/GJ

CAPEX= $US 9,153,503.01
Rt= *Income* – OPEX
Income= $US 2,073,557.19
OPEX= $US 998,390.16
Rt= $US 1,075,167.03
i= 10%
t= Time in years
N= 20
NPV = - $US 9,153,503.01 + 9,153,503.01
= $US 0

Year	$(1+i)^t$	Rt/$(1+i)^t$
1	1.10	977,424.57
2	1.21	888,567.79
3	1.33	807,788.90
4	1.46	734,353.55
5	1.61	667,594.13
6	1.77	606,903.76
7	1.95	551,730.69
8	2.14	501,573.35
9	2.36	455,975.78
10	2.59	414,523.43
11	2.85	376,839.48
12	3.14	342,581.35
13	3.45	311,437.59
14	3.80	283,125.08
15	4.18	257,386.44
16	4.59	233,987.67
17	5.05	212,716.06
18	5.56	193,378.24
19	6.12	175,798.40
20	6.73	159,816.73
		9,153,503.01

Appendix B

Supplementary material chapter 4

Table B.1. Pig manure characteristics and methane yields

Type of pig	% Moisture	DS (%FM)	VS (%DS)	Methane yield m^3/tVS
Gestating sow manure	71	28.9	74.7	282.4
Nursery and weaned Manure	76.4	23.6	76.6	328.7
Growing manure	73	26.9	75.2	263.5
Mixed manure	73.8	26.1	76.9	354.7

Table B.2. Manure excreted by pig type per day

Animal type	Average weight of animal (kg)	DS (kg/day)	VS (kg/day)	kgVS per 1000 kg live weight
Nursery pig	12.5	0.13	0.11	8.8
Grow-finisher pig	70	0.46	0.375	5.36
Gestating sow	200	0.50	0.45	2.25
Lactacting sow*	192	1.2	1	5.21
Boar	200	0.38	0.34	1.7
* Includes contribution of nursing pigs			Average	4.66

Table B.3. Pig population and average weight of Conkal municipality

Type of Pig	Number of heads	Average weight (kg)*	Live weight per type of pig (kg)
Sow	1,234	200	246,800
Growing	6,025	12.5	75,313
Finisher	17,487	70	1,224,090
	24,746		1,546,203

*ASAE average weight

Table B.4. Farm slurry and VS generation of scenario 3

	Animal population (heads)					VS per day (kg)			Plant VS per day	Slurry	VS per day	Slurry per year
	Farm	Sows	Growing	Finishers	UAP	Sows	Growing	Finishers	(kg)	(m^3/day)	(t)	(m^3)
Plant 1	1	0	0	4,000	2,800			1,500.0	2,157	44.8	787	39,016
	2	0	0	100	70			37.5		2.5		
	3	30	0	0	60	13.5				2.1		
	4	15	30	75	86	6.8	3.3	28.1		3.0		
	5	190	0	0	380	85.5				13.3		
	6	0	320	500	390		35.2	187.5		13.7		
	7	120	100	0	253	54.0	11.0			8.8		
	8	211	129	0	438	95.0	14.2			15.3		
	9	0	778	0	97		85.6			3.4		
Plant 2	10	0	0	4,500	3,150			1,687.5	3,066	50.4	1,119	39,550
	11	450	1,500	1,384	2,056	202.5	165.0	519.0		32.9		
	12	0	563	573	471		61.9	214.9		16.5		
	13	0	1,955	0	244		215.1			8.6		
Plant 3	14	0	0	5,500	3,850			2,062.5	2,553	61.6	932	36,738
	15	41	80	120	176	18.5	8.8	45.0		6.2		
	16	77	137	305	385	34.7	15.1	114.4		13.5		
	17	0	213	0	27		23.4			0.9		
	18	60	150	150	244	27.0	16.5	56.3		8.5		
	19	40	70	280	285	18.0	7.7	105.0		10.0		

Sow average weight 200 kg, Sow VS production 0.45 kg/200 kg of live weight

Growing pig average weight 12.5 kg, VS production 0.11 kg/12.5 kg of live weight

Finisher pig average weight 70 kg, VS production 0.375 kg/70 kg of live weight

For farms greater than UAP > 2001, slurry production is 0.016 m^3/UAP

For farms smaller than UAP< 681, slurry production is 0.035 m^3/UAP

Table B.5. Electricity consumption and costs

Scenario		Farm	Mixer	Irrigation pump	Pit pump	Upgrading plant	Total consumption	Consumption cost	Maximum demand	Maximum demand cost	Eelectricity cost
		(kWh/year)	(kWh/year)	(kWh/year)	(kWh/year)	(kWh/year)	(kWh/year)	($US/year)	(kW/month)	($US/year)	($US/year)
Scenario 1		109,500	61,320				170,820	14,468	47	6,903	21,372
Scenario 2		109,500	105,120	47,052			261,672	22,164	75	11,032	33,196
Scenario 3	Plant 1		227,760	70,360	10,804		308,924	26,166	62	9,087	35,253
	Plant 2		245,280	99,331	10,804		355,415	30,104	76	11,100	41,204
	Plant 3		219,000	82,776	10,804		312,580	26,476	66	9,746	36,222
	Biomethane Plant		692,040	252,467	32,412	766,500	1,743,419	147,668	291	42,785	190,453

Farm electricity consumption is 300 kWh per day

Mixers working 100% of the time. Scenario 1, 7kw. Scenario 2, 12 kw.For plant 1 mixer has a capacity of 26 kW; plant 2, 28 kW and plant 3,25 kW

Irrigation consumption is given at 0.7087 kWh/ha. The pump works 8 hours per day 365 days

Pit pump has a capacity of 3.7 kW working 8 hours per day 365 days

Upgrading plant consumption is given at 0.25 kWh/m^3

Price of kWh is given at $US 0.0847/kWh. Maximum demand cost $US 12.24/kW

Table B.6. NPV calculation of scenario 1

	Analysis without grants				Analysis including grants			
Years	CAPEX	R_t	$(1+i)^t$	$Rt/(1+i)^t$	CAPEX	R_t	$(1+i)^t$	$Rt/(1+i)^t$
1	148,331	16,804	1.08	15,632	77,813	8,815	1.08	8,200
2		16,804	1.16	14,541		8,815	1.16	7,628
3		16,804	1.24	13,527		8,815	1.24	7,096
4		16,804	1.34	12,583		8,815	1.34	6,601
5		16,804	1.44	11,705		8,815	1.44	6,140
6		16,804	1.54	10,888		8,815	1.54	5,712
7		16,804	1.66	10,129		8,815	1.66	5,313
8		16,804	1.78	9,422		8,815	1.78	4,943
9		16,804	1.92	8,765		8,815	1.92	4,598
10		16,804	2.06	8,153		8,815	2.06	4,277
11		16,804	2.22	7,584		8,815	2.22	3,979
12		16,804	2.38	7,055		8,815	2.38	3,701
13		16,804	2.56	6,563		8,815	2.56	3,443
14		16,804	2.75	6,105		8,815	2.75	3,203
15		16,804	2.96	5,679		8,815	2.96	2,979
OPEX per year*	27,963				27,963			
Revenues per year**	44,767				36,778			
Cumulative cash flow				148,331				77,813
NPV	0				0			

Prices in $US

*Considering no increases in maintenance rates

** A tariff of $ 0.115 kWh_e for the no grant condition and $ 0.096 kWh_e for the grant condition. $ 0.0081/kWh_e for the use of the grid

Table B.7. NPV Calculation for scenario 2

	Analysis without grants				Analysis including grants			
Years	CAPEX	R_t	$(1+i)^t$	$Rt/(1+i)^t$	CAPEX	R_t	$(1+i)^t$	$R_t/(1+i)^t$
1	378,457	42,874	1.08	39,883	258,969	29,338	1.08	27,291
2		42,874	1.16	37,101		29,338	1.16	25,387
3		42,874	1.24	34,512		29,338	1.24	23,616
4		42,874	1.34	32,104		29,338	1.34	21,968
5		42,874	1.44	29,865		29,338	1.44	20,436
6		42,874	1.54	27,781		29,338	1.54	19,010
7		42,874	1.66	25,843		29,338	1.66	17,684
8		42,874	1.78	24,040		29,338	1.78	16,450
9		42,874	1.92	22,363		29,338	1.92	15,302
10		42,874	2.06	20,802		29,338	2.06	14,235
11		42,874	2.22	19,351		29,338	2.22	13,241
12		42,874	2.38	18,001		29,338	2.38	12,318
13		42,874	2.56	16,745		29,338	2.56	11,458
14		42,874	2.75	15,577		29,338	2.75	10,659
15		42,874	2.96	14,490		29,338	2.96	9,915
OPEX per year*	63,558				63,558			
Revenues per year**	106,432				92,896			
Cumulative cash flow				378,457				258,969
NPV	0				0			

Prices in $US

*Considering no increases in maintenance rates

** A tariff of $ 0.129 kWh_e for the no grant condition and $ 0.114 kWh_e for the grant condition. $ 0.0081/kWhe for the use of the grid

Table B.8. NPV calculations for scenario 3 electricity and biomethane

	Plant 1				Plant 2				Plant 3				Biomethane			
Years	CAPEX	R_t	(1+i)t	$R_t/(1+i)^t$	CAPEX	R_t	$(1+i)^t$	$R_t/(1+i)^t$	CAPEX	R_t	$(1+i)^t$	$R_t/(1+i)^t$	CAPEX	R_t	$(1+i)^t$	$R_t/(1+i)^t$
1	812,072	101,250	1.08	94,186	997,438	122,215	1.08	113,689	884,136	109,380	1.08	101,749	2,079,647	263,253	1.08	244,886
2		101,250	1.16	87,615		122,215	1.16	105,757		109,380	1.16	94,650		263,253	1.16	227,801
3		101,250	1.24	81,502		122,215	1.24	98,379		109,380	1.24	88,046		263,253	1.24	211,908
4		101,250	1.34	75,816		122,215	1.34	91,515		109,380	1.34	81,904		263,253	1.34	197,124
5		101,250	1.44	70,527		122,215	1.44	85,130		109,380	1.44	76,189		263,253	1.44	183,371
6		101,250	1.54	65,606		122,215	1.54	79,191		109,380	1.54	70,874		263,253	1.54	170,578
7		-34,250	1.66	-20,644		-12,785	1.66	-7,706		-25,620	1.66	-15,443		-141,747	1.66	-85,439
8		101,250	1.78	56,771		122,215	1.78	68,526		109,380	1.78	61,329		263,253	1.78	147,606
9		101,250	1.92	52,810		122,215	1.92	63,746		109,380	1.92	57,051		263,253	1.92	137,308
10		101,250	2.06	49,126		122,215	2.06	59,298		109,380	2.06	53,070		263,253	2.06	127,729
11		101,250	2.22	45,698		122,215	2.22	55,161		109,380	2.22	49,368		263,253	2.22	118,817
12		101,250	2.38	42,510		122,215	2.38	51,313		109,380	2.38	45,924		263,253	2.38	110,528
13		101,250	2.56	39,544		122,215	2.56	47,733		109,380	2.56	42,720		263,253	2.56	102,816
14		101,250	2.75	36,785		122,215	2.75	44,403		109,380	2.75	39,739		263,253	2.75	95,643
15		101,250	2.96	34,219		122,215	2.96	41,305		109,380	2.96	36,967		263,253	2.96	88,970
OPEX per year*	136,250				153,600				132,734				500,687			
Revenues per year**	237,500				275,816				242,113				763,939			
Cumulative cash flow				812,072				944,200				884,136				2,079,647
NPV	0				0				0				0			

Prices in $US

*Considering no increases in maintenance rates , electricity and fuels

**Plant 1 electricity tariff $0.195/kWhe. Plant 2 $0.161/kWhe.Plant 3 $0.169/kWhe. Biomethane $11.60/GJ

Appendix C

Supplementary material chapter 6

Table C.1. AFB modified medium (composition in 1 litre)

Glucose	20g
Tryptone	3g
Yeast extract	1g
NaCl	3g
K_2HPO_4	2.5g
$MgCl_2$	0.1g
$FeCl_2$	0.1g
L-Cysteine	0.5g
Glutamatic acid	0.1g
Ascorbic acid	0.025g
Riboflavin	0.025g
Citric acid monohydrate	0.01g
Folic acid	0.1g
p-Aminobenzoic acid	0.01g
Creatine	0.025g
$MnCl_2$	0.01g
$ZnCl_2$	0.01g
H_3BO_3	0.01g
$CaCl_2$	0.01g
Na_2MoO_4	0.01g
$CoCl_2\,6H_2O$	0.2g
$AlK\,(SO_4)_2$	0.01g
$NiCl_2\,6H_2O$	0.01g

Table C.2. Reactor configuration

The two-stage fermentation system comprised of a hydrolysis reactor and a methane reactor. The reactors had a total volume of 5 L with an internal diameter of 0.15 m and a height of 0.4 m. A third system, a single-stage reactor with the same dimensions as the methane reactor of the two-stage system was also employed. A temperature controller unit was installed to maintain a constant temperature in the reactors at mesophilic conditions. An outer heating blanket supplied the heat. A wet gas metre recorded gas flow automatically. Collected biogas was stored in a gas bag for compositional analysis. Mixing was provided by a stirring mechanism, consisting of a vertical shaft with height adjustable paddles at the upper and lower end. A variable speed motor drove the shaft. The shaft of the stirrer was surrounded by a top mounted pipe, which sealed the top of the reactor with the rotating stirrer. The reactors were equipped with a submerged pipe on top of the reactor to prevent gas leakage and oxygen entry during the feeding process.