

**Financial Viability of Microalgal Biodiesel Production –
A Case Study in Southern Ontario, Canada**

by

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August 2018

A thesis submitted to McGill University in partial fulfillment of the
requirement for the degree of Master of Science in Renewable Resources

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Abstract

Biofuels are an alternative to fossil fuels with potential to help in the fight against climate change. With transportation accounting for 14% of global greenhouse gas emissions and most of the world's transportation energy coming from gasoline and diesel, biodiesel has the potential to replace fossil fuels, particularly in the road transportation sector. However not all biodiesel is created equal, due to the large number of potential feedstocks and conversion processes that may be followed to produce biodiesel. A suitable biodiesel must be environmentally and socially sustainable, and the manufacturing process must be financially cost-effective, before it can be considered as a replacement for conventional diesel fuel. This thesis evaluated the financial viability of a novel biodiesel source, produced from microalgae feedstock grown in wastewater with CO₂ additions from flue gas. The case study was based on Canada-specific data. The optimal geographic location for producing microalgae feedstock in a municipal wastewater treatment facility was determined using climatic data on insolation and temperature effects, whilst also taking into account the location of existing wastewater treatment plants and biodiesel infrastructure in southern Ontario, Canada. Methods to grow the microalgae, whether in open ponds or photobioreactors, were determined from the literature review. Characteristics of the wastewater were derived from anonymous data provided by three wastewater treatment plants in Alberta, Canada. This was compared to microalgae growth requirements, to determine if sufficient nutrients and CO₂ would be provided by the wastewater and flue gas to support microalgae growth in the case study, based on strains that were recommended for cold climates. Harvesting, dewatering and conversion pathways were determined from literature review and consultation with experts, and the most cost-effective options were retained for analysis. Financial metrics were estimated from

literature, public and private data, and from consultation with industry professionals. The net present value (NPV), internal rate of return (IRR), breakeven date and investment multiple of the flue gas and wastewater co-utilization (FWC) system were then derived. A five case multi-scenario analysis was then conducted to identify the impact on the profitability of the proposed microalgae biodiesel production system in southern Ontario, Canada, with a $\pm 25\%$ revenues or costs estimation error. Then, single-variable sensitivity analysis was conducted to assess the impact on the profitability of the system of a percentage change in key revenue or cost variables, notably: the percentage of nitrogen (N) and phosphorus (P) removal savings passed on to a private company, the price of diesel, the Ontario energy price, the quantity of biodiesel and methane produced, as well as capital expenses (CapEx) and operational expenses (OpEx). In conclusion, the FWC systems was found to be profitable with a base case NPV of roughly \$256.5M, an IRR of 38.77%, a 3.8X investment multiple and a 3-year payback period. Given that these results are primarily dependent on the percentage of N and P removal savings passed on to a private company, this type of project was found not to be suited for private businesses and investors but rather governments and wastewater treatment plant operators who would incur 100% of wastewater treatment savings generated.

Résumé

Les biocarburants, qui représentent une alternative aux combustibles fossiles, ont le potentiel de participer à la lutte contre le changement climatique. C'est dans le secteur du transport, qui représente 14% des émissions de gaz à effet de serre, que le biodiesel a le plus grand potentiel. En effet, il offre la possibilité d'y remplacer les combustibles fossiles, puisque l'énergie des transports provient principalement de l'essence et du diesel. Cependant, tous les biodiesels ne se valent pas, compte tenu de la panoplie de matières premières et de processus de conversion qui permettent de produire du biodiesel. Pour être adéquat, un biodiesel doit être durable sur le plan environnemental et social. Le processus de fabrication doit aussi être rentable pour pouvoir être considéré comme une solution de remplacement pertinente au diesel. La présente thèse mesure la viabilité financière d'une nouvelle source de biodiesel produite à partir de microalgues cultivées dans des effluents avec un ajout en CO₂ à partir de gaz de combustion. Cette étude de cas se fonde sur des données propres au Canada. La position géographique optimale pour la production des microalgues dans l'usine de traitement d'eau d'une municipalité a été établie à partir de données concernant l'effet de l'ensoleillement et de la température, tout en prenant acte de l'emplacement d'usines de traitement d'eau et d'infrastructures dédiées au biodiesel en Ontario, Canada. Les méthodes pour cultiver la microalgue, dans des bassins à ciel ouvert ou dans des photobioréacteurs ont été déterminés sur la base de la littérature existante. Les propriétés des effluents ont été tirées de données anonymes provenant de trois usines de traitement des effluents en Alberta, Canada. Ces données ont été comparées avec les exigences de croissance des microalgues afin d'établir si la quantité de nutriments et de CO₂ des effluents et des gaz de combustion serait suffisante

pour répondre aux besoins des microalgues dans cette étude, tout en tenant compte de la contrainte du climat froid. Les méthodes de collecte, d'assèchement et de transformation ont été déterminées sur la base de la littérature existante et de consultations avec des experts. Les options les plus rentables ont été retenues pour l'analyse. Les paramètres financiers ont été estimées à partir de la littérature, de données privées et publiques, et des consultations avec des professionnels de l'industrie. La valeur actuelle nette (VAN), le taux de rendement interne (TRI), le seuil de rentabilité et le multiple d'investissement du système de co-utilisation de gaz de combustion et d'effluents (CGF) en ont ensuite été tirés. Une analyse de multiples scénarios a été effectuée afin de cerner l'impact sur la rentabilité du système de production de biodiesel à partir de microalgues au sud de l'Ontario, Canada. L'estimation de l'erreur concernant les revenus ou les coûts est $\pm 25\%$. Une analyse de sensibilité à variable simple a ensuite été effectuée pour évaluer l'effet d'un changement de pourcentage des variables des principaux revenus ou coûts. Notamment le pourcentage d'épargne concernant la suppression du nitrogène (N) et du phosphore (P) en évitant le coût de traitement par une compagnie privée, le prix du diesel, le prix de l'énergie en Ontario, la quantité de biodiesel et de méthane produite ainsi que les dépenses en capital (DeCap) et les dépenses de fonctionnement (DeFon). Pour conclure le système de CGF s'est révélé profitable dans le scénario de base avec une VAN d'environ 256.5M\$, un TRI de 38.77%, un multiple d'investissement de 3.8X et une période de remboursement de trois ans. Puisque ces résultats dépendent principalement aux gains générés par la suppression de N et P, ce type de projet ne convient pas aux entreprises et investisseurs privés, mais plutôt aux gouvernements ainsi qu'aux usines de traitements des eaux qui profiteraient de 100% des gains générés par le traitement des effluents.

Preface

This master's project was conducted under the supervision of Dr. Joann K. Whalen at the Macdonald campus of McGill University. The objective of this project was to assess the financial viability of microalgal biodiesel production in Canada by conducting a case study of southern Ontario based on existing literature. These case studies are beneficial for evaluating the long-term prospects of a given technology and are essential in determining the most effective way for Canada to achieve its greenhouse gas emission reduction targets.

This thesis is composed of 3 chapters preceded by an introduction that provides context, the problem statement, and the overall objectives of the thesis. Chapter 1 is a literature review in which I take a detailed look at related papers, discuss the current body of literature on the subject of biodiesel production from microalgae and the current research gap that this thesis will try to address. Then, Chapter 2 reviews the methodology used to build the model used to determine the financial viability of 3rd generation biodiesel production in Canada, the results of which are presented and discussed in Chapter 3: Results & Discussion. The thesis then concludes with a summary of key findings and recommendations for further research.

Contribution of Authors

This research is funded by BioFuelNet Canada, a network focusing on the development of advanced biofuels and a member of the Networks of Centers of Excellence of Canada program. The candidate wrote the literature review in chapter 1 and undertook all of the original research in chapters 2 & 3, including data collection, analysis and interpretation. Professor Whalen provided financial support, advisory guidance on the research, and editorial supervision. Professor Smith provided technical and editorial comments.

Acknowledgements

I would like to thank my supervisor, Dr. Joann K Whalen, for giving me the opportunity to conduct this research under her supervision. Her constant support, mentorship, guidance and patience throughout my master's degree were not only central to the development of this thesis but also to my betterment as a young adult.

I'm also lucky enough to have had Dr. Donald L Smith and Prof. Robert Cairns as co-supervisors for my thesis. Their incredible help in the early stages of my research and vast networks have been essential in determining the scope and content of my thesis.

A huge thanks goes to everyone in the Natural Resource Sciences department, notably Dr. Benoit Cote for all of your guidance and administrative help and Dr Sebastien P. Faucher for organizing all of those memorable graduate seminars.

In addition, I could not have completed this work without the help of numerous experts from the BioFuelNet. I would like to thank BioFuelNet for helping me compete at the 2016 MIT Energy conference, present a poster at the 2016 Advanced Biofuels Symposium and present at the 2016 Energy Information Agency's conference.

Special thanks go to my fellow McGill Macdonald Campus graduate students, notably Mitalie Makhani and Monica Soria Baledon for their endless support and encouragement throughout this period.

I also would like to thank my family and friends for their love, patience and support throughout this entire process. This thesis is dedicated to my grandmother Marie-France Dominci who lost the fight to cancer on May 2nd 2018.

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Introduction

In October 2016, Canada ratified the Paris climate agreement and communicated its target to reduce GHG emissions by 30% below 2005 levels by 2030, a reduction of approximately 523 Mt of annual CO₂ equivalent emissions. At the time the country's existing policies and measures were insufficient to reach this target (Figure 1). However the Trudeau government has since announced the “Pan-Canadian Framework on Clean Growth and Climate Change” that describes economy-wide measures to implement carbon pricing schemes, plans to phase out traditional coal power plants and develop cleaner fuels (Environment and Climate Change Canada, 2016b). This push for cleaner fuels builds upon the previous government's Renewable Fuel Regulations (RFR) which mandates that at least 5% of gasoline and 2% of heating oils and diesel be from a renewable source (EPA, 2012). These regulations, alongside the Paris agreement, highlight Canada's continued commitment to expanding the production and use of biofuels such as biodiesel and ethanol.

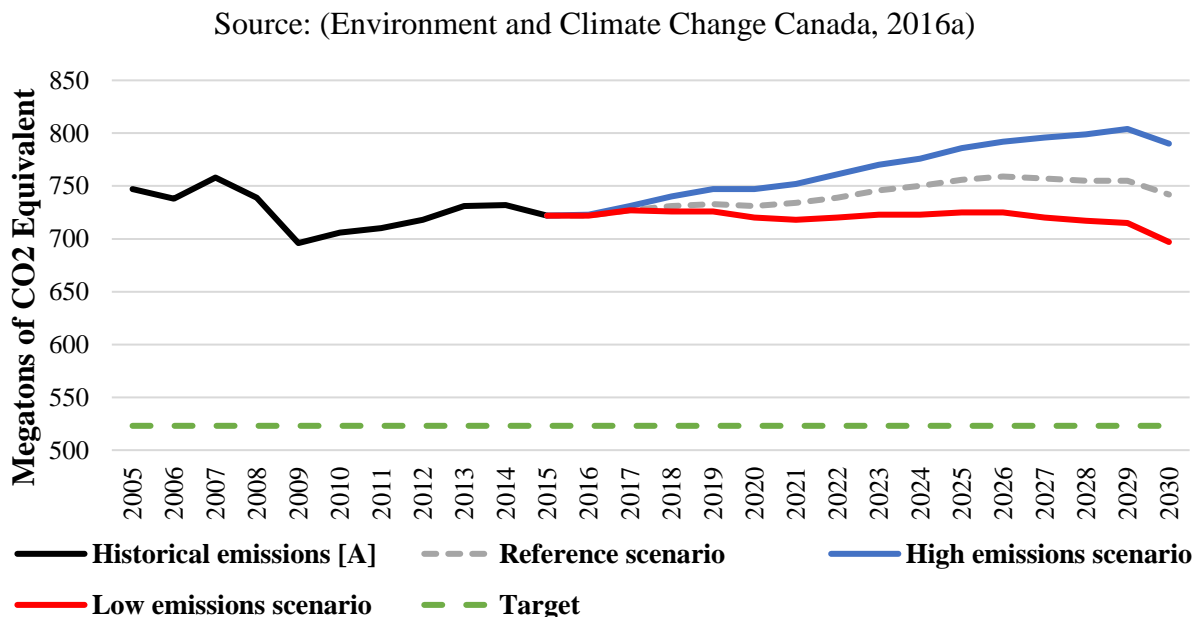


Figure 1: Canadian GHG historical emissions and projections to 2030 with policies and measures as of November 2016

Microalgae can contribute to Canada's efforts to increase biofuel production because they do not require arable land to grow, thus avoiding the "Food vs Fuel" concerns, and have significantly higher yields than other biofuel feedstocks (Najafi, Ghobadian, & Yusaf, 2011). Generally, lipids are extracted from microalgae and upgraded into biodiesel, a renewable fuel with relatively high energy density and energy use efficiency. However, the major barrier to widespread production of microalgal biodiesel is the high cost of production (Mallick, Bagchi, Koley, & Singh, 2016). Other challenges include: selecting the right strain to cultivate, where and when to grow the algae, nutrient and CO₂ sourcing, growth and harvesting techniques, and finally fuel extraction and refining (Hannon, Gimpel, Tran, Rasala, & Mayfield, 2010). There is limited information available to make the most cost-effective decisions for each of these required steps in cultivating microalgae for biodiesel. Some researchers have proposed to use municipal wastewater treatment ponds as the growth medium for microalgae, which would provide nutrients, and the water could be enriched in CO₂ to support microalgae growth. Therefore, the purpose of this thesis is to evaluate the financial viability of Canadian 3rd generation biodiesel produced from microalgae grown in wastewater, relying on the wastewater to provide nutrients and flue gas as a source of dissolved CO₂.

Chapter 1: Literature review

Since the 1900s, greenhouse gas (GHG) emissions have increased exponentially (CDIAC, 2014), so much so that atmospheric CO₂ concentrations reached 400 parts per million (ppm) in March 2015 (National Oceanic and Atmospheric Administration, 2016). Scientists and governments agree that atmospheric GHG concentrations must stabilize at a level below 400 ppm to avoid global warming by 2°C above pre-industrial levels by 2100 (Centre & Office, 2013). This 2°C figure represents the start of “catastrophic climate change” where flooding and severe weather caused by storms, droughts and hurricanes occur more often and with more intensity (Pachauri, Meyer, & Team, 2014).

Despite a decreasing energy intensity of the world economy, the world’s energy consumption has been steadily increasing for decades (Enerdata, 2016). Greater energy consumption is attributed predominantly to non-OECD countries (outside the Organization for Economic Cooperation and Development) and is fueled by the strong economic growth and increasing population in the non-OECD countries (DOE/EIA, 2016). Despite a continued and rapid growth of the renewable energy sector, it only accounted for 2.8% of global primary energy consumption in 2015. Fossil fuels still account for 86% of the world’s energy consumption, however the gradual shift away from coal in favor of natural gas has partially constrained the GHG emissions attributed to fossil fuel burning (Markewitz, Marx, Schreiber, & Zapp, 2013).

The carbon intensity of the economy, which is the ratio of GHG emissions produced to gross domestic product, has significantly decreased during the latter half of the 20th century. However, carbon intensity remains stagnant at around 76 KtCO₂ per billion 2005 US\$ since the beginning of the 21st century (Shift Project Data Portal, 2015). This may indicate that we have reached the

limits of carbon savings in the fossil fuel-based economy. Alternatives to fossil fuels will play a crucial role in our transition towards a more sustainable world energy mix (Fulton, Lynd, Körner, Greene, & Tonachel, 2015) and likely lead to economic benefits and increase energy security in OECD (Cherp, Vinichenko, Jewell, Suzuki, & Antal, 2017) and non-OECD countries (Sharma & Singh, 2017).

Currently renewable energy sources range from wind and solar energy to biofuels and hydrogen fuel cells, all of which contribute to a low carbon economy. However, these technologies require significant advances in energy storage and energy density before they will supplant fossil fuels: renewable energy use (wind, solar etc.) at a large scale requires the development efficient grid scale storage (Kies, Schyska, & Von Bremen, 2016), which has yet to be developed. In addition, despite the recent advances in commercial battery technology and the development of hydrogen fuel cells, these solutions fall far short of conventional fuels like diesel and gasoline in terms of energy density (Today in Energy, 2013), thus limiting their use in transportation.

Due to their similar chemical composition, some biofuels share near identical characteristics with their fossil fuel counterparts (AFDC, 2014). This, combined with the potential for GHG emission reductions with biofuels, makes them an interesting alternative to fossil fuels. However not all biofuels are created equal. Depending on the feedstock and conversion process used, the resulting biofuels may generate much lower carbon savings than expected (Pachauri et al., 2014). For this reason, it is important to define the three major types of biofuels:

1st Generation Biofuels: First generation biofuels are produced directly from the edible portions of food crops, grown on agricultural land, by extracting the oils for use in biodiesel or producing bioethanol through starch/sugar fermentation. Crops such as wheat, sugar beet and corn are the

most widely used feedstocks for bioethanol while canola oil has proved a very effective crop for use in biodiesel (Technology, 2008).

2nd Generation Biofuels: Second generation biofuels are produced from non-food crops grown in soil such as non-marketable wood residues (i.e. branches, bark, dead wood, etc.), food crop residues (e.g. corn stalks) and purpose-grown bioenergy crops (e.g. jatropha) (Technology, 2008). Many of these bioenergy crops can be grown on marginal agricultural land, however in practice most are grown on agricultural land or deforested land (Walmsley, Bailis, & Klein, 2016).

3rd Generation Biofuels: Third generation biofuels are also produced from non-edible biomass, predominantly microalgae, but grow in water rather than soil. Until recently, microalgae biofuels were categorized as a 2nd generation biofuel, however they were differentiated based on their capacity to generate much higher yields with potentially lower inputs (Technology, 2008).

Despite being well known technology and easy to produce, 1st and 2nd generation biofuels have yet to capture a significant share of the energy market (Markewitz et al., 2013). This is mostly because of their higher cost (Alternative Fuels Data Center, 2015), but is also due to the “food vs fuel” debate (de Gorter & Drabik, 2016). The “Food versus Fuel” argument applies to both 1st generation biofuels and 2nd generation biofuels derived from purpose bioenergy crops since they use arable land (Walmsley et al., 2016). Despite proof of the contrary, policy makers are afraid to allocate arable land for biofuel feedstock production for fear it will increase the price of food. Paradoxically, the policy of non-OECD countries that restrict food exports and incentivize imports when faced with increased food prices is self-defeating, resulting in both higher world food grain prices and volatility (de Gorter & Drabik, 2016).

Third generation biofuels circumvent these issues by growing the feedstock on non-arable land and thus do not compete with food production (Singh, Nigam, & Murphy, 2011). Microalgae also are more efficient at capturing CO₂ (Sayre, 2010) and have a higher energy density per unit of land area than other feedstocks (Najafi et al., 2011). Moreover, microalgae has the potential to produce biomass rapidly, and many strains may double their cell counts within hours (Singh et al., 2011). As hundreds of thousands of strains of microalgae exist or could be developed (Guiry, 2012), there is ample species diversity to identify the most promising biofuel production strains that match their feedstock requirements.

1.1 Microalgae: potential for commercialization as a feedstock for biodiesel production

The development and commercialization of microalgae biofuels must not only consider environmental and social impacts of the technology but also its economic implications. To do so, a wide range of papers have conducted a techno-economic analysis to assess the feasibility of various traditional microalgae to biodiesel conversion pathways (Table 1).

These studies show the high variability of biodiesel fuel cost estimates derived from differing assumptions on cultivation systems, products, oil contents, production capacities and conversion technologies to name just a few parameters. In addition, these assessments were conducted by extrapolating laboratory-scale experiments which often do not represent commercial or even pilot scale operations. Overall however, the consensus seems to be that due to high initial and production costs, the biodiesel produced using these pathways cannot compete with conventional fossil-fuel diesel currently priced at around US\$0.85 / L (Energy Information Administration, 2018).

Table 1: Techno-economic results from literature for the production of biodiesel

Culture System	Biomass Productivity	Oil Content (%)	Capacity	Conversion technology	Biodiesel Selling price	Reference
Solar lit PBR			500 ha	Solvent Extraction	US\$21.72 / L	(Amer, Adhikari, & Pellegrino, 2011)
Open Pond			500 ha		US\$3.55 / L	(Amer et al., 2011)
Open Pond	25 g/m ² /day	20% – 50%	333.3 ha	Hydro-Treating	US\$1.68 / L	(Delrue et al., 2012)
PBR	0.65 – 1.9 kg/m ³ /d	20% – 50%		Hydro-Treating	US\$2.8 / L	(Delrue et al., 2012)
Raceway + PBR		20% – 50%		Hydro-Treating	US\$2.69 / L	(Delrue et al., 2012)
Open Pond	1.25 kg/m ³ /day	25%			US\$3.91 / L	(Richardson, Johnson, & Outlaw, 2012)
PBR	25 g/m ² /day	25%			US\$9.89 / L	(Richardson et al., 2012)
Open Pond	60 g/m ² /day	50%		Oil Extraction and Transesterification	US\$0.42 / L	(Nagarajan, Chou, Cao, Wu, & Zhou, 2013)
Open Pond	30 g/m ² /day	50%			US\$0.97 / L	(Nagarajan et al., 2013)

PBR = Photobioreactor

Wastewater contains water, nitrogen, phosphorus, organic carbon and other nutrients which makes it suitable for microalgae growth. By providing these nutrients, the wastewater is treated by the microalgae as they grow, resulting in the phytoremediation of the water. Such an integrated system would derive a wide range of benefits such as a reduced wastewater treatment and biomass production costs, due to the biological nutrient removal by algal cultivation, replacing expensive and non-sustainable fertilizers, and reducing the energy usage and environmental impact of substituted wastewater treatment processes (Ren et al., 2017). Furthermore, flue gas emissions from nearby industries or from a combined heat and power generator can be bubbled up into the wastewater to increase its carbon content and facilitate algal growth (Ferrell & Sarisky-Reed, 2010). Revenues could also be derived from wastewater treatment, biodiesel production, carbon credits and other byproducts, and would further improve the economic soundness of a flue gas and wastewater co-utilization (FWC) system (Mata, Mendes, Caetano, & Martins, 2014).

Research has primarily been focused on technological issues which focus on biomass productivity, lipid content improvement and nutrient removal. The results of these studies have a large degree of variability primarily induced from the fact that different microalgae strains were grown in different wastewater conditions. These results have been summarized in a review paper by Zhou et al. (Zhou et al., 2014). As a result of this focus, many technological issues related to wastewater-based algal biofuel production have been resolved. However economic studies and related data are incomplete and outdated. The techno-economic viability of an integrated system combining wastewater treatment, CO₂ capture, microalgae cultivation, and biomass conversion to biodiesel hasn't been discussed much, especially based on pilot-scale data. Examples of some of these studies have been summarized in Table 2.

The studies listed in Table 2 tend to show that even with the cost of nutrients and CO₂ being obliterated, the economics of a flue gas and wastewater co-utilization (FWC) system are unfavorable and highly dependent on the fuel market value (Doshi, Pascoe, Cogle, & Rainey, 2016). However, these studies fail to account for the cost savings generated at the wastewater treatment plant into their financial analysis of the project. They point towards other key contributory factors associated with the financial viability of the biofuels being produced. These include:

- Plant location and size
- Algae cultivation method used
- Feedwater quality and optimal growth conditions
- The algae strain selected and algal lipid productivity
- Ancillary processes, such as biomass harvesting, drying and converting techniques

These factors reflect the complex and multi-faceted nature of this research topic which not only encompasses engineering and microbiological aspects of the technology but also the ambient conditions, environmental impacts and financial constraints of building a sustainable biofuel production facility.

Table 2: Combined CO₂ and nutrient cost analysis examples since 2011

Reference	Primary system facets/variables considered, or study objectives	Key correlation (s)/output (s) Costs
(Acién et al., 2012; Gabriel Acién Fernandez et al., 2012)	All key fundamental factors contributing to cost for OP and PBR	Costs of installation at small and large-scale; cost benefit of zero-cost CO ₂ and nutrient feed
(Delrue et al., 2013; Slade & Bauen, 2013)	<ul style="list-style-type: none"> ○ Energy and carbon accounting; EIA ○ Water and nutrient recycling; excess nutrient demand met by supplementation 	<ul style="list-style-type: none"> ○ OP vs PBR; cost benefit of zero-cost CO₂ and nutrient feed. ○ OP vs PBR at different productivities and lipid content.
(Orfield, Keoleian, & Love, 2014)	Co-location of multiple flue gas and wastewater sources at national scale.	Potential production capacity and selling price of biofuel in US with reference to nutrient and CO ₂ availability and infrastructure requirements.
(Ramos Tercero, Domenicali, & Bertucco, 2014)	Covered 100 ha pond	Full CapEx and OpEx determination
(Kang et al., 2015)	OP; Algal species; light wavelength (blue, green, red, white)	Impact of (a) zero-cost nutrient and CO ₂ supply, and (b) light wavelength on biofuel selling price.
(Quinn & Davis, 2015)	Algal growth; lipid productivity; scalability; environmental impact	Summary of published lipid yields, biofuel costs. Impact of productivity on fuel cost, and biomass processing technology on GHG emissions.
(Rizwan, Lee, & Gani, 2015)	Optimization based on maximum algal yield and maximum operating margin	Impact of (a) 90% nutrient recycling and (b) zero cost CO ₂ supply Impact
(Hernández-Calderón et al., 2016)	Multiple flue gas and wastewater sources at regional scale.	Impact of regional decentralization of processing facilities on cost
(Rezvani et al., 2016)	PBR technology (OP & PBR); Power plant technology (3 types); irradiation intensity; CO ₂ fixation rate.	BESP of the generated electricity (\$/MW) and CO ₂ avoidance cost (\$/t) against the selling price of the algal biomass (\$/t)
(Xin et al., 2016)	All key fundamental factors contributing to cost (incl carbon credit) for WwTW centrate feed.	Cost sensitivity to algal concentration, biofuel yield, chemicals cost, PBR CAPEX, nutrient feed, harvesting technology, and productivity

PBR = Photobioreactor, OP = Open Pond, CapEx = Capital Expenditures, OpEx = Operating expenses

Within the scope of these studies, there has been many different approaches to quantifying the cost benefit of a FWC system. The most common of these involve determining the breakeven sell price of either the biomass or the derived biofuel for overall cost neutrality (Acién et al., 2012; Gabriel Acien Fernandez et al., 2012; Kang et al., 2015; Quinn & Davis, 2015; Rezvani et al., 2016). There has also been a number of studies looking at the energy balance of the system, more specifically at the energy return of investment, which have recently been reviewed by Quinn et al. (Quinn & Davis, 2015). More recently still, studies have provided an energy balance for algal open ponds used for wastewater treatment (Mehrabadi, Craggs, & Farid, 2015), as well as a comparison of the CO₂ balance of biofuel production from algae grown in an open pond to that of conventional diesel production (Manganaro & Lawal, 2016).

However, these studies often do not provide outputs in terms of the breakeven selling price and therefore are hard to compare with other studies. Furthermore, the use of carbon accounting seems to be complicated by the range of energy to carbon conversion factors used, which can range from 0.26 kg CO₂ / kWh or lower for “carbon neutral” conditions (Quinn & Davis, 2015) to 0.95 kg CO₂ / kWh for a coal-fired power station (Rickman, Pellegrino, Hock, Shaw, & Freeman, 2013). More recent studies have started to incorporate environmental impacts into their cost models with varying degrees of success in order to try and offer a more robust breakeven selling price for the biodiesel produced (R. E. Davis et al., 2014; Delrue et al., 2012; Xin et al., 2016).

For the purposes of this thesis however, utilizing a breakeven selling price of the biodiesel produced would be a flawed metric for determining the viability of a FWC system since it would give the false impression that the financial returns are primarily generated from the sale of biodiesel rather than alternative revenue streams such as wastewater treatment. Therefore, the financial analysis of the FWC system determined in this thesis will rely on more standard project

finance metrics such as the net present value (NPV), internal rate of return (IRR), breakeven year and investment multiple which are all better at evaluating the financial sustainability of the whole project.

1.2 Summary and research questions

The preceding literature review highlights some of the numerous traditional pathways that can be utilized to grow and convert algal biomass to biodiesel with varying degrees of success and resulting in estimated biodiesel selling prices to range from US\$0.42 / L to US\$21.72 / L. Given this, traditional pathways do not seem to be able to compete with conventional diesel currently priced at US\$0.85 / L. A further look at existing literature shows that merging algal biomass production with wastewater treatment and carbon capture from flue gas emissions could not only reduce the cost of biomass production, but also help generate other sources of revenues. However, given researcher's focus on technological issues, there is little information on the financial viability of these FWC systems with no studies looking at their viability in a Canadian context.

It is clear then that there remains a fundamental gap in the knowledge of the economics of such a system in a Canadian context. I can therefore set about defining the key parameters of the growth system envisaged in this thesis to subsequently model the flue gas and wastewater co-utilization (FWC) system and evaluate its financial viability in Canada. To do so, I must answer the following series of questions derived from the literature review:

1. Where in Canada should the case study be located to provide optimal insolation and temperatures for the growth of microalgae?
2. What method should be used to grow the microalgae?

3. Are sufficient nutrients and CO₂ provided to the microalgae by the wastewater to ensure their optimal growth?
4. What strain of algae is best suited to grow under these growth conditions?
5. How would the algal biomass be harvested and dried?
6. How would the algal biomass be converted to biodiesel?

In Chapter 2, I will use these questions to guide the selection of parameters relevant to model the proposed microalgae biodiesel production system. Then, I will use this information to undertake a financial analysis of my case study, a hypothetical microalgae biodiesel production system operating in southwestern Ontario, Canada.

Chapter 2: Methodology

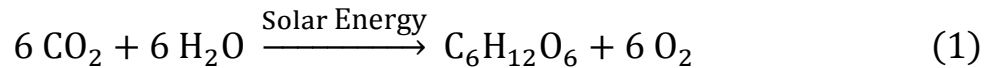
2.1 Geographic determination: Where in Canada should the case study be located to provide optimal insolation and temperatures for the growth of microalgae?

Not all regions are created equal when it comes to growing microalgae for use in biofuels production. The amount of solar radiation (i.e. insolation or irradiance), water & nutrient availability, and seasonal variations in temperature are amongst the most important parameters when determining the optimal geographic location to grow microalgae (Banerjee & Ramaswamy, 2017).

2.1.1 Insolation

2.1.1.1 The effect of insolation

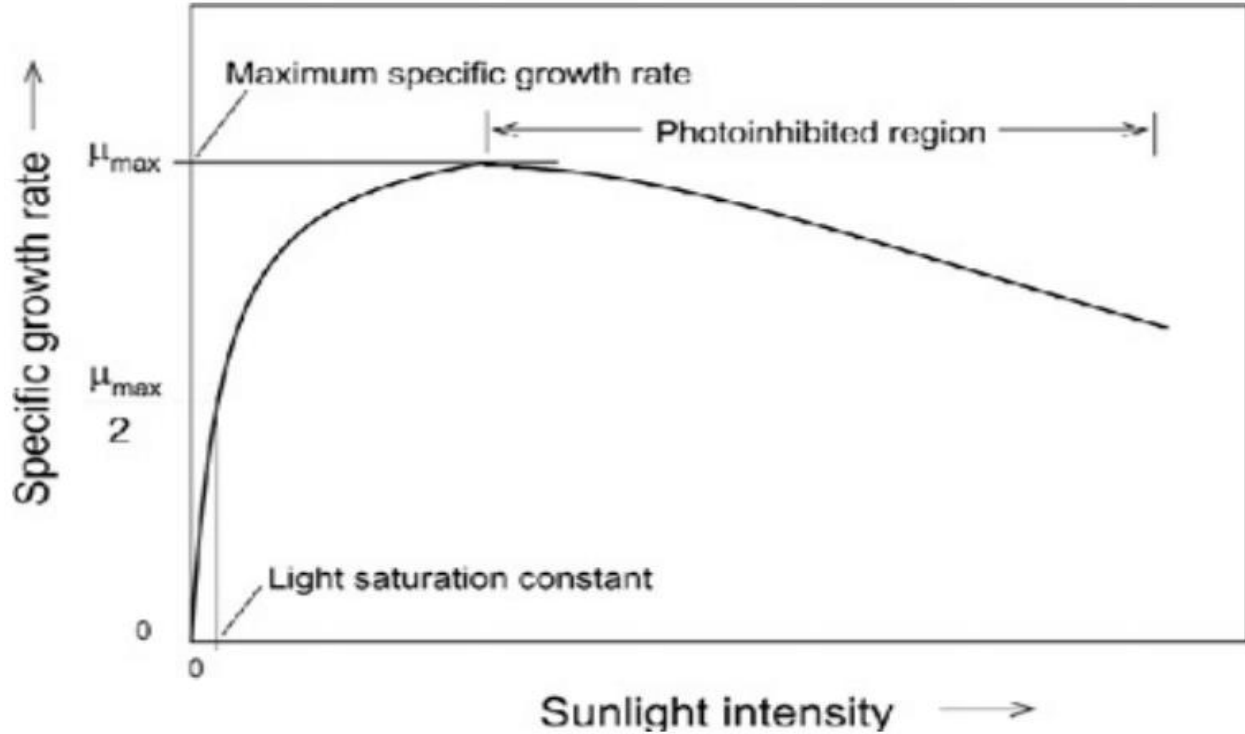
Microalgae obtain energy through photosynthesis, which converts carbon dioxide and water into glucose and oxygen when they are exposed to light through solar energy (Equation 1).



Since microalgae cannot store solar energy for later use, light must be supplied on a continuous basis for photosynthesis to occur. For this reason, microalgae specific growth rates are directly related to sunlight intensity (Figure 2). The relationship shows that microalgae will rapidly achieve their maximum specific growth rate as the intensity of solar radiation increases up to a certain point, the determinants of which aren't well known, beyond which additional sunlight causes light stress (Demmig-Adams & Adams, 2003). This process is known as photoinhibition and is the result of excess photons inactivating or degrading the photosystem II reaction center required for

photosynthesis (Demmig-Adams & Adams, 2003; Nikolaou, Hartmann, Sciandra, Chachuat, & Bernard, 2016). This process is often reversible and does not cause a long-term impact on the microalgae's growth rate (Camacho Rubio et al., 2004).

Figure 2: Effect of light intensity on the specific growth rate of microalgae



Source: (Chisti, 2008)

The light saturation constant of a species of microalgae is defined as the intensity of light at which the specific biomass growth rate is equal to half of its maximum value. It is commonly measured in $\mu E/m^2/s$, otherwise known as photosynthetically active radiation (PAR), where E represents the energy in one mole of photons and is otherwise known as an Einstein. This unit can in turn can be converted to W/m^2 , $kWh/m^2/day$, Lux (lx) and Foot Candles (fc) using the following approximate conversion rates for sunlight of:

$$1 W/m^2 \approx 0.024 kWh/m^2/day \approx 4.57 \mu E/m^2/s \approx 0.249 lx \approx 23.16 fc \quad (2)$$

These conversion rates vary by light source (Table 3; Sager & McFarlane, 1997). Although the light saturation constant varies per species and overtime through a process known as photoacclimation, studies have identified a wide range of microalgae species with light saturation constants between $150 \mu\text{E}/\text{m}^2/\text{s}$ and $250 \mu\text{E}/\text{m}^2/\text{s}$ (Chisti, 2008; Talbot, Thébault, Dauta, & de la Noüe, 1991). Therefore, the region that I select for our model must maximize the number of months where the mean daily insolation is above $250 \mu\text{E}/\text{m}^2/\text{s} \approx 54.7 \text{ W}/\text{m}^2 \approx 1.31 \text{ kWh}/\text{m}^2/\text{day}$.

Table 3: Approximate conversion values for radiation of 400 – 700 nm

Multiply by Indicated Value

Radiation Source	$\mu\text{E}/\text{m}^2/\text{s}$ to W/m^2	W/m^2 to $\mu\text{E}/\text{m}^2/\text{s}$	$\mu\text{E}/\text{m}^2/\text{s}$ to lux	Lux to $\mu\text{E}/\text{m}^2/\text{s}$	W/m^2 to lux	Lux to W/m^2
Sunlight	0.219	4.57	54	0.019	0.249	4.02
Cool White Fluorescent	0.218	4.59	74	0.014	0.341	2.93
Plant Growth Fluorescent	0.208	4.80	33	0.030	0.158	6.34
High-pressure Sodium	0.201	4.98	82	0.012	0.408	2.45
High-pressure metal Halide	0.218	4.59	71	0.014	0.328	3.05
Low-pressure Sodium	0.203	4.92	106	0.009	0.521	1.92
Incandescent 100W tungsten Halogen	0.200	5.00	50	0.020	0.251	3.99

Source: (Sager & McFarlane, 1997)

2.1.1.2 Solar radiation mapping

Natural Resources Canada provides mean daily insolation per month for over 3500 municipalities in Canada (Table 4). Of the 13 Canadian provinces and territories, only New Brunswick, Nova Scotia, Ontario, and Prince Edward Island provide a mean daily insolation of

$\geq 250 \mu\text{E}/\text{m}^2/\text{s}$ for eleven months of the year. Hence, those four provinces are the most promising locations for microalgae production since they receive sufficient light, year-round, for optimal microalgae growth. However, significant variation in insolation occurs within any particular province. The mean daily insolation for all municipalities in the candidate regions during the months of January and February (Figures 3 & 4), shows that the southernmost region of Ontario receives the most solar radiation in Canada during winter months when solar radiation is lowest.

Table 4: Mean daily insolation by province per month

Mean daily insolation in $\mu\text{E}/\text{m}^2/\text{s}$

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Annual
AB	192	371	649	913	1088	1174	1184	984	663	410	211	146	667
BC	163	308	564	837	1037	1118	1157	983	689	391	187	129	632
MB	236	417	681	943	1094	1148	1160	952	654	398	226	175	675
NB	269	447	656	815	967	1064	1045	920	684	430	255	205	647
NL	204	361	585	772	918	1016	990	843	621	367	205	153	587
NT	44	163	458	830	1087	1190	1055	744	433	184	60	23	524
NS	266	431	627	776	957	1068	1062	944	709	456	264	205	648
NU	28	125	416	845	1149	1191	1003	659	351	136	37	12	498
ON	283	464	684	864	1038	1138	1148	962	705	452	252	217	685
PEI	259	433	650	809	960	1067	1050	922	689	431	250	195	644
QC	269	452	683	853	995	1080	1059	902	650	405	238	203	650
SK	229	414	677	935	1105	1179	1190	987	686	428	234	171	688
YT	54	185	464	822	1020	1100	967	755	470	208	74	30	514

Source: (Natural Resources Canada, 2017)

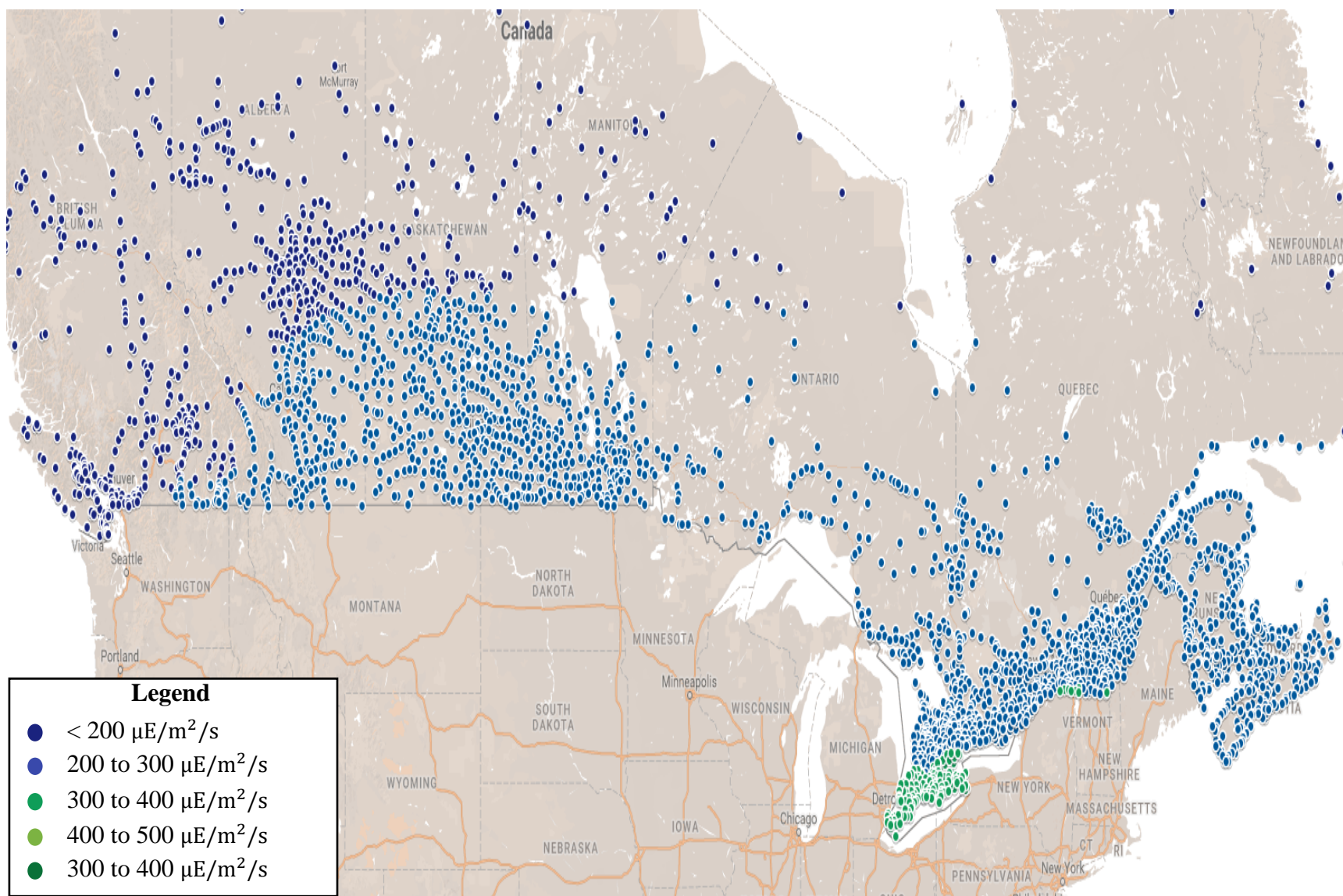


Figure 3: Map of mean daily insolation of Canadian municipalities - January

Source: (Natural Resources Canada, 2017)

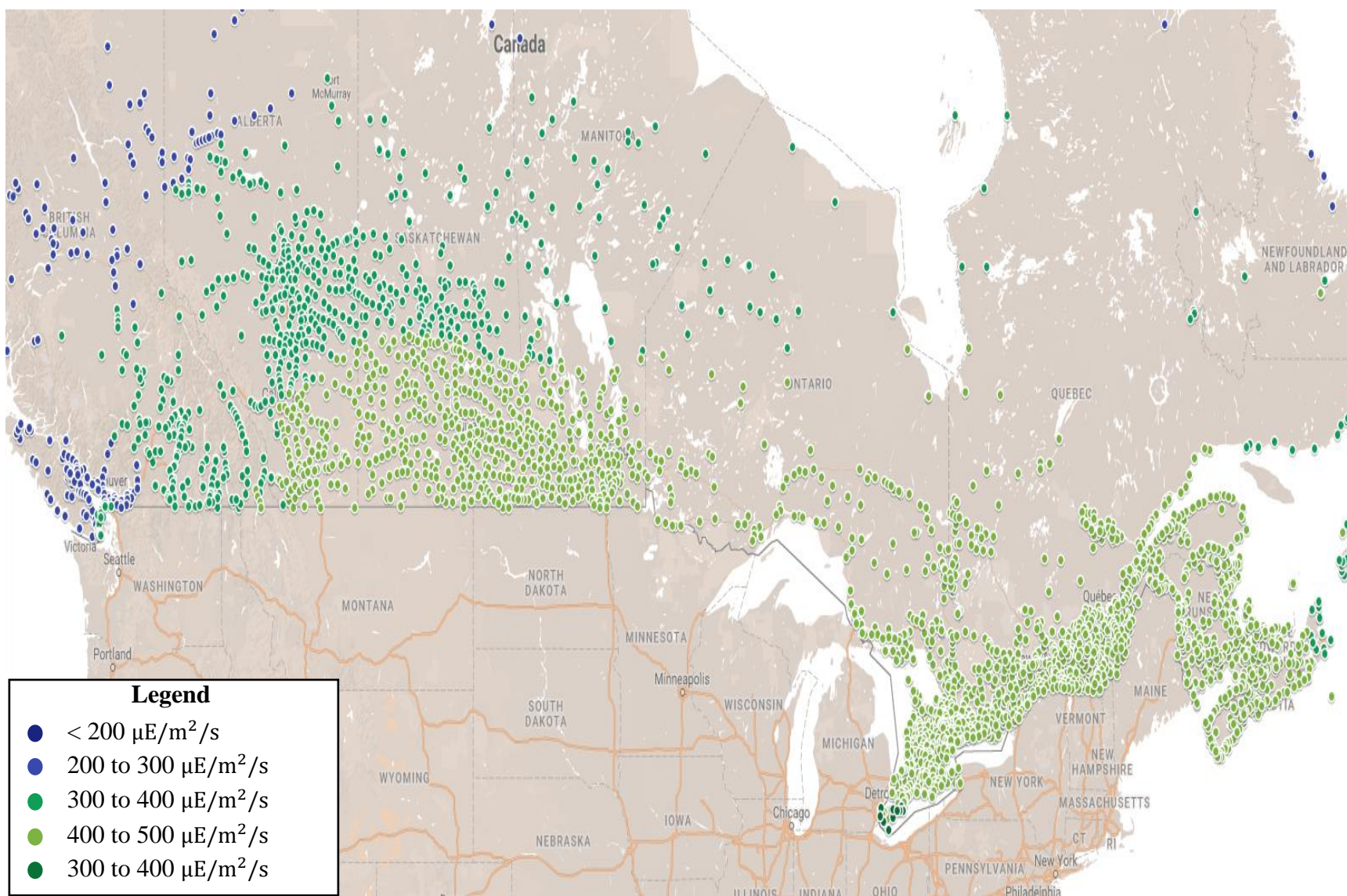


Figure 4: Map of mean daily insolation of Canadian municipalities - February

Source: (Natural Resources Canada, 2017)

2.1.2 Temperature

2.1.2.1 The effect of temperature

Temperature significantly impacts the growth rate and chemical composition of microalgae with higher temperatures generally resulting in higher lipid and carbohydrate contents that in turn are essential for increased biofuel yields (Converti, Casazza, Ortiz, Perego, & Del Borghi, 2009; Renaud, Thinh, Lambrinidis, & Parry, 2002). There is however an optimal temperature, which varies with species, after which an increase in temperature will result in a lower photosynthetic efficiency due to some enzymes being denatured and therefore temporarily or irreversibly damaged by heat (Talbot et al., 1991). Inversely, despite having some degree of adaptation and acclimation, if water temperatures drop below a specie's chilling stress temperature it will see its photosynthetic efficiency significantly decrease and will ultimately cease once the water freezes (ÖQUIST, 1983). For these reasons it is essential that the microalgae be maintained at a somewhat constant and elevated temperature throughout the year so that a species with the right optimal temperature may be selected.

2.1.2.2 Temperature mapping

Unfortunately, there isn't a map tracking average water temperature across Canada throughout the year. However, since water temperature is most prominently affected by solar radiation and air temperature (Fondriest Environmental, 2014), I can estimate using these parameters the areas in Canada where water temperatures will be the highest during the winter, thus helping to avoid a reduction in microalgae growth due to temperatures dropping below our selected specie's chilling stress point. In addition to the pacific coast of Canada, I can identify the southern tip of Ontario as a region with one of the highest average temperatures in winter (Figure 5). Having also already

identified this region as having the most solar radiation in Canada, I can conclude that southern Ontario also likely has the highest average water temperature in Canada throughout the year.

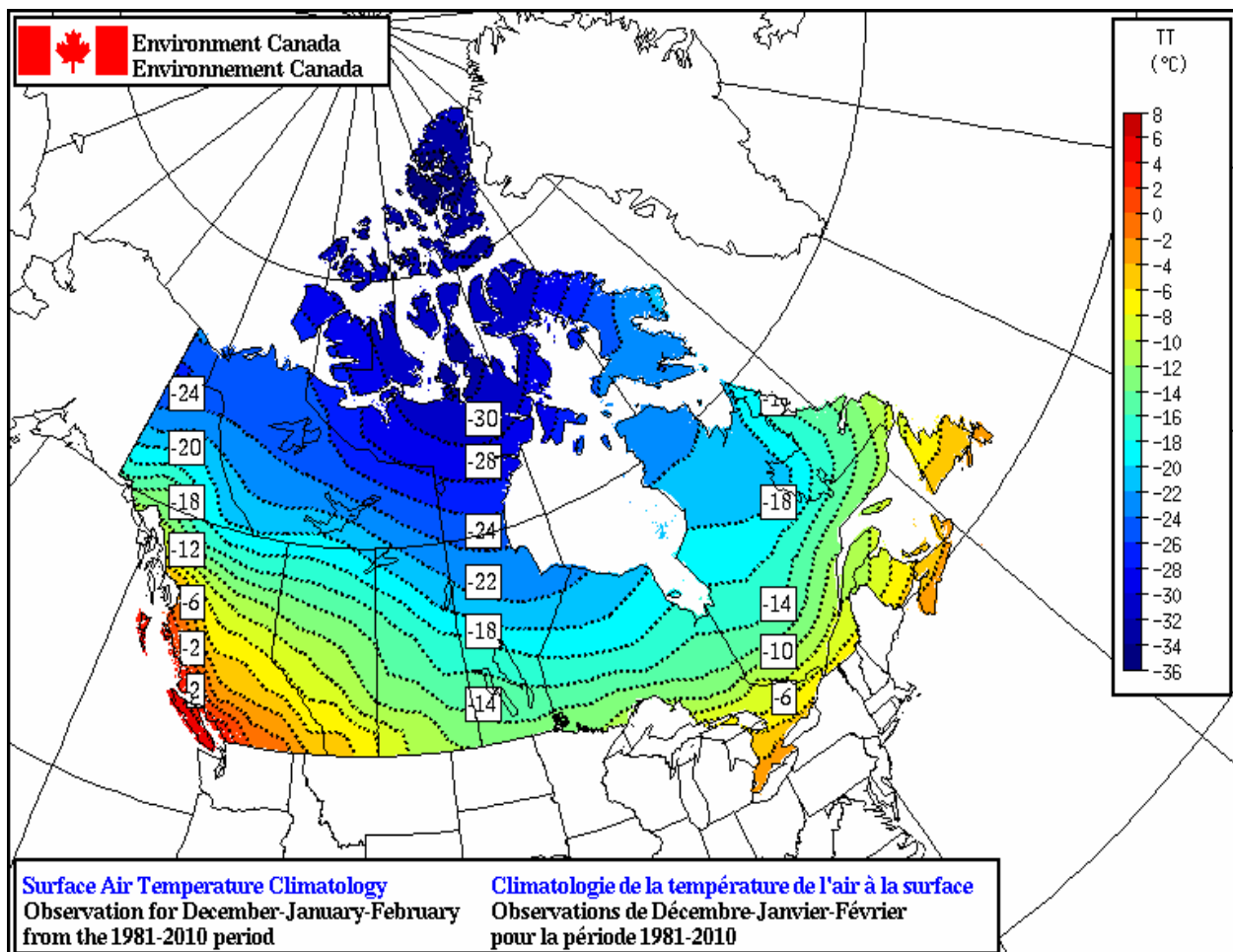


Figure 5: Average temperature map of Canada – December to February (Winter)

Source: (Environment Canada, 2017)

In addition, looking at anonymized data from two wastewater treatment plants in southern Alberta showing the seasonal variation in wastewater temperature (Figure 6), I can see that temperatures tend vary between 12°C and 19°C with a yearly average just above 15°C. Given that southern Ontario consistently receives more sunlight and has a higher air temperature than southern Alberta (Figures 3, 4 & 5), I can safely assume that water temperatures in the region will follow a similar pattern, albeit at a slightly higher temperature, than those of southern Alberta.

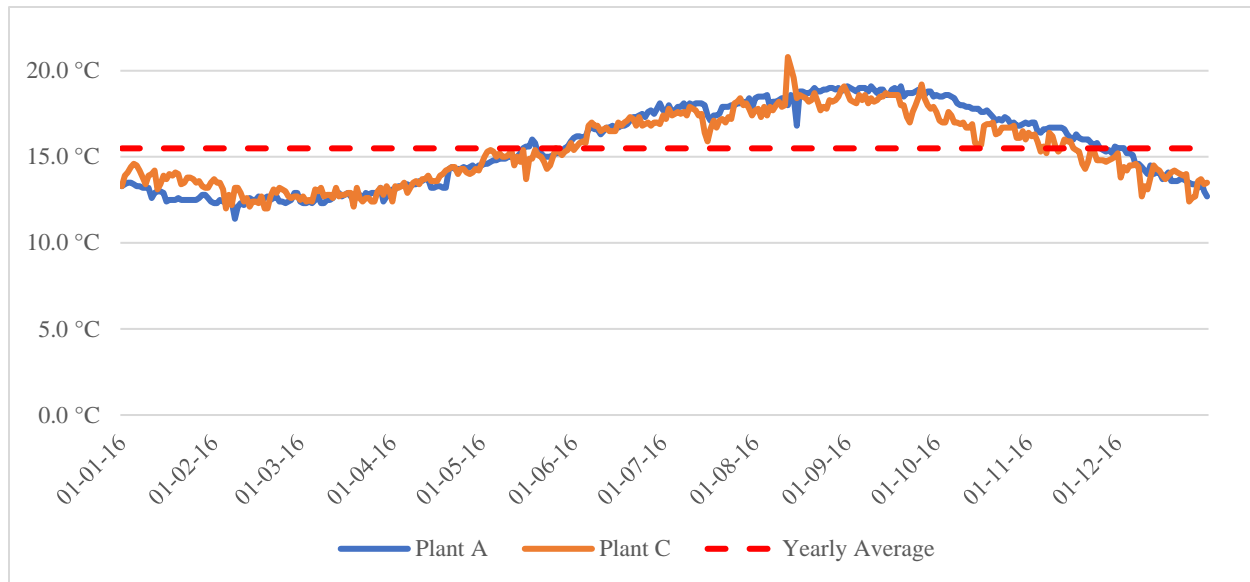


Figure 6: Daily recorded wastewater temperatures for two treatment plants in southern Alberta

Source: (Anonymous, 2017)

2.1.3 Supply chain considerations and geographic region selection

When determining the geographic region of our study, I must consider the both the location of our major production inputs (i.e. wastewater) and the destination of our production outputs (i.e. biorefineries) to help minimize transportation and other logistical costs. Mapping out the location of most wastewater treatment plants and biorefineries across in and around southern Ontario, I can identify a cluster of facilities near the U.S and Canadian border (Figure 7). Given the presence of these facilities and the region's yearly sunlight and temperature, I can determine that southern Ontario is an optimal location in Canada for 3rd generation biofuel production and therefore will be the region of interest for this study.

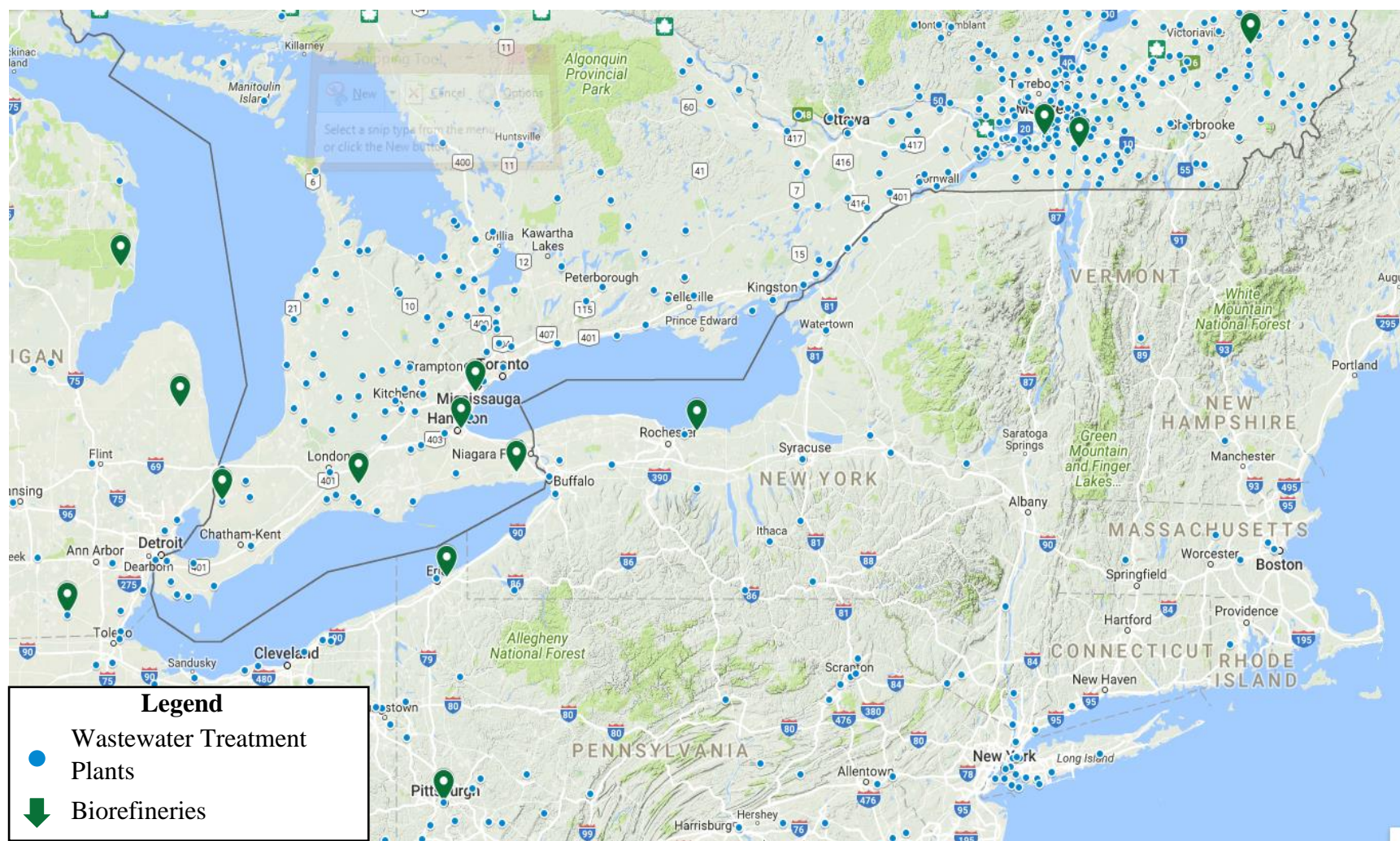


Figure 7: Map of major wastewater treatment plants and biorefineries near southern Ontario

Source: (American Biogas Council, 2016; Biodiesel Magazine, 2016; Canada Water and Wastewater Association, 2001; Canadian Government, 2011; Greenfuels, 2016; Sustainable america, 2016)

2.2 Microalgal cultivation method selection: What method should be used to grow the microalgae?

Open ponds and photobioreactors (PBRs), defined below, are the two primary methods used for microalgae cultivation in wastewater (Tolieng, Prasirtsak, Sitdhipol, Thongchul, & Tanasupawat, 2017).

Open Ponds: In these systems, wastewater is left to settle in basins 0.2 to 1 m deep and be treated by various organisms that breakdown the pollutants for use as nutrients. A variation of the open pond design is known as High Rate Algae Ponds (HRAPs) where a mechanism, often a paddle wheel, is used to mix the water and generate a hydraulic flow (Verbyla, von Sperling, & Maiga, 2017). Microalgae grown in these ponds provide oxygen for and absorb carbon dioxide from the respiration of aerobic bacteria. Open ponds have been widely implemented in wastewater treatment plants around the world due to their low operational cost, simple design and decent performance in treating wastewater (Alamgir, Khan, & Shaukat, 2016). However, these systems suffer from a number of disadvantages including a high contamination risk where other organisms outcompete the microalgae for nutrients, limited control of the growth environment (pH, temperature and nutrients) and evaporation (Austin, 2010).

Photobioreactors: PBRs are closed systems where all essential nutrients, water, light and CO₂ must be introduced into the system and managed in order for the microalgae to grow (Oilgae, 2010). Given the additional control of all parameters affecting microalgae growth, PBRs can maintain optimal growth conditions, resulting in a higher yield of biomass (Xin et al., 2016), and can be used to grow and harvest biomass on a continuous basis (Acien et al., 2012). However, this

control comes at the cost of significantly higher capital and operational expenditures (Power Plant CCS, 2010).

2.2.1 Discussion and selection

The cultivation system for microalgae production will be a significant factor in determining the system-wide economic viability. Most information on the cost of establishing and operating open ponds vs. PBRs is derived from lab-scale or pilot-scale trials spanning a brief period of time, the results of which are extrapolated to commercial sized facilities. In the southwest United States, the financial viability of PBRs was substantially lower than for open ponds where, in their base case, the average production cost of lipids for PBRs and open ponds was \$8.35 \$/L and \$4.68 \$/L (\$31.61 \$/gal and \$12.73 \$/gal) respectively (Richardson et al., 2012). Similarly, Norsker et al. estimated in 2011 that biomass production costs for open ponds and flat-panel PBRs was an estimated \$7.25 CAD/Kg and \$8.73 CAD/Kg (4.95 €/Kg and 5.96 €/Kg) (Norsker, Barbosa, Vermuë, & Wijffels, 2011). The general consensus is that open ponds can cultivate biomass at a 30% to 60% lower price than PBRs. As a result, I assume that the microalgae will be grown in an open-pond system attached to the wastewater treatment plant.

2.3 Nutrient and CO₂ sourcing: Are sufficient nutrients and CO₂ provided to the microalgae by the wastewater to ensure their optimal growth?

The main purpose of wastewater treatment is to remove nutrients, notably nitrogen (N) and phosphorus (P) which, if left untreated, may lead to the eutrophication of downstream rivers and lakes, resulting in their oxygen depletion and subsequent loss of species diversity (Kontas, Kucuksezgin, Altay, & Uluturhan, 2004). Microalgae have been proposed as a means to reduce the nutrient load in wastewater treatment systems for more than 60 years with some of the earliest work having been conducted at the Sanitary Engineering Research Laboratory of University of

California (Oswald, Gotaas, Golueke, & Kellen, 1957). Microalgae grown in the wastewater treatment open ponds would utilize these nutrients in order to grow, and therefore the subsequent harvest of the algal biomass for biofuel production would effectively remove the nutrients from the wastewater (García et al., 2006). The U.S. department of energy has already identified these potential synergies, stating that “Inevitably, wastewater treatment and recycling must be incorporated with algae biofuel production”. (Barry, Wolfe, English, Ruddick, & Lambert, 2016; Ferrell & Sarisky-Reed, 2010). Indeed, algae-based wastewater treatment could be deployed relatively quickly since most of the infrastructure is already in place, algal biomass costs would be reduced by 15% to 30% due to having access to “free” nutrients (Figure 8), and wastewater treatment revenues could offset algae production costs.

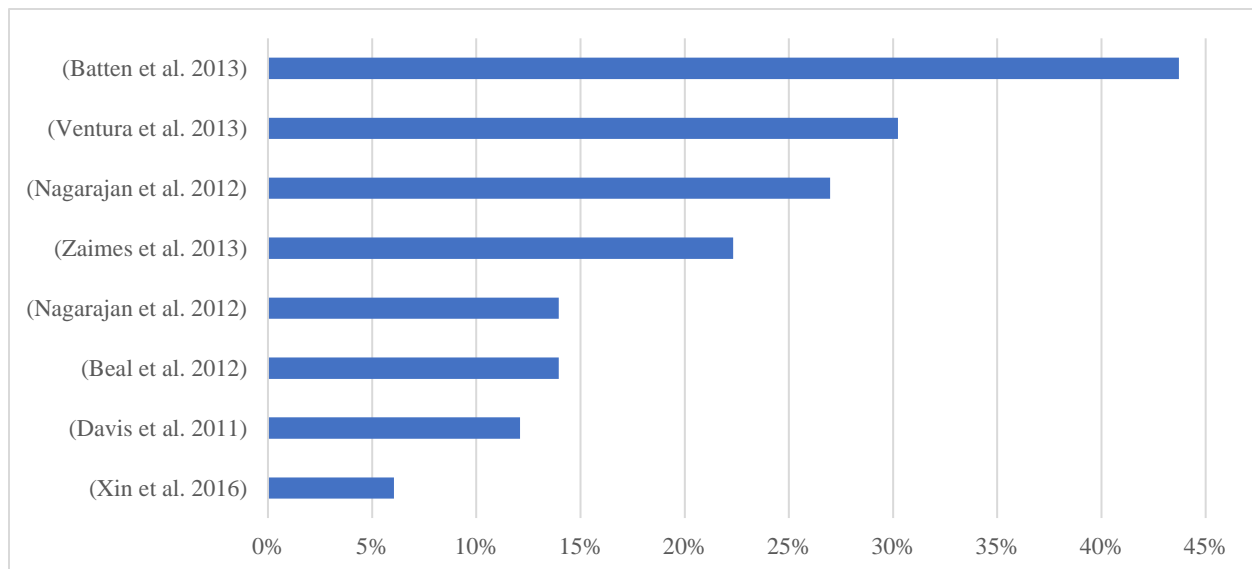


Figure 8: % Decrease in algal biomass cost from nutrient cost offset

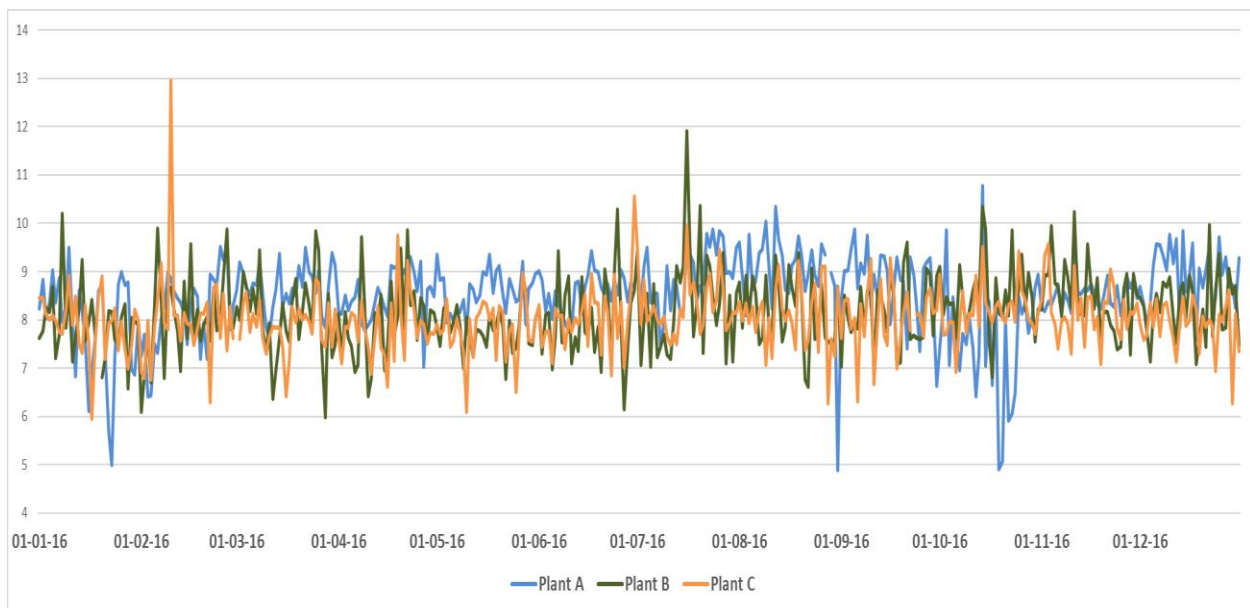
To grow efficiently, microalgae require CO₂, N and P in constant proportions. Although plankton have an optimal Redfield ratio of 106:16:1 of carbon (C):N:P, freshwater microalgae are relatively more flexible in their N and P requirements, depending on the strain, with optimal ratios

ranging between 53:8:1 to 298:45:1 C:N:P depending on the species (Enriquez, Duarte, & Sand-Jensen, 1993; Hillebrand & Sommer, 1999). While nutrients are already dissolved in wastewater, the amount of atmospheric CO₂ that dissolves in water is severely limited by its low concentration in the air and the relatively high surface tension of water (Van Den Hende, Vervaeren, & Boon, 2012). As a result, the availability of CO₂ in wastewater treatment high-rate algal ponds is primarily determined by the heterotrophic oxidation of organic compounds by bacteria present in the wastewater (Oswald, 1988).

2.3.1 Discussion and selection

Anonymized data provided by three wastewater treatment plants in southern Alberta (Figure 9) shows that influent wastewater has a yearly N:P ratio of mean 8.2 with regular daily variations of ± 1 .

Figure 9: Variation of the nitrogen to phosphorus (N:P) ratio for three treatment plants in Alberta



Source: (Anonymous, 2017)

The report does not provide dissolved organic carbon (DOC) concentrations, however independent literature suggests the C:N ratio in domestic wastewater is around 5:1 (Bashaar, 2004). Therefore, for this thesis, I assume that wastewater influent has a C:N:P ratio of 205:41:5 and that it is similar in southern Alberta and southern Ontario. Clearly, additional CO₂ dissolved in the wastewater is required to maintain optimal microalgal growth. Depending on the microalgae strain grown, additional N may have to be provided as well. The most direct way to assure that microalgae have ample CO₂ would be to pump CO₂ emitted from nearby industries into the water. The concept presented here envisions that CO₂ will be obtained from a power generator attached to the wastewater facility, that burns biogas produced from the anaerobic digestion of algal biomass residue (Figure 10).

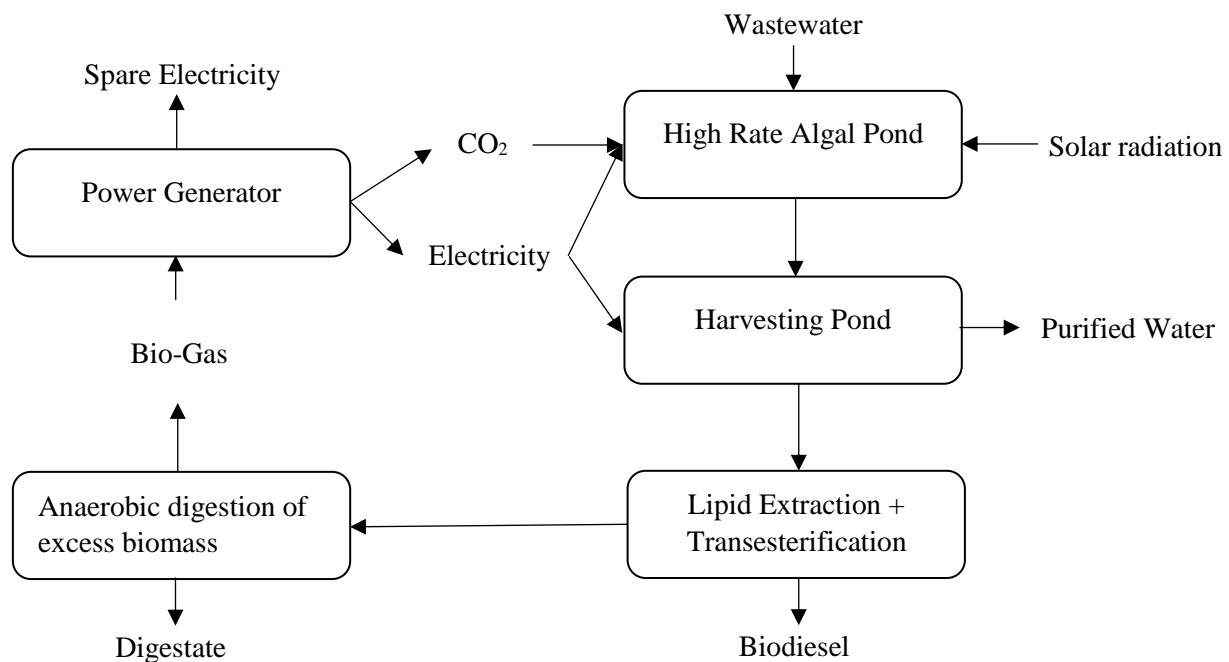


Figure 10: Flow-diagram of the FWC system

2.4: Microalgae strain selection: What strain is best suited to grow under these growth conditions?

After defining the operational parameters of temperature, insolation, and nutrient and CO₂ availability in the wastewater treatment plant, the next factor to consider is the strain of microalgae to cultivate in the facility. Microalgae species diversity is estimated to be in the hundreds of thousands, selecting the optimal strain to grow in each growth environment should be possible (Guiry, 2012). Most biofuels studies have focused on a small subset (< 30) of species with very few of these publications being comparative in nature (Larkum, Ross, Kruse, & Hankamer, 2012). Despite there being a wide range of promising strains for biodiesel production, researchers believe that the limits of biomass production are primarily determined not by the strain selected but rather by external factors such as insolation, photosynthetic and metabolic limitations (Grobelaar, 2010). There are however many other potential advantages that should be considered when evaluating different strains such as ease of harvesting, resistance to predation, and most importantly for this study temperature tolerances. More recently scientists have focused on finding or engineering strains with a high-lipid content since these triglycerides can be quickly and easily converted to biodiesel using a chemical process known as trans-esterification (Klinthong, Yang, Huang, & Tan, 2015). However very few studies have looked at the production of microalgae feedstock growth in colder climates such as Canada. Fortunately, in a recent study published in 2014, researchers from University of Montreal used a novel screening technique to identify 100 strains of microalgae present in the local (Quebec) freshwater lakes and rivers (Abdelaziz, Leite, Belhaj, & Hallenbeck, 2014). This study also provides biomass and lipid productivity for these strains in wastewater versus a synthetic medium under varying temperatures ($10 \pm 2^{\circ}\text{C}$ and $22 \pm 2^{\circ}\text{C}$) and identifies the top performing strains under each condition with regards to biomass

growth, high lipid content and nutrient removal capacity. Given the unique nature of this study and its geographic proximity to our area of study, the data provided will act as the foundation for our microalgae strain selection.

2.4.1 Discussion and selection

In their study, Abdelaziz et al. identified seven strains and five strains as high lipid producers in wastewater at 10°C and 22°C respectively (Abdelaziz et al., 2014) (Figure 11).

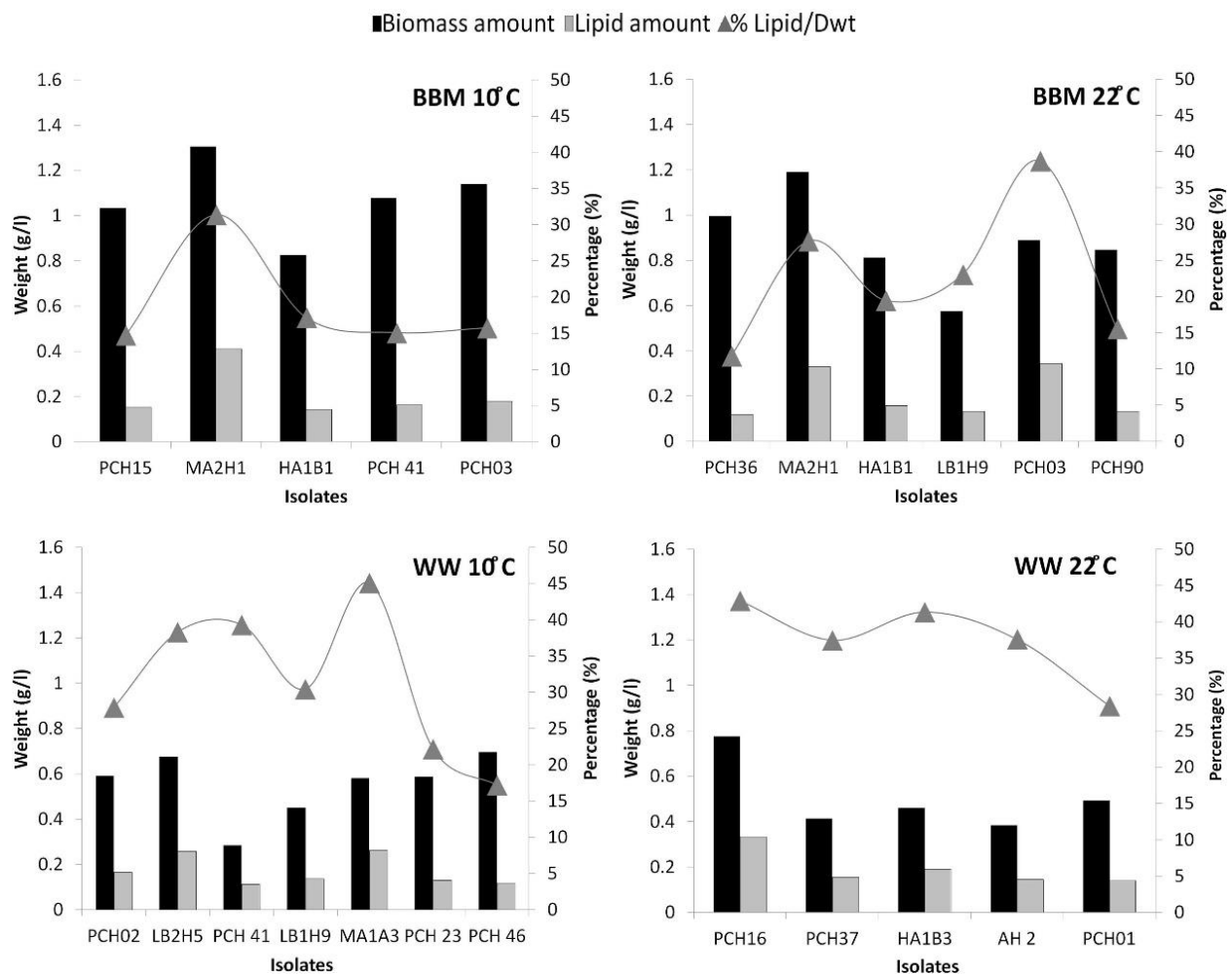


Figure 11: Biomass and lipid content of selected strain in wastewater (WW) and a synthetic medium (BBM) at different temperatures

Source: (Abdelaziz et al., 2014)

Data from the three wastewater treatment plants in southern Alberta indicate that the wastewater there varies between roughly 13°C and 19°C. Given the similar atmospheric temperature in southern Alberta and southern Ontario, I assume that wastewater temperatures would be analogous between the two regions (Figure 5). Therefore, I will focus on the seven top performing strains at 10°C since, as described in Section 2.1.2, the photosynthetic performance of microalgae significantly decreases at below optimal temperatures versus only a slight decrease at temperatures above optimal. Out of the seven-species identified (PCH02, PCH23, PCH41, PCH46, MA1A3, LB2H5 & LB1H9), the four starting with PCH are of the *Chlorella* genus, a member of the Chlorellaceae family of green algae and well-known for its adaptability. In a later study conducted by the same research department, Hallenbeck et al. determined the lipid content and growth rates of all 100 strains of microalgae identified by Abdelaziz et al. under the same growth conditions (Hallenbeck, Leite, & Abdelaziz, 2014). Out of these 100 strains, Table 5 summarizes the specific growth rates (i.e. Daily percentage growth of biomass) and lipid contents of the top performing strains grown in wastewater at varying temperatures. Given its relatively high growth rate and lipid content when grown in wastewater at both 10°C and 22°C, I select *Chlorella* MA1A3 as the microalgae for this study.

Table 5: Growth rate and lipid content of microalgae species identified by Abdelaziz et al. grown in wastewater and under different temperature conditions.

Genus	Identification	Specific Growth Rate 10°C	Lipid content 10°C (% of dry mass)	Specific Growth Rate 22°C	Lipid content 22°C (% of dry mass)
<i>Chlorella</i>	PCH02	0.63	28%	0.65	11.8%
<i>Chlorella</i>	PCH23	0.79	21.8%	1.26	8.87%
<i>Chlorella</i>	PCH41	0.73	39.36%	0.53	28.1%
<i>Chlorella</i>	PCH46	0.66	17.4%	0.69	15.3%
<i>Chlorella</i>	MA1A3	0.74	45.5%	0.98	9.16%
<i>Anabaena</i>	LB2H5	0.74	33.3%	0.69	12.3%
<i>Pseudochlorella</i>	LB1H9	0.43	23.4%	0.39	23.5%

Source: (Hallenbeck et al., 2014)

2.5 Microalgae harvesting and dewatering: How would the algal biomass be harvested and dried?

Due to the small cell size (3 – 30 μm) of unicellular microalgae strains such as the *Chlorella* genus and their relatively dilute concentration in water (200–600 mg/L dry biomass), separation of the algae from the wastewater remains a major hurdle to any commercial scale operation (Molina Grima, Belarbi, Acién Fernández, Robles Medina, & Chisti, 2003; Uduman, Qi, Danquah, Forde, & Hoadley, 2010). Methods to remove microalgae from drinking water are well established (Henderson, Parsons, & Jefferson, 2008), but there is less information on how to harvest microalgae and recover biomass for biofuel production. Microalgae collection methods often involve a two-step harvesting process where the algal cells are first aggregated through flocculation, subsequently collected via sedimentation or flotation, and then dewatered in a second step using either centrifugation or filtration followed by thermal processes (Wiley, Campbell, & McKuin, 2011).

Flocculation is a chemical process used to aggregate microalgal cells in order to increase the effective “particle” size and ease sedimentation, centrifugal recovery, and filtration depending on the composition of the wastewater and the species of algae. Flocculants, or flocculant agents, can be divided into two groups: bioflocculants that do not contaminate the algal biomass for later use as animal feed and are effective in lower concentrations than their inorganic flocculant counterparts, such as alum and iron chloride, that are more efficient but require higher concentrations to be effective (Becker, 1994; R. P. Singh et al., 2000). Since flocculants are often specific, not all flocculants will necessarily function on specific algal cell types (Bratby, 2004). Lately cationic starch has attracted interest as an alternative for inorganic and synthetic flocculants because of its low cost (\$US 100 – 600 per metric ton) and effectiveness during the liquid-solid

separation processes present in wastewater treatment and paper production (Shirzad-Semrari, Kulicke, & Lotz, 1993). As shown in Table 6, which summarizes data from a recent study by Letelier et al., the *Chlorella* flocculation efficiency of cationic starch is high (>80%) when introduced in concentrations around 40 mg/L even with biomass concentrations as low as 0.44 g/L. In addition, this same study found that increasing flocculant concentrations beyond 40 mg/L or biomass concentrations to above 0.77 g/L did not significantly increase flocculation efficiency ($P > 0.05$) (Letelier-Gordo, Holdt, De Francisci, Karakashev, & Angelidaki, 2014).

Table 6: Settling phase flocculation efficiencies for different biomass and flocculant concentrations.

Flocculant concentrations (mg/L)	Efficiency at various biomass concentration (g/L)		
	0.44 g/L	0.56 g/L	0.77 g/L
0 mg/L	58%	68%	39%
2.5 mg/L	61%	74%	49%
5 mg/L	61%	69%	56%
10 mg/L	65%	77%	76%
20 mg/L	61%	79%	89%
40 mg/L	84%	89%	90%

Source: (Letelier-Gordo et al., 2014)

This proportional relationship between *Chlorella* flocculation efficiency and flocculant concentrations was in line with the previous findings of other studies that focused on a wide range of other genera such as *Scenedesmus*, *Phaeodactylum* and *Nannochloropsis* (Bayat Tork, Khalilzadeh, & Kouchakzadeh, 2017; Hansel, Guy Riefler, & Stuart, 2014; C. Peng, Li, Zheng, Huang, & Li, 2017; Vandamme, Foubert, Meesschaert, & Muylaert, 2010). Once cells have aggregated as a result of flocculation, the resulting flocs will form a sediment that can subsequently be collected for dewatering.

Dewatering is the process by which water is removed from the collected microalgae biomass so that it can be used in downstream processing and product isolation and conversion. Centrifuges and belt filters are the most commonly used dewatering processes with the primary difference

between the two being their method of separation: whereas centrifuges use centrifugal effect to separate the algal biomass from its surrounding liquid, belt feeder systems achieve separation via gravity drainage followed by compression shear (Udom et al., 2013).

Centrifugation is often used as a dewatering technique due to its speed, efficiency and ease of use (Molina Grima et al., 2003). However the high gravitational and shear forces applied to the microalgae cells can lead to cell disruption and structural damage, thus preventing their use in biofuel conversion processes (Knuckey, Brown, Robert, & Frampton, 2006). In addition, although centrifugation might be feasible for high-value products (such as pigments, proteins and antioxidants), it is far too costly for low-value products such as biodiesel produced from integrated systems, such as the one presented in this study (Dassey & Theegala, 2013). Indeed, the U.S. Department of Energy considers the current level of technology for this process is cost-prohibitive for large-scale use and has indicated that significant cost and energy savings must be achieved for the widespread use of centrifugation as a dewatering technique (Ferrell & Sarisky-Reed, 2010).

Various forms of filtration have been found satisfactory at recovering large or flocculated algal cells (Molina Grima et al., 2003). Although there is a wide range of filter designs, these can be categorized by filter pore sizes with macro filtration using pores larger than 10 μm , micro-filtration using pores between 0.1–10 μm , ultrafiltration between 0.02–0.2 μm and reverse osmosis using pores smaller than 0.001 μm . Due to the aforementioned small cell-sizes (3 – 30 μm) of unicellular microalgae strains, micro-filtration would seem to be the most appropriate pore size for microalgae filtration (Wicaksana, Fane, Pongpairaj, & Field, 2012), however, after flocculation filters with pore sizes as large as 25 μm have been found to be effective (Shelef & Sukenik, 1984). Since the pressure required to force fluid through a membrane, and therefore the operational energy cost, generally decreases as the membrane pore size increases, being able to use macro-filtration instead

of micro-filtration significantly reduces operational energy costs and decreases costs associated with replacing the membrane (Chatsungnoen & Chisti, 2016; Pittman, Dean, & Osundeko, 2011).

Following flocculation and filtration, the microalgae biomass forms a slurry that must then be dried into a stable and storable form using thermal processes, without which the slurry would spoil within a few hours at room temperature (Milledge & Heaven, 2013; Shelef & Sukenik, 1984). In order for this slurry to be used in the oil extraction and biodiesel conversion processes, this additional dewatering process must increase the slurry's solid content up to around 90 % (Lardon, Helias, Sialve, Steyer, & Bernard, 2009). A wide range of methods have been developed to dry microalgae for subsequent processing, the main methods being solar drying, convective/oven drying, spray drying and freeze drying (Milledge & Heaven, 2013).

Solar drying is considered to be the cheapest and most energy efficient (2.8 MJ/ton of algae) drying technique with the lowest net GHG emissions (0.2 Kg CO₂ eq./ dry ton of algae) due to its utilization of solar energy (Udom et al., 2013). However it is weather dependent and requires a large amount of space and time since only around 140g of dry matter can be produced from each square meter of drying space per 3 to 5 hour cycle in warm climates (Prakash et al., 1997). In addition, the content of free fatty acids (FFAs) in the oil extracted from microalgae dried in sunlight is significantly higher than those dried using other techniques such as freeze or convective/oven drying (Balasubramanian, Yen Doan, & Obbard, 2013). These FFAs quickly react with the alkaline catalysts sometimes used during transesterification, thus reducing its efficiency and resulting in solar drying likely not being suitable for biodiesel production from microalgae (Ramadhas, Jayaraj, & Muraleedharan, 2005).

Spray drying and freeze drying are widely used techniques in research and the food industry since they do not rupture the cell walls of the microalgae (Guldhe, Singh, Rawat, Ramluckan, &

Bux, 2014). These processes are therefore the preferred methods of drying high value micro-algal products, with microalgal biomass treated by spray drying less susceptible to lipolysis in storage whilst freeze drying minimizes protein loss in the treated cells (Oliveira, Rosa, Moraes, & Pinto, 2009; Ryckebosch, Muylaert, Eeckhout, Ruysen, & Foubert, 2011). However the high energy intensity of these processes result in them being cost-prohibitive for large scale commercial recovery of algal biomass (Molina Grima et al., 2003).

Convective/oven drying is the process of drying microalgae through heated air circulation and is a popular choice for microalgal biomass drying for use in biodiesel production. By managing the temperature and flow rate of the air, the drying speed of the algal biomass can be optimized whilst minimizing the loss of algal oil and the amount of FFAs (Bennamoun, Afzal, & Léonard, 2015; Desmorieux & Decaen, 2005). Although not as cheap and environmentally conscious as solar drying, convective/oven drying remains faster and cheaper than spray or freeze drying (Klinthong et al., 2015)

2.5.1 Discussion and selection

Harvesting and dewatering of algal biomass are crucial steps in the commercial viability of microalgae for biofuel production. Indeed, because of the small cell size (3 – 30 μm) of unicellular microalgae strains such as the *Chlorella* genus and their relatively dilute concentration in water (200–600 mg/L dry biomass), studies have shown that some of the most cost and energy intensive steps in algal biomass production are harvesting and dewatering (Guldhe et al., 2014; Uduman et al., 2010), with harvesting accounting for 20% - 30% of the total cost of producing the biomass (Molina Grima et al., 2003) . Given the wide range of harvesting and dewatering techniques existing, a combination of several methods generally results in significant savings in cost, energy

demand and total emissions (Beach, Eckelman, Cui, Brentner, & Zimmerman, 2012; Bilad, Vandamme, Foubert, Muylaert, & Vankelecom, 2012; Khoo et al., 2011; Weschler, Barr, Harper, & Landis, 2014).

Amongst the harvesting techniques, flocculation – sedimentation and filtration is often considered the method of choice for biomass recovery from sewage-based processes (Molina Grima et al., 2003). Therefore, for the purposes of this study, the microalgae cultivated will first be flocculated using cationic starch due to its low cost and high flocculation efficiency in wastewater (Vandamme et al., 2010), after which they will be collected through sedimentation and dewatered through macro-filtration to minimize costs.

2.6 Biodiesel production: How will the algal biomass be converted to biodiesel?

Depending on the microalgae species and its growth conditions, lipids can constitute anywhere between 2% to 60% of the total dry cell mass (Abdo et al., 2014; Bohutskyi et al., 2014). These microalgae-derived lipids contain long-chain fatty acids and triglycerides that can be converted to biofuels through various thermochemical and biochemical processes depending on the desired output, most of which are summarized in Figure 12 (Amin, 2009; Brennan & Owende, 2010; Sukahara & Awayama, 2005).

My thesis focuses on the conversion of microalgae lipids to biodiesel, which is primarily composed of Fatty Acid Methyl Ester (FAMES), as a “drop-in” or substitute fuel for conventional diesel. There are three conversion pathways to transform microalgae lipids to biodiesel, namely

direct secretion, extraction - conversion, and processing of whole algal biomass (Ferrell & Sarisky-Reed, 2010).

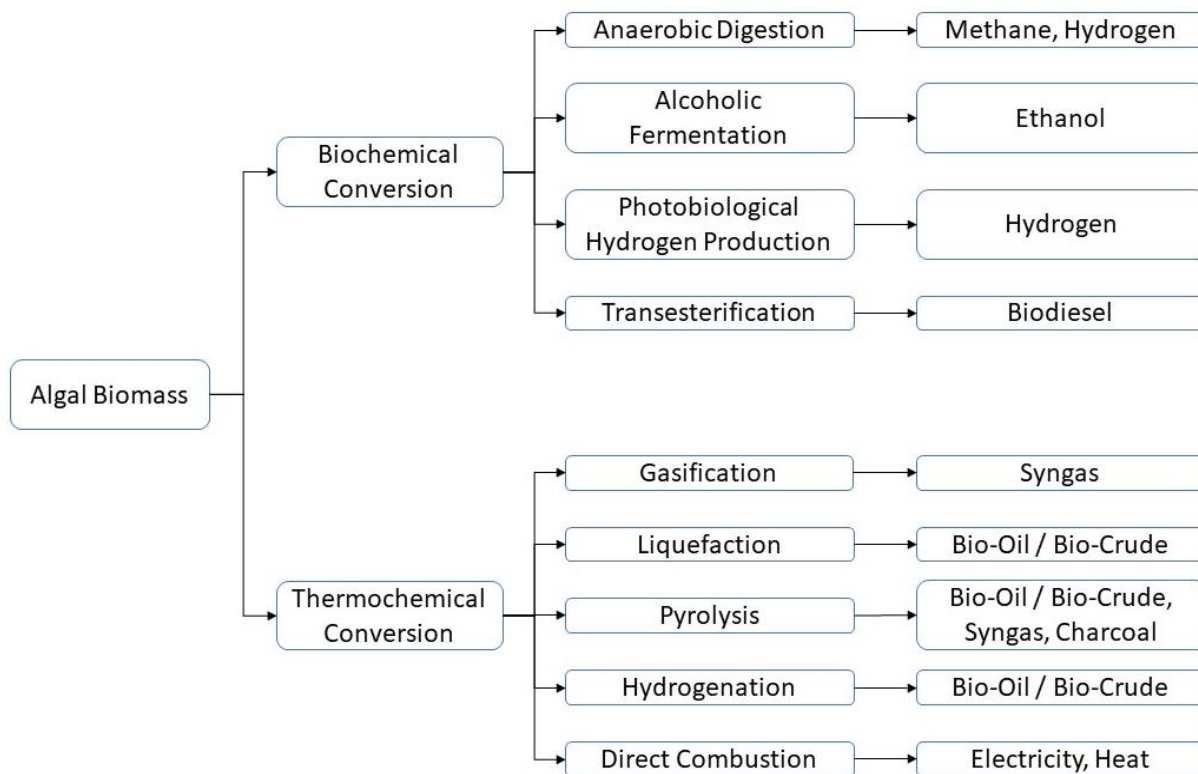


Figure 12: Potential algal biomass conversion processes

Source: (Amin, 2009; Sukahara & Awayama, 2005)

2.6.1 Direct secretion

A small subset of microalgae species secrete bio-oil or alcohol directly into their growth medium, therefore bypassing the need for extraction and conversion processes (Radakovits, Jinkerson, Darzins, & Posewitz, 2010). These secretions have the advantage of being easily convertible to biofuels, however the very low growth rates of these algal species make them unsuitable for conventional biofuels production (Metzger & Largeau, 2005).

2.6.2 Extraction – conversion

2.6.2.1 Extraction

Extraction methods are categorized as wet or dry, depending on the amount of dewatering required for the successful extraction of bio-oil/bio-crude from the microalgae biomass. These extraction techniques were reviewed in detail by Cooney, Young, & Nagle (2009) and Mercer & Armenta (2011).

2.6.2.1.1 Solvent extraction

Solvent extraction relies on chemical solvents to isolate and separate compounds with different polarity and molecular size. In the context of microalgae biomass, a solvent or co-solvent mixture is used to extract crude lipids from the rest of the cell biomass (Grima, González, & Giménez, 2013). The classic mixture for lipid extraction (i.e., the Bligh and Dyer method) is composed of a chloroform and methanol co-mixture that relies on chloroform to dissolve the triglycerides and methanol to dissolve the polar membrane lipids (Bligh & Dyer, 1959; Cooney et al., 2009; Mercer & Armenta, 2011). When applied at a large scale however, the Bligh and Dyer method requires that large amounts of solvent be evaporated, resulting in a high energy consumption of 46.069 MW/mol for ethanol (Resa, Goenaga, Iglesias, Gonzalez-Olmos, & Pozuelo, 2006), and environmental concerns given the solvent's toxic nature (Breil, Abert Vian, Zemb, Kunz, & Chemat, 2017).

The Soxhlet extraction method is also appropriate to extract lipids from microalgae. A solvent, typically n-hexane, is vaporized, condensed, then percolated repeatedly through the algal biomass to dissolve and extract the lipids. Despite initially promising life cycle assessment of Soxhlet extraction, , long processing times coupled with the relatively poor extraction of polar lipids and

the large amounts of solvent required make this method unsuitable for large scale lipid extraction from microalgae (Ranjan, Patil, & Moholkar, 2010).

In response to the challenges associated with traditional solvent extraction methods, new processes utilizing supercritical fluids such as supercritical CO₂ were developed (Mercer & Armenta, 2011). CO₂ has a moderate critical point (31.1 °C and 72.9 atm), is non-toxic and is a gas at room temperature. Supercritical CO₂ extraction has several key advantages over other solvent extraction methods, notably no contamination of the algal biomass, lower energy consumption and processing temperatures, easy solvent recovery, and selectivity for triglycerides (Cheng et al., 2011; Mendes, Nobre, Cardoso, Pereira, & Palavra, 2003). Despite these advantages over organic solvents, supercritical CO₂ performs poorly in terms of the quantity of lipids extracted, achieving only half of the yields obtained with a traditional Soxhlet extraction on the same algal biomass, resulting in this method being unfeasible for large-scale biodiesel production from microalgae (Bjornsson, MacDougall, Melanson, O’Leary, & McGinn, 2012).

2.6.2.1.2 Cell disruption

For solvent extraction to be effective, the solvent must first penetrate the solid matrix surrounding the lipid, then make physical contact with the lipid and finally dissolve it (Cooney et al., 2009). Unfortunately, most microalgae cells have a thick cell wall that limits the solvent’s access to the lipids contained inside. A wide range of cell disruption techniques have been developed to remove the cell wall barrier, such as mechanical pressing, bead beating, ultrasound / sonication, osmotic shock, and microwave heating.

Mechanical pressing is often used for high oil recovery from oil seeds like canola, palm, and coconut. The principal behind this process is the application of a high mechanical pressure to

disrupt algal cells and squeeze out the oil that they contain. Applying the right amount of pressure is crucial to maximizing extraction efficiency to around 70-75%. However the application of too much pressure would result in increased heat generation, choking and decreased lipid recovery (Topare et al., 2011). In addition, press methods are expensive due to high maintenance costs, can involve long processing times and oil extraction from microalgae is hindered by their rigid cell structure and small size (Boldor et al., 2010; Johnson & Wen, 2009).

Bead beating uses grinding and attrition to damage algal cells by spinning the algal biomass at high speeds with fine beads (Geciova, Bury, & Jelen, 2002; S. J. Lee, Yoon, & Oh, 1998). There are two common types of bead mills, shaking vessels and agitated beads: whereas in the shaking vessel type the entire culture vessel is shaken, the agitated bead types provide better disruption and extraction efficiencies by agitating the beads with the algal biomass. These processes do not require the prior dewatering of the algal biomass and can successfully disrupt over 90% of cells, however the high specific energy consumption of bead mills relative to other disruption techniques suggests that it is unlikely to be used for biofuel production from algal biomass (Doucha & Lívanský, 2008; A. K. Lee, Lewis, & Ashman, 2012).

Ultrasound assisted extraction, otherwise known as sonication, causes damage to the algal cells through two major mechanisms: cavitation and acoustic streaming. Cavitation is the process by which the applied ultrasound causes the production of microbubbles, which in turn create pressure on the cells, breaking them up, whereas acoustic streaming facilitates mixing of the algal culture (Khanal, Grewell, Sung, & Van Leeuwen, 2007; Suslick & Flannigan, 2008). This process is simple, easy to use, minimizes denaturation of biomolecules by only generating low amounts of heat, and does not require beads or chemicals that would have to be removed in downstream processes (Harrison, 1991). However, prolonged exposure to sonication can lead to the generation

of free radicals in algal biomass, which in turn can degrade lipids through oxidation (Ghasemi Naghdi, González González, Chan, & Schenk, 2016).

Osmotic shock causes algal cells to burst as a result of an abrupt lowering of osmotic pressure. This is often achieved by placing marine algae in fresh water (hypotonic shock) or fresh water algae in a highly saline environment (hypertonic shock) (Mercer & Armenta, 2011). Despite being an ecological process, osmotic shock was experimentally shown to be ineffective in comparison to other cell disruption techniques such as sonication and microwave heating (Prabakaran & Ravindran, 2011).

First reported in the 1980s, microwave extraction has allowed the development of rapid and safe lipid extraction techniques which remain some of the most effective, simple and economically viable processes used to date (Dai, Chen, & Chen, 2014; J. Y. Lee, Yoo, Jun, Ahn, & Oh, 2010). Applicable to both wet and dry algal biomass, microwave radiation generates intracellular heating, resulting in the formation of water vapor which in turn disrupts the cell from within. This triggers the electroporation effect, further opening the cell membrane and thereby facilitating the efficient extraction of lipids (Ma et al., 2014). The main disadvantages of microwave assisted lipid extraction are the associated maintenance costs when used at a commercial scale, however a study has shown that the sequential use of domestic microwave ovens generates similar results without the need for specialized and expensive units (Mahesar et al., 2008).

2.6.2.1.3 Cell disruption with solvent extraction

Following cell disruption, it is possible for bio-oil to separate naturally from the biomass and water, given enough settling time. However, this is inefficient. Industrial processors rely on solvent extraction methods for rapid recovery of bio-oil from the mixture containing water, lipids and

cellular debris. Many studies have examined the lipid yields for various types of cell disruption and solvent extraction techniques, and report varying degrees of lipid extraction efficiency (J. Y. Lee et al., 2010; Prabakaran & Ravindran, 2011). These findings highlight the need to select the appropriate cell disruption method for a particular microalgae species, and that cell disruption techniques coupled with solvent extraction can increase lipid recovery efficiency over solvent extraction methods alone.

2.6.2.2 Conversion

Once triglycerides have been extracted from microalgae, they can then be transformed into biodiesel through transesterification (Johnson & Wen, 2009; Lemões et al., 2016; Mallick et al., 2016). Alternative processing method, called in-situ methods, avoid the costs, labor and time associated with disruption and solvent extraction techniques by using alcohols as both the solvent and the reactant in the transesterification process (Chen et al., 2015; Wahlen, Willis, & Seefeldt, 2011; Zhang, Li, Zhang, & Tan, 2015).

2.6.2.2.1 In-situ conversion of dry biomass

The first report of in-situ conversion relied upon sulphuric acid as a catalyst to break down the cell walls of dry algal biomass, whilst the triglycerides and cell membrane lipids were converted to biodiesel with an abundance of methanol (Wahlen et al., 2011). Compared to the traditional two-step extraction and transesterification process, direct transesterification of dry algal biomass has been found to result in 10% - 20% higher biodiesel yields (28% vs 22.2%) and has the potential of simplifying conversion process, reduce the heat requirement as well as overall costs (Johnson & Wen, 2009; Li et al., 2011). Despite these advantages over more traditional two-step conversion

pathways, further improvements must be made to further optimize the biodiesel yield, control the fuel quality and sulfur content in order for these technologies become more widespread.

2.6.2.2.2 In-situ conversion of wet biomass

Drying of microalgae biomass prior to transesterification can be avoided by using I supercritical methanol transesterification of wet algal biomass to simultaneously extract and convert lipids to biodiesel (Patil et al., 2011). Similar to the in-situ conversion of dry biomass, methanol was introduced in abundance (9:1 methanol/biomass ratio), however the reaction had to be maintained at a temperature of 400 °C which may offset energy saving resulting from the removal of the drying phase (Marulanda, 2012). Levine et al. has proposed an alternative process involving the subcritical hydrolysis of algal biomass paired with the supercritical ethanol esterification of fatty acids retained from the hydrolysis.

2.6.3 Total algal biomass conversion

To circumvent the complexity, energy intensity and capital costs with extraction – conversion processes, many researchers have proposed the direct conversion of the entire algal biomass. These methods can be divided into biological and thermochemical processes.

2.6.3.1 Biological conversion

Anaerobic digestion is a biological process where bacteria digest the algal biomass and produce methane (i.e. biogas) that can then be used to produce methanol or burned to generate heat and/or power. The digestibility of algal biomass is strongly correlated with the strength of the cell wall and therefore the strain of microalgae being digested (Mussnug, Klassen, Schlüter, & Kruse, 2010). Since extraction- conversion is not necessary, the energy balance is improved with anaerobic digestion, however, due to transportation difficulties and the low-value nature of the

biogas generated, it is unlikely that anaerobic digestion will ever be commercially viable as a standalone process. However, anaerobic digestion can be used to upgrade low-value byproducts of the biodiesel production process, therefore potentially improving the economic viability of biodiesel production pathway.

Data provided by Ward et al. indicate that the C:N ratio of various microalgae species varies between 4.16 and 7.82, which is far below the 20 C:N ratio required to balance the carbon and nitrogen requirements of the bacterial community. This imbalance results in the excess nitrogen being released as ammonia, that in turn causes volatile fatty acids to accumulate in the digester, therefore decreasing the digester's efficiency (Ward, Lewis, & Green, 2014). By mixing glycerol, a biproduct of biodiesel production, into the microalgae residues, the C:N ratio of the mixture is raised to 12.44 resulting in a significant increase (>50%) in the production of biogas, as compared to when the residues are digested alone (Ehimen, Sun, Carrington, Birch, & Eaton-Rye, 2011).

2.6.3.2 Thermochemical conversion

The total conversion of algal biomass into liquid fuels can be achieved via thermochemical processes, namely gasification, pyrolysis and liquefaction (Biller & Ross, 2011). Gasification is the partial oxidation, at high temperatures (700–1,300 °C), of algal biomass to produce syngas, that in turn is typically converted to a liquid using the Fischer Tropsch process, and then upgraded into a liquid fuel using hydro-processing (Swanson, Platon, Satrio, & Brown, 2010). Standard gasification and advanced gasification methods (e.g., with supercritical and subcritical water (Caputo, Rubio, Scargiali, Marotta, & Brucato, 2016; Onwudili, Lea-Langton, Ross, & Williams, 2013) require a dry feedstock, so the algal biomass must be dried before transformation. The cost of drying a wet feedstock, coupled with the additional cost to upgrade syngas and perform hydro-processing, means that this technology has limited commercial application. Typically, the cost of

producing liquid fuel via gasification cannot compete with the low-cost natural gas resources that are widely available (Brandenberger, Matzenberger, Vogel, & Ch, 2013; G. Peng, 2015).

Occurring in an oxygen-free environment heated to 400-600 °C, pyrolysis is the process by which dry biomass is turned into bio-oil, syngas and charcoal (biochar). The bio-oil generated via this technique is acidic, viscous, contains chemically dissolved water and is unstable (Chiaramonti, Oasmaa, & Solantausta, 2007). It is therefore considered as an intermediary product since it must be further upgraded in a biorefinery to be utilized as a transport fuel substitute (Chiaramonti, Prussi, Buffi, Casini, & Rizzo, 2015). Hydrothermal liquefaction differs from pyrolysis because it occurs in a high pressure environment (5 – 20 MPa) and requires hydrogen alongside a catalyst (Goyal, Seal, & Saxena, 2008). Reactors used for this process are complex and expensive but provide the significant advantage of being able to convert wet algal biomass (Hise et al., 2016). Like pyrolysis, the bio-oil generated via hydrothermal liquefaction must first be upgraded so that it can be used as a transportation fuel.

The major issue with the three aforementioned methods of processing whole algal biomass into liquid fuels is that they yield low-value products that must be upgraded before being brought to market as a transportation fuel. Despite numerous studies highlighting the energetic viability of these methods, even when including upgrading pathways, yields are reduced whilst capital and operational costs increase (Chiaramonti et al., 2015; Goyal et al., 2008; A. Lee, Lewis, Kalaitzidis, & Ashman, 2016).

2.6.4 Discussion and selection

Numerous biodiesel and organic synthesis studies have shown that microwave-assisted extraction and transesterification processes can be conducted at atmospheric pressures, with much

shorter reaction times and at temperatures close to the boiling point of methanol, resulting in lower costs and equivalent yields than more traditional pathways (Azcan & Danisman, 2007; Barnard, Leadbeater, Boucher, Stencel, & Wilhite, 2007; Hernando, Leton, Matia, Novella, & Alvarez-Builla, 2007; Nicholas E. Leadbeater & Lauren M. Stencel, 2006; Patil, Gude, Camacho, & Deng, 2010; Refaat, 2010). Consequently, I assume that a microwave-assisted single stage in-situ conversion of dry-biomass process is utilized, resulting in a lipid to biodiesel conversion efficiency of 99.9% (Koberg, Cohen, Ben-Amotz, & Gedanken, 2011).

2.7 Financial analysis metrics

2.7.1 Net Present Value (NPV)

A core principal of finance is the time value of money (TVM): money currently available is worth more than the same amount in the future because of inflation or its earning potential due to interest. TVM, also known as the present discounted value of money, is given in Equation 3:

$$PV = \frac{FV}{\left(1 + \frac{i}{n}\right)^{nt}} \quad (3)$$

where “PV” is the present value of money, “FV” the future value of money, “i” is the interest rate, “n” is the number of compounding periods per year and “t” is the number of years (Investopedia, 2017g). I assume that $n = 1$, meaning that interest is compounded annually rather than quarterly. As a result, Equation 3 becomes:

$$PV = \frac{FV}{(1 + i)^t} \quad (4)$$

Applying this principal to the profitability analysis of a project, I determine the present values of all the projected cash inflows and outflows over the lifecycle of the project and therefore calculate its net present value (NPV) as:

$$NPV = \sum_{t=1}^T \left(\frac{C_t}{(1+r)^t} \right) - C_0 \quad (5)$$

where C_t is the net cash flow at time “t”, C_0 is the net cash flow at the time of investment $t = 0$, “t” is the number of time periods and “r” is the discount rate (Investopedia, 2017c). The discount rate reflects the risk exposure for an investor so that the higher the risk, the higher the discount rate must be to compensate. For the base case of this study, a discount rate of 5% was chosen. A positive NPV indicates that a project’s projected earnings outstrip the project’s projected costs in present dollar terms and as such is profitable. NPV is often used as an investment decision rule so that only projects with a negative NPV should likely be rejected. In calculating the NPV of the FWC system, neither interest nor taxes were included: interest was not considered because including the cost of financing the project would give a distorted view of the profitability of the underlying project given that financing costs are highly dependent the source of financing. Taxes were not included because corporate taxes are calculated on the net profit or loss of all a company’s profits; therefore, including taxes in the valuation of the FWC system would not properly reflect its revenue generation potential. In addition, supply-chain costs associated with selling the produced biodiesel were not modeled given the high degree of uncertainty that would be associated with estimating them and the fact that they would not reflect the profitability of the project itself, instead these costs are assumed to be covered by the buyer and are included in the model by using the wholesale price of diesel instead of the commercial price.

2.7.2 Internal Rate of Return (IRR) and Payback Period

Another benchmark often used in capital budgeting is the internal rate of return (IRR) which represents the interest rate at which the NPV is equal to zero. The formula for the IRR is a variation on Equation 5:

$$NPV = \sum_{t=1}^T \left(\frac{C_t}{(1 + IRR)^t} \right) - C_o = 0 \quad (6)$$

IRR indicates the expected rate of growth of a project. Although this will often vary from the actual rate of return, projects with an IRR substantially higher than alternative solutions are more likely to experience strong growth (Investopedia, 2017b).

The final financial metric used in this thesis is the payback period. It represents the time required to recover the cost of an investment. Unlike NPV, IRR and other methods of capital budgeting, the payback period ignores the time value of money (TVM), instead focusing on the nominal value of cash flows. The payback period is another tool at a financial analyst's disposal in determining whether or not to undertake a project, given that longer payback periods are likely not desirable for investment positions (Investopedia, 2017e).

2.7.3 Scenario and sensitivity analysis

Given that NPV, IRR and payback period rely on a project's estimated net cash flows, that in turn are derived from a wide range of estimates and assumptions, they may vary significantly due to changes in the underlying independent variables used to calculate the project's profitability. A sensitivity analysis, also known as a what-if or simulation analysis, is used in conjunction with a multiple scenario analysis to evaluate the variability and risk associated with the profitability estimates (Investopedia, 2017f). An outcome variable's sensitivity to changes in an input variable

is given by the slope of the curve where the outcome variable is on the Y-axis and the input variable is on the X-axis. The sensitivity analysis of NPV and IRR to percentage changes in key revenue and key cost variables, where slopes (i.e. sensitivity factor) were estimated using linear regression. A positive slope indicates a positive correlation between the two variables, and inversely for a negative slope, whilst the absolute value of the slope illustrates the sensitivity of the outcome variable to a change in the input variable. The key variables that were considered in this study were:

- Price of Diesel
- Ontario Electricity Price (due to case study location)
- Quantity of Biodiesel Produced
- Quantity of Methane Produced
- % of N & P removal savings passed on to a private company
- Capital Costs
- Operational Costs

These parameters are self-explanatory, except for “% of N & P removal savings passed on to private a company”. Indeed, a project like this would save a great deal of money for the wastewater treatment plant by reducing or eliminating their need for conventional processes to remove nutrients in the wastewater. If this project were to be implemented by the owners of a wastewater treatment plant, the option selected for this thesis, 100% of those savings would be incurred. However, if a 3rd party private company were to present this project to a separate treatment plant owner, they would not be able to charge for 100% of the savings generated, since that would be a financial disincentive for the treatment plant operator. It is likely the private company would have to settle for a percentage of the cost savings generated for the wastewater treatment plant.

Multiple scenario analysis looks at the impact on NPV, IRR and payback periods of changing base case revenues and costs by $\pm 25\%$. These scenarios are broken down as follow:

- Overly Conservative: capital and operational costs increased by 25%, operational revenues decreased by 25%
- Conservative: Capital and operational costs increased by 25%
- Base Case: No change to underlying assumptions
- Optimistic: Operational revenues increased by 25%
- Overly Optimistic: Operational revenues increased by 25%, capital and operational costs decreased by 25%.

Chapter 3: Results and Discussion

3.1 Key assumptions and calculations for base case scenario

Capital expenditures (CapEx) are funds used by companies to purchase, maintain or upgrade physical assets and are often used by firms to undertake new projects or investments (Investopedia, 2017a). Operating expenses (OpEx) on the other hand are the set of expenses that a company or project incurs through its normal business operations such as equipment and inventory costs, rent, labor, insurance and so on (Investopedia, 2017d). The case study assumes that all capital expenditures are incurred at time $t = 0$ and that yearly operating revenues and expenses remain constant over the lifespan of the FWC system. This study estimates Capex and OpEx based on literature review and industry standards. Estimates rely on the assumption that CapEx and OpEx are proportional to the size of the FWC system, however to minimize uncertainty, I selected data from systems of a comparable size as my case study (85 hectare wastewater treatment facility, see justification below) with a dry equivalent annual biomass production of 40 000 metric tons per year. In addition, currencies were converted to Canadian Dollars (CAD) using the 1-year average market exchange rate as of December 31st, 2017. Currency conversion rates and other conversion factors are reported in Table 7.

Table 7: Various conversion rates used in financial modelling

Conversion	Conversion rate	Inverse Conversion	Inverse Conversion rate	Source
Currency Conversion rates				
USD to CAD	1.297	CAD to USD	0.771	(OFX, 2018)
EUR to CAD	1.464	CAD to EUR	0.683	(OFX, 2018)
GBP to CAD	1.669	CAD to GBP	0.599	(OFX, 2018)
AUD to CAD	1.020	CAD to AUD	0.980	(OFX, 2018)
Biofuel Conversion rates				
Biodiesel Gal to L	3.78541	Biodiesel L to Gal	0.26	(Aqua Calc, n.d.)
Biodiesel Barrel to L	158.987	Biodiesel L to Barrel	0.0063	(Aqua Calc, n.d.)
Methane Kg to MJ	52.5	Methane MJ to Kg	0.019	(World Nuclear Association, 2018)
Methane Kg to L	0.424	Methane L to Kg	2.36	(Aqua Calc, n.d.)
Energy Conversion rates				
GJ to kWh	277.78	kWh to GJ	0.0036	(Unit Juggler, n.d.)

3.1.1 CapEx

Economies of scale for microalgae production in open ponds are limited when the treatment facility is beyond a certain size. For instance, only a 1% decrease in production cost was observed per 10 000 metric tons/year increase in plant capacity beyond 40 000 metric tons/year (Brownbridge et al., 2014). This confirms previous observations showing that a 10x increase in pond size from 100 ha to 1 000 ha only resulted in an estimated 0.5% decrease in algal biomass production costs (Rickman et al., 2013; Sun et al., 2011). Indeed, open pond system cost contributions, such as land and mixing energy, are often directly proportional to total algal biomass produced (Norsker et al., 2011). Assuming that the target annual biomass production capacity is 40 000 metric tons/year to maximize economies of scale, that would translate to $(40\,000)/365 = 109.59 \text{ metric tons/day}$. Given the specific growth rate of *Chlorella* MA1A3 in wastewater at 10°C of 0.74 (Table 5), there would need to be $109.59/0.74 =$

148.09 *metric tons* of “permanent” algal biomass in the system in order to sustainably harvest 109.59 *metric tons/day*. Since *Chlorella* MA1A3 has a biomass density in wastewater of approximately 580 *mg/L* or $0.58 * 10^{-6}$ *metric tons/L* (Abdelaziz et al., 2014), the proposed open pond must be able to contain $\frac{148.09}{0.58*10^{-6}} = 255.33 * 10^6$ *Litres* = $255.33 * 10^3$ *m*³ of water. To maximize light penetration in the culture environment, the open pond is limited to a culture depth of 0.25 – 0.3 meters (Chisti, 2016), therefore the footprint of the open pond would have to be:

$$255\,333/0.3 = 851\,110\,m^2 = 85.11\,hectares.$$

Unfortunately, a central repository for land prices across Canada does not exist, however there are a range of reports, studies and articles on farmland prices. Assuming that regular land prices would be at or below those for farmland, I will use data provided by Farm Credit Canada (FCC) and Statistics Canada to set our base case land price in southern Ontario at \$2.718 *CAD/m*² (\$11 000 *CAD/acre*) for 2017 based on an average 2016 annual price of \$2.609 *CAD/m*² (\$10 588 *CAD/acre*) and 3.9% an estimated annual price growth rate across the region (Canada Statistics, 2017; Farm Credit Canada, 2017). This gives an estimated land cost of $851\,110 * 2.718 = \$2\,313\,450$ *CAD* for the base case of the proposed open pond.

All other capital costs, with the exception of the contingency, which was set at 10% (Zamalloa, Vulsteke, Albrecht, & Verstraete, 2011), were estimated from literature by dividing data by the surface area used in hectares or metric tons of dry biomass equivalent produced in the study, converting to Canadian dollars using the conversion rates found in Table 7, and then multiplying the result by 85.11 hectares or 40000 metric tons to get the estimates used in this thesis.

3.1.2 OpEx

Most operational costs were estimated from literature using the same method outlined above in the CapEx section. However, the cationic starch concentration was estimated as follows. Given that *Chlorella* MA1A3 has a biomass density in wastewater of approximately 580 mg / L (Abdelaziz et al., 2014), that a cationic starch concentration of 40 mg / L is the most efficient at flocculating *Chlorella* at concentrations around 560 mg / L (Table 6), and lastly that 255.33 * 10⁶ Litres of water are being treated per day in the FWC system, a total of $\frac{(40 * 255.33 * 10^6) * 365}{10^9} = 3727.8$ metric tons/year of cationic starch would be consumed at the facility. Based on an estimated cationic starch cost of \$1556 CAD / metric ton (Made-in-China.com, 2018; Vandamme et al., 2010), I get an estimated base case yearly cationic starch cost of 3727.8 * 1556 = \$5 802 051 CAD / year.

Maintenance was set at 5% of total capital costs, whilst supervision costs were set at 2% of total labor costs (R. Davis, Aden, & Pienkos, 2011). The lifespan of the project was estimated at 20 years (Barlow, Sims, & Quinn, 2016; R. Davis et al., 2011; Grima et al., 2013; Lardon et al., 2009; Ramos Tercero et al., 2014). Depreciation was calculated using a straight-line depreciation method assuming a 20% salvage rate and $annual\ depreciation\ rate = \frac{1}{project\ lifespan} = 5\%$ so that:

$$Depreciation = (1 - Salvage\ Rate) * CapEx * annual\ depreciation\ rate$$

$$\Leftrightarrow Depreciation = (1 - 20\%) * 69,679,997 * 5\% = \$253,382\ CAD$$

Finally, electricity cost was derived by conservatively doubling the electrical requirement per hectare from an estimated 50 kWh/ha/day (Dai et al., 2014; Klinthong et al., 2015; Zamalloa et al.,

2011) to 100 kWh/ha/day in order to achieve a total yearly electrical requirement of $100 * 365 * 85.11 = 3,106,515 \text{ kWh/year}$. This was then multiplied by Ontario's July 2017 mid-peak electricity price of 9.5 ¢ per kWh (Ontario Energy Board, n.d.) to get an estimated electricity cost for the facility of \$295,199 CAD.

3.1.3 Revenues and cost savings

Despite Ontario joining a common carbon market in January 2018 with Quebec and California, I do not include the market value of carbon credits produced in the valuation of the case study. This decision was made primarily because of the high degree of inaccuracy associated with estimating net GHG emissions of the FWC system and carbon price variations anticipated during the lifespan of the project. Similarly, I did not account for the revenues generated from the sale byproducts due to the lack of information about the type and quantity of byproducts that could be produced by the FWC system.

As the microalgae system is expected to produce 40 000 metric tons of dry algal biomass per year, I can derive revenues generated from the sale of biodiesel and electricity produced through the combustion of biogas, as well as cost savings at the wastewater treatment plant for the removal of N and P:

- Biodiesel revenues: From Table 5, I know that the lipid content of the microalgae will be around 45.5% (Hallenbeck et al., 2014), meaning that $45.5\% * 40\,000 = 18\,200 \text{ metric tons}$ of lipids are produced annually. Given the estimated 99.9% lipids to biodiesel conversion rate of the proposed conversion process (Koberg et al., 2011), assuming a biodiesel density of 0.801 kg/L (Christenson & Sims, 2011) this translates to $99.9\% * 18\,200 = 18\,182 \text{ metric tons} = 5\,996\,408 \text{ Gallons} = 22\,698\,863 \text{ L}$ (Table 7) of

biodiesel produced annually. I also assumed that the biodiesel would be sold at the same price as conventional diesel ($\$3.58 \text{ CAD} / \text{Gallon} = \$0.946 \text{ CAD} / \text{L}$) rather than the subsidized higher price available to current biodiesel ($\$4.38 \text{ CAD} / \text{Gallon} = \$1.16 \text{ CAD} / \text{L}$). This resulted in annual revenues from the sale of biodiesel being equal to $0.946 * 22\,698\,863 = \$21\,465\,739 \text{ CAD} / \text{year}$

- Methane combustion revenues: Following the biodiesel conversion, there remains $40\,000 - 18\,200 = 21\,800 \text{ metric tons}$ of lipid extracted algae (LEA) that can be converted to methane via anaerobic digestion with an efficiency of 0.4 mL/g (Bohutskyi et al., 2014), to produce $3\,697\,280 \text{ kg} = 194\,107 \text{ GJ}$ of methane (Table 7). This methane is in turn combusted in a combined heat and power unit with an electrical conversion efficiency of 40% (Clarke Energy, n.d.), to produce $21\,465\,739 \text{ kWh}$ of electricity (Table 7) with the heat generated being redirected to heating the water in the algal ponds. This electricity can then be sold at the aforementioned Ontario mid-peak electricity price of 9.5 ¢ per kWh (Ontario Energy Board, n.d.) to generate an additional $((21\,465\,739 * 9.5))/100 = \$2\,048\,911 \text{ CAD}$ in revenues. Potential revenues from the sale of the methane were also considered, but these were negligible relative to revenues generated from methane combustion and therefore were not considered a revenue for the wastewater treatment facility.
- N & P removal cost savings: Anonymized data provided by three wastewater treatment plants in southern Alberta indicate that untreated wastewater contains 49.08 mg N / L and 6.01 mg P / L , on average. Wastewater treatment plants are required to remove most if not all of these nutrients from the wastewater at an estimated cost of $\$5.12 \text{ CAD/Kg}$ for nitrogen and $\$3.51 \text{ CAD/Kg}$ for phosphorus (Zamalloa et al., 2011). Assuming that the microalgae have a 99% removal efficiency for both N and P (Abdelaziz et al., 2014), this translates to:

4 528 010 *kg/year* = \$23 203 566 *CAD/year* in cost savings from N removal

554 885 *kg/year* = \$1 949 816 *CAD/year* in cost savings from P removal

In the base case of this study it is assumed that local governments, which currently operate wastewater treatment facilities across Canada, would invest in the proposed microalgae project, and therefore they would realize 100% of the calculated cost savings. If a private company would be the one to invest in this project, they would be able to charge a percentage of the cost savings to the wastewater treatment plant. A sensitivity analysis of the project to this percentage amount has been conducted and will be discussed later in this case study.

3.2 Financial analysis, scenario-based analysis and sensitivity analysis

The financial analysis of the base case model is presented with the scenario-based analysis in Table 8, with the key base case parameters being:

Annual Algal Biomass produced: 40,000 metric tons

Biodiesel Produced: 22 698 863 *L / year*

Electricity from methane combustion: 21,567,484 kWh / year

Price of Diesel: \$0.946 CAD / L

Ontario Energy Price: 9.5 cents / kWh

Electrical Requirement per hectare: 100 kWh/ha/day

Land Used: 85.11 ha

% of N & P removal savings passed on to a private company: 100%

Operational Costs (OpEx): \$21,610,581 CAD / year

Capital Costs (CapEx): \$69,681,366 CAD / year

Revenues (incl. cost savings): \$48,668,032 CAD / year

Project Lifespan: 20 years

Discount Rate: 5%

The base case model the FWC system is highly profitable, as shown by the positive NPV of roughly \$267.5M CAD, an IRR of 38.77% and a payback period of 3 years (Table 8). Even in the overly conservative case, where revenues were decreased and costs were increased by 25%, the project is found to be profitable with a positive NPV of roughly \$165.8M CAD, an IRR of 22.92% and a payback period of 5 years (Table 8).

Table 8: Key financial outcomes of studied scenarios

<i>Scenarios</i>	Overly conservative	Conservative	Base	Optimistic	Overly Optimistic
<i>Net Present Value (NPV)</i>	\$ 165,795,027	\$ 250,093,939	\$ 267,514,280	\$ 351,813,192	\$ 369,233,534
<i>Internal Rate of Return (IRR)</i>	22.92%	30.92%	38.77%	48.52%	64.71%
<i>Payback Period (years)</i>	5	4	3	3	2
<i>Investment Multiple</i>	1.9X	2.9X	3.8X	5.X	7.1X

The IRR of each scenario follows a natural logarithmic curve with an inflection point in either year 3 or year 4 of the project (Figure 13). This means that the rate at which the profitability of the project increases, for a unit increase in the time period, decreases with time. Changing the expected lifespan of the project by ± 10 years would therefore have little impact on the expected IRR of the project under the various scenarios. Running a linear regression analysis (not shown) on the NPVs of the different scenarios resulted in a high R-squared score of 0.9831, which is consistent with the fact that the yearly net profits of the project are held constant over the project's lifespan and are offset by the 5% discount rate. As such, any change in the lifespan of the project would have a near-linear impact on the NPV of the project.

The sensitivity analysis showed that the higher the number in absolute terms, the more the profitability index is sensitive to that underlying variable (i.e. stronger slope); and a positive value indicates a positive relationship between the variable and the profitability metric (i.e. positive slope) whereas a negative number indicates the opposite (i.e. negative slope). This analysis (Table 9) shows that the profitability of this project is affected primarily by (1) the price and quantity of biodiesel produced (Figure 14), (2) the percentage of N and P removal savings passed on to a private company (Figure 15), and (3) operational costs (OpEx) (Figure 16). Understandably, capital costs (CapEx) have a smaller impact on the profitability of the project since they are only incurred once and are amortized over the lifespan of the project. However, the provincial electricity price and the quantity of methane produced had little effect on the profitability of the project because the methane generated is combusted to produce electricity and that revenues and/or cost savings generated from that electricity account for approximately 4.2% of the FWC system's annual revenues.

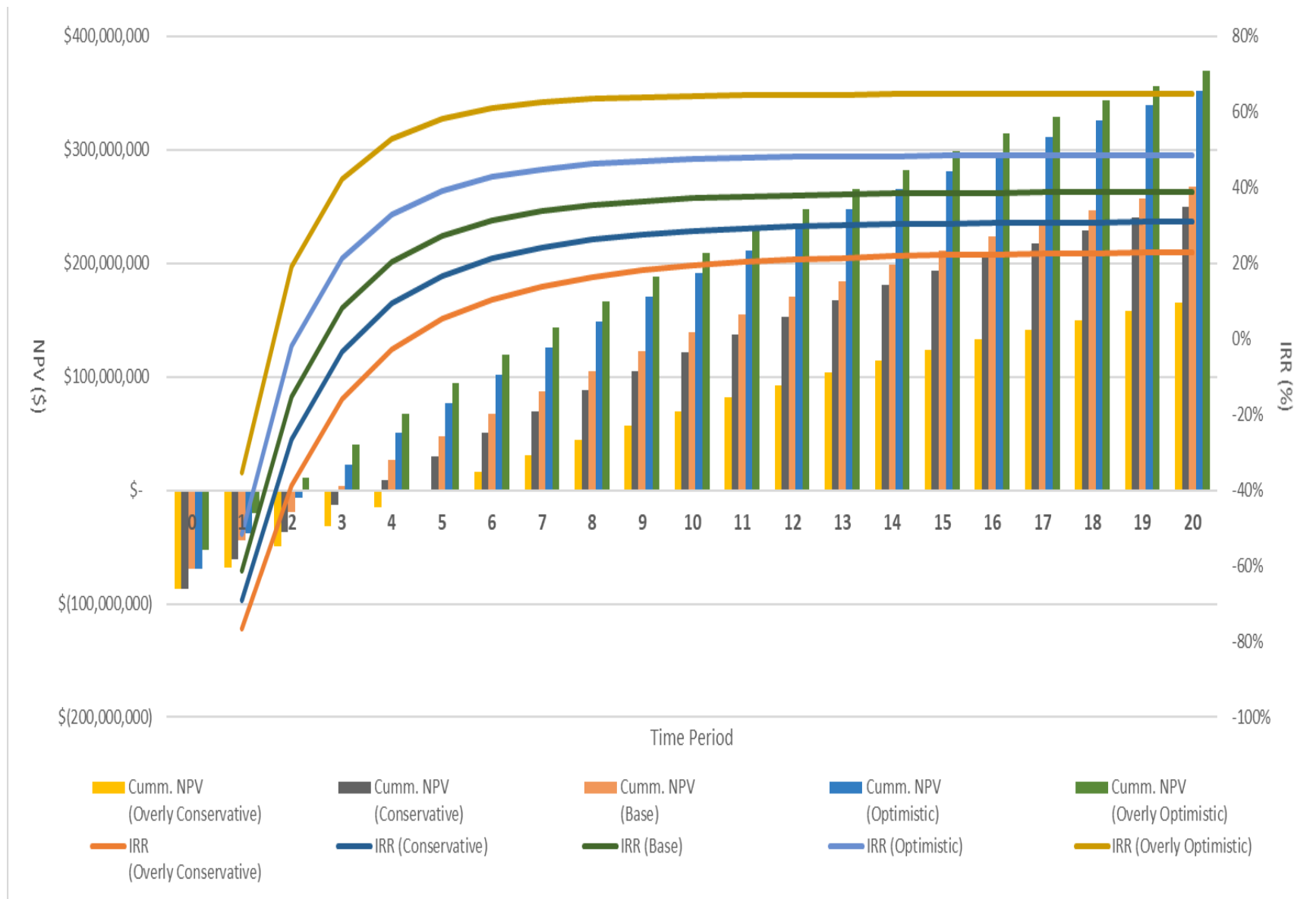


Figure 13 NPV & IRR of different scenarios over lifespan of the project

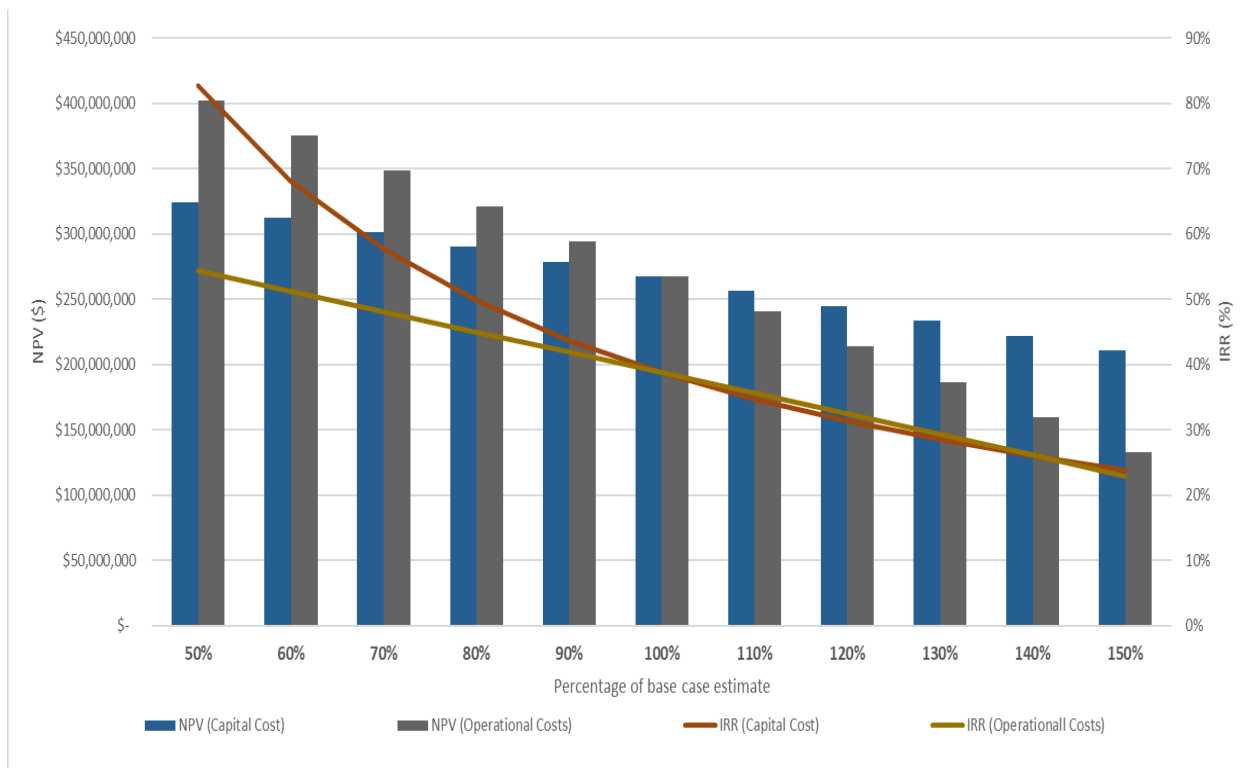


Figure 14: Base Case NPV & IRR sensitivity to changes in key revenue variables

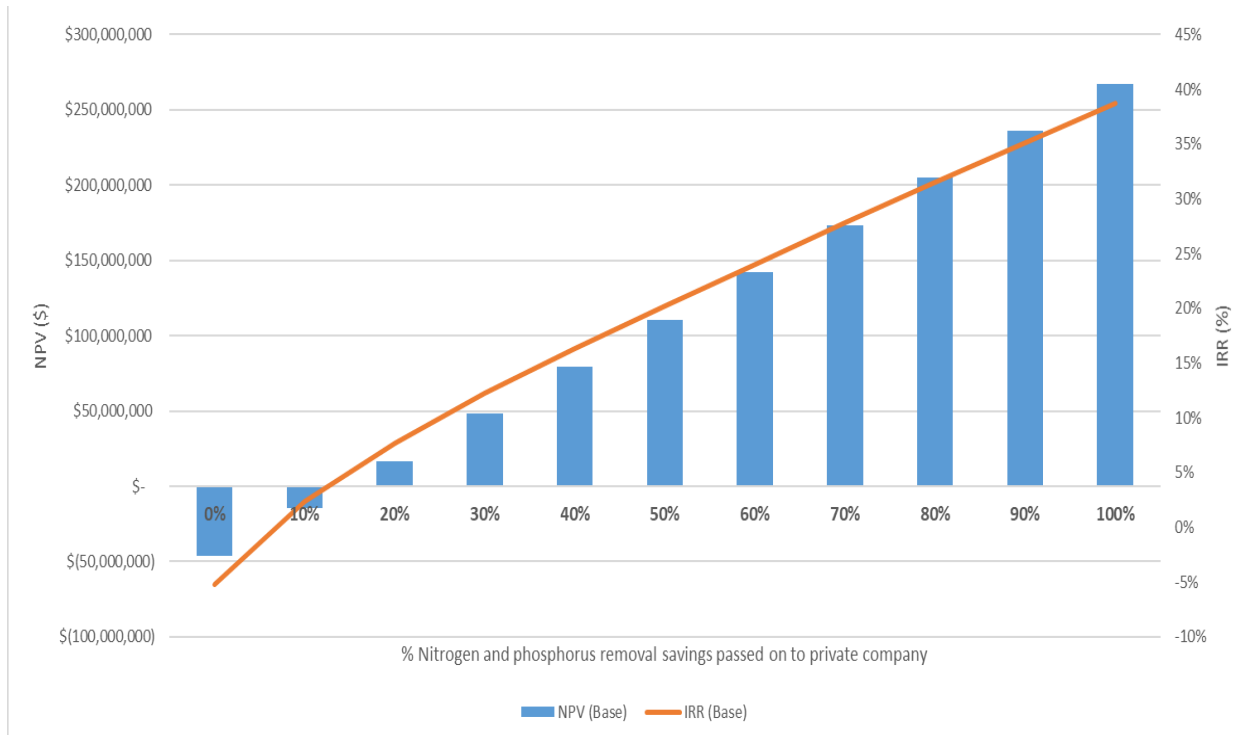


Figure 15: Base Case NPV & IRR as a function of % Of nitrogen and phosphorus removal savings passed on to a private company

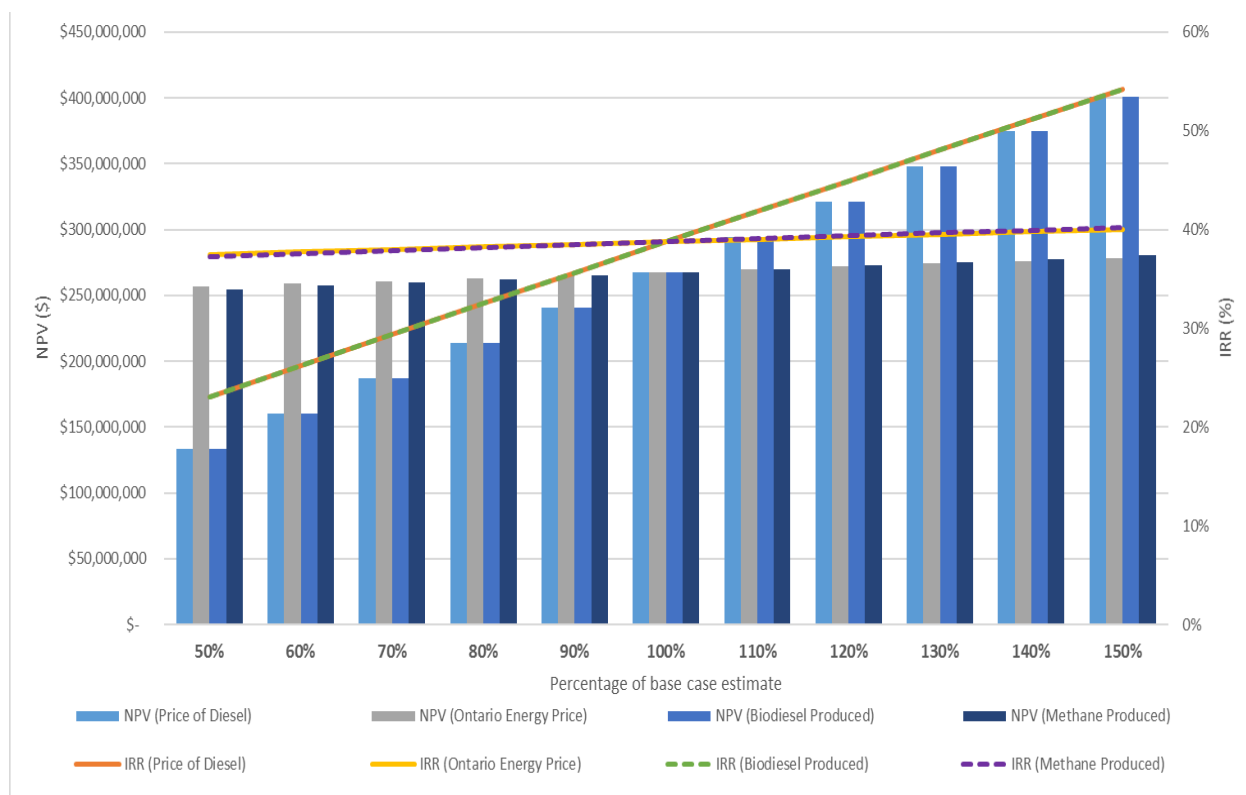


Figure 16: Base Case NPV & IRR sensitivity to changes in key cost variables

Table 9: Base Case NPV & IRR sensitivity to changes in key cost variables

	<i>NPV</i>	<i>IRR</i>
<i>Of N & P removal savings passed on to a private company</i>	313466736	0.396
<i>Price of Diesel</i>	267510557	0.311
<i>Ontario Energy price</i>	21856125	0.025
<i>Biodiesel Produced</i>	267510557	0.311
<i>Capital Costs</i>	-113100559	-0.541
<i>Operational Costs</i>	-269315606	-0.313
<i>Methane produced</i>	25533959	0.030

3.3 Outlook and limitations of the case study

Given the innovative nature of the FWC system, there are no comparable facilities in operation or proposed as study cases in the scientific literature. Therefore, the financial analysis should be interpreted cautiously, as it is a best guess estimate of the costs and revenues associated with this specific system. However, each component of the FWC system was studied in detail and economic

outcomes were already determined. Given that most of the parameters in my financial model were determined by selecting conservative data from the literature, it is reasonable to assume that the results are robust. In addition, a deviation in the base case that increased costs and reduced revenues by 25% (i.e., the overly conservative scenario) would not make the FWC system unprofitable. This confirms the reliability of the profitability results.

The results of the sensitivity analysis are consistent with existing literature that also identifies the price and quantity of biodiesel sold, CapEx and OpEx as being some the largest variables affecting the profitability of biodiesel production from algae (Banerjee & Ramaswamy, 2017; Hise et al., 2016; Richardson et al., 2014; Xin et al., 2016; Zamalloa et al., 2011). The caveat to this however is that no other available study considers the parameter that has the largest impact on the profitability of the system, namely the “% of N & P removal savings passed on to a private company”. The only comparable to my thesis is a 2014 study published by Orfield et al. which found that, similar to my results, biological oxygen demand (BOD) removal credits were a key parameter affecting the financial viability of algal bio-oil production potential through flue gas and wastewater co-utilization (Orfield et al., 2014). As shown in Figure 13, including these cost savings in the valuation of the project is essential to generating a positive return on investment, and as such a higher degree of precision on this parameter is required.

Conclusions and future directions

My thesis presents a novel approach to assessing the financial viability of a FWC system which incorporates cost savings generated for the wastewater treatment plant. All major aspects of the FWC system had to first be defined including its location, the cultivation method used, the sourcing of nutrients and CO₂, the selection of the optimal microalgae to grow, the biomass harvesting and dewatering processes used, and finally the conversion pathways that would be employed to convert the algal biomass to biodiesel. Then, a financial model was built in Excel to assess the profitability of the system using parameters primarily estimated from academic literature in combination with online research and industry contacts and complemented by scenario-based and sensitivity-based analysis.

The results found by this study point towards the financial viability of microalgal biodiesel production in Canada when paired with the phytoremediation of wastewater and the combustion of biogas generated from the anaerobic digestion of lipid-extracted algae (LEA). However, this type of project isn't suited for private businesses and investors. Figure 13 shows that the profitability of the FWC system for a private company is highly sensitive to the percentage of N & P removal cost savings generated for the wastewater treatment plant that can be earned as income for the private company. In the likely scenario that the business is able to charge the treatment plant for 50% of the cost savings generated, it would see the NPV of the project decrease by 58% to \$110.78M CAD (down from \$267.51M CAD) which, given the estimated capital costs of \$69.68M CAD, is significantly less attractive given the operational and logistical risks associated with running such a project over its 20-year lifespan. Even assuming that government run wastewater treatment plants are willing to pay for 100% of cost savings generated, a risk averse

private investor would be reticent to invest in a infrastructure project that generates a low investment multiple (base case 3.8x) over a long period of time (lifespan of 20 years). However, government run wastewater treatment plants would not face any of these issues since they would incur 100% of the cost savings generated and do not require high returns on investment. In addition, government investors would benefit from a plethora of non-financial benefits such as a decreased reliance on fossil fuels, an improved public image and helping to achieve their GHG emission reduction targets. The results of this study therefore indicate that the FWC system would be, at a first glance, incredibly attractive for the government.

This study however is not exhaustive, and a great deal of additional research and work would have to be conducted prior to a system like the one proposed here seeing widespread implementation. Firstly, the financial modelling would have to be refined by increasing the number and precision of the underlying parameters. A more extensive financial analysis could then be conducted to include more financial performance metrics, better detailed scenarios, and a multi-parameter sensitivity analysis. Secondly, a life-cycle analysis (LCA) would have to be conducted to include the economic and environmental impacts of the FWC system. Subsequently, a wastewater treatment plant would have to be selected, the technical feasibility of integrating a system like this would then be assessed, and supply chains would have to be created. Finally, once the pilot facility has been built and the business model sufficiently proven to be profitable, further studies would have to be conducted to evaluate other areas in Canada where such a system could be implemented, government policies and incentives programs would have to be drafted, and nationwide LCA analyses would have to be conducted.

References

- Abdelaziz, A. E. M., Leite, G. B., Belhaj, M. A., & Hallenbeck, P. C. (2014). Screening microalgae native to Quebec for wastewater treatment and biodiesel production. *Bioresource Technology*, 157, 140–148. <https://doi.org/10.1016/j.biortech.2014.01.114>
- Abdo, S. M., Ahmed, E., El-enin, S. A., Din, R. S. El, Diwani, G. El, & Ali, G. (2014). Qualitative and quantitative determination of lipid content in microalgae for biofuel production. *Journal of Algal Biomass Utilization*, 5(3), 23–28.
- Acien, F. G., Fernández, J. M., Magán, J. J., & Molina, E. (2012). Production cost of a real microalgae production plant and strategies to reduce it. *Biotechnology Advances*, 30(6), 1344–1353. <https://doi.org/10.1016/j.biotechadv.2012.02.005>
- AFDC. (2014). Fuel Properties Comparison. Retrieved from http://www.afdc.energy.gov/fuels/fuel_properties.php
- Alamgir, A., Khan, M. A., & Shaukat, S. S. (2016). Algal growth and waste stabilization ponds performance efficiency in a sub-tropical climate. *Pakistan Journal of Botany*, 48(1), 377–385.
- Alternative Fuels Data Center. (2015). Alternative Fuel Price Report. Retrieved from <http://www.afdc.energy.gov/fuels/prices.html>
- Amer, L., Adhikari, B., & Pellegrino, J. (2011). Technoeconomic analysis of five microalgae-to-biofuels processes of varying complexity. *Bioresource Technology*, 102(20), 9350–9359. <https://doi.org/10.1016/j.biortech.2011.08.010>
- American Biogas Council. (2016). Operational Biogas Systems in the U.S. Retrieved December 12, 2016, from https://www.americanbiogascouncil.org/biogas_maps.asp
- Amin, S. (2009). Review on biofuel oil and gas production processes from microalgae. *Energy Conversion and Management*, 50(7), 1834–1840. <https://doi.org/10.1016/j.enconman.2009.03.001>
- Anonymous. (2017). Data from three wastewater treatment plants in southern alberta. Retrieved

November 28, 2016, from Personal Communication

Aqua Calc. (n.d.). Volume to Weight conversions for common substances and materials.

Retrieved March 9, 2018, from <https://www.aqua-calc.com/calculate/volume-to-weight>

Austin, A. (2010). Open Ponds Versus Closed Bioreactors. *Biomass Magazine*, 4. Retrieved from <http://biomassmagazine.com/articles/3618/open-ponds-versus-closed--bioreactors>

Azcan, N., & Danisman, A. (2007). Alkali catalyzed transesterification of cottonseed oil by microwave irradiation. *Fuel*, 86(17–18), 2639–2644.
<https://doi.org/10.1016/j.fuel.2007.05.021>

Balasubramanian, R. K., Yen Doan, T. T., & Obbard, J. P. (2013). Factors affecting cellular lipid extraction from marine microalgae. *Chemical Engineering Journal*, 215–216, 929–936.
<https://doi.org/10.1016/j.cej.2012.11.063>

Banerjee, S., & Ramaswamy, S. (2017). Dynamic process model and economic analysis of microalgae cultivation in open raceway ponds. *Algal Research*, 26, 330–340.
<https://doi.org/10.1016/j.algal.2017.08.011>

Barlow, J., Sims, R. C., & Quinn, J. C. (2016). Techno-economic and life-cycle assessment of an attached growth algal biorefinery. *Bioresource Technology*, 220, 360–368.
<https://doi.org/10.1016/j.biortech.2016.08.091>

Barnard, T. M., Leadbeater, N. E., Boucher, M. B., Stencel, L. M., & Wilhite, B. A. (2007). Continuous-flow preparation of biodiesel using microwave heating. *Energy and Fuels*, 21(3), 1777–1781. <https://doi.org/10.1021/ef0606207>

Barry, A., Wolfe, A., English, C., Ruddick, C., & Lambert, D. (2016). 2016 National Algal Biofuels Technology Review. *U.S. Department of Energy*, (June).
<https://doi.org/10.2172/1259407>

Bashaar, Y. A. (2004). Nutrients requirements in biological industrial wastewater treatment. *African Journal of Biotechnology*, 3(4), 236–238. <https://doi.org/10.5897/AJB2004.000-2042>

Batten, D., Beer, T., Freischmidt, G., Grant, T., Liffman, K., Paterson, D., ... Threlfall, G.

- (2013). Using wastewater and high-rate algal ponds for nutrient removal and the production of bioenergy and biofuels. *Water Science and Technology*, 67(4), 915–924.
<https://doi.org/10.2166/wst.2012.618>
- Bayat Tork, M., Khalilzadeh, R., & Kouchakzadeh, H. (2017). Efficient harvesting of marine *Chlorella vulgaris* microalgae utilizing cationic starch nanoparticles by response surface methodology. *Bioresource Technology*, 243, 583–588.
<https://doi.org/10.1016/j.biortech.2017.06.181>
- Beach, E. S., Eckelman, M. J., Cui, Z., Brentner, L., & Zimmerman, J. B. (2012). Preferential technological and life cycle environmental performance of chitosan flocculation for harvesting of the green algae *Neochloris oleoabundans*. *Bioresource Technology*, 121, 445–449. <https://doi.org/10.1016/j.biortech.2012.06.012>
- Beal, C. M., Stillwell, A. S., King, C. W., Cohen, S. M., Berberoglu, H., Bhattarai, R. P., ... Hebner, R. E. (2012). Energy Return on Investment for Algal Biofuel Production Coupled with Wastewater Treatment. *Water Environment Research*, 84(9), 692–710.
<https://doi.org/10.2175/106143012X13378023685718>
- Becker, E. (1994). *Microalgae: biotechnology and microbiology*. cambridge: Cambridge University Press. Retrieved from
https://books.google.ca/books?hl=en&lr=&id=KAKx4I7NWEYC&oi=fnd&pg=PA1&ots=028rZLKXjS&sig=SbZCf7BQf3NRks6Eov2Uv8hVJZk&redir_esc=y#v=onepage&q&f=false
- Bennamoun, L., Afzal, M. T., & Léonard, A. (2015). Drying of alga as a source of bioenergy feedstock and food supplement - A review. *Renewable and Sustainable Energy Reviews*, 50, 1203–1212. <https://doi.org/10.1016/j.rser.2015.04.196>
- Bilad, M. R., Vandamme, D., Foubert, I., Muylaert, K., & Vankelecom, I. F. J. (2012). Harvesting microalgal biomass using submerged microfiltration membranes. *Bioresource Technology*, 111, 343–352. <https://doi.org/10.1016/j.biortech.2012.02.009>
- Biller, P., & Ross, A. B. (2011). Potential yields and properties of oil from the hydrothermal liquefaction of microalgae with different biochemical content. *Bioresource Technology*,

102(1), 215–225. <https://doi.org/10.1016/j.biortech.2010.06.028>

Biodiesel Magazine. (2016). Biodiesel plants map. Retrieved October 1, 2016, from

<http://www.biodieselmagazine.com/plants/map/>

Bjornsson, W. J., MacDougall, K. M., Melanson, J. E., O’Leary, S. J. B., & McGinn, P. J.

(2012). Pilot-scale supercritical carbon dioxide extractions for the recovery of triacylglycerols from microalgae: A practical tool for algal biofuels research. *Journal of Applied Phycology*, 24(3), 547–555. <https://doi.org/10.1007/s10811-011-9756-2>

Bligh, E. ., & Dyer, W. . (1959). A Rapid method of total lipid extraction and purification.

Canadian Journal of Biochemistry and Physiology, 37(8), 911–917.

Bohutskyi, P., Kula, T., Kessler, B. A., Hong, Y., Bouwer, E. J., Betenbaugh, M. J., & Allnutt, F.

C. T. (2014). Mixed Trophic State Production Process for Microalgal Biomass with High Lipid Content for Generating Biodiesel and Biogas. *Bioenergy Research*, 7(4), 1174–1185. <https://doi.org/10.1007/s12155-014-9453-5>

Boldor, D., Kanitkar, A., Terigar, B. G., Leonardi, C., Lima, M., & Breitenbeck, G. A. (2010).

Microwave assisted extraction of biodiesel feedstock from the seeds of invasive chinese tallow tree. *Environmental Science and Technology*, 44(10), 4019–4025.

<https://doi.org/10.1021/es100143z>

Brandenberger, M., Matzenberger, J., Vogel, F., & Ch, L. (2013). Producing synthetic natural

gas from microalgae via supercritical water gasification : A techno-economic sensitivity analysis, 1–9.

Bratby, J. (2004). *Coagulation and Flocculation in Water and Wastewater Treatment* (2nd ed.).

London: IWA publishing. Retrieved from

https://books.google.ca/books?hl=en&lr=&id=dIzbCwAAQBAJ&oi=fnd&pg=PR5&ots=GHdbk1_02Q&sig=rz-wi01I8fBuGfhhb9SKtzBtyFKw&redir_esc=y#v=onepage&q&f=false

Breil, C., Abert Vian, M., Zemb, T., Kunz, W., & Chemat, F. (2017). “Bligh and Dyer” and

Folch methods for solid–liquid–liquid extraction of lipids from microorganisms.

Comprehension of solvation mechanisms and towards substitution with alternative solvents. *International Journal of Molecular Sciences*, 18(4), 1–21.

<https://doi.org/10.3390/ijms18040708>

- Brennan, L., & Owende, P. (2010). Biofuels from microalgae-A review of technologies for production, processing, and extractions of biofuels and co-products. *Renewable and Sustainable Energy Reviews*, 14(2), 557–577. <https://doi.org/10.1016/j.rser.2009.10.009>
- Brownbridge, G., Azadi, P., Smallbone, A., Bhave, A., Taylor, B., & Kraft, M. (2014). The future viability of algae-derived biodiesel under economic and technical uncertainties. *Bioresource Technology*, 151, 166–173. <https://doi.org/10.1016/j.biortech.2013.10.062>
- Camacho Rubio, F., Sánchez Mirón, A., Cerón García, M. C., García Camacho, F., Molina Grima, E., & Chisti, Y. (2004). Mixing in bubble columns: a new approach for characterizing dispersion coefficients. *Chemical Engineering Science*, 59(20), 4369–4376. <https://doi.org/10.1016/j.ces.2004.06.037>
- Canada Statistics, C. (2017). Value per acre of farm land and buildings, at July 1. Retrieved February 21, 2018, from <http://www5.statcan.gc.ca/cansim/a26?lang=eng&id=20003>
- Canada Water and Wastewater Association. (2001). National Survey of Wastewater Treatment Plants. Retrieved September 26, 2016, from http://www.cwwa.ca/pdf_files/freepub_wastewaterreport.PDF
- Canadian Government. (2011). 2009 Water Use Summary Database. Retrieved September 21, 2016, from https://www.ec.gc.ca/eau-water/ED0E12D7-1C3B-4658-8833-347B527C688A/MWWS_2009_Water_Use_Summary_Database_EN_publishedOct2011.xls
- Caputo, G., Rubio, P., Scargiali, F., Marotta, G., & Brucato, A. (2016). Supercritical water gasification of microalgae and their constituents in a continuous reactor. *The Journal of Supercritical Fluids*, 107, 450–461. <https://doi.org/10.1016/j.supflu.2015.09.022>
- CDIAC. (2014). Global CO2 emissions from 1751 to 2013. Retrieved from <http://www.statista.com/statistics/264699/worldwide-co2-emissions/>
- Centre, H., & Office, M. (2013). International symposium on the stabilisation of greenhouse gases. Retrieved from papers2://publication/uuid/F9A1E5A7-A145-4BBB-8A57-5E3BABF936D7

- Chatsungnoen, T., & Chisti, Y. (2016). Harvesting microalgae by flocculation-sedimentation. *Algal Research*, 13, 271–283. <https://doi.org/10.1016/j.algal.2015.12.009>
- Chen, C. L., Huang, C. C., Ho, K. C., Hsiao, P. X., Wu, M. S., & Chang, J. S. (2015). Biodiesel production from wet microalgae feedstock using sequential wet extraction/transesterification and direct transesterification processes. *Bioresource Technology*, 194, 179–186. <https://doi.org/10.1016/j.biortech.2015.07.021>
- Cheng, C.-H., Du, T.-B., Pi, H.-C., Jang, S.-M., Lin, Y.-H., & Lee, H.-T. (2011). Comparative study of lipid extraction from microalgae by organic solvent and supercritical CO₂. *Bioresource Technology*, 102(21), 10151–10153. <https://doi.org/10.1016/j.biortech.2011.08.064>
- Cherp, A., Vinichenko, V., Jewell, J., Suzuki, M., & Antal, M. (2017). Comparing electricity transitions: A historical analysis of nuclear, wind and solar power in Germany and Japan. *Energy Policy*, 101, 612–628. <https://doi.org/10.1016/j.enpol.2016.10.044>
- Chiaramonti, D., Oasmaa, A., & Solantausta, Y. (2007). Power generation using fast pyrolysis liquids from biomass. *Renewable and Sustainable Energy Reviews*, 11(6), 1056–1086. <https://doi.org/10.1016/j.rser.2005.07.008>
- Chiaramonti, D., Prussi, M., Buffi, M., Casini, D., & Rizzo, A. M. (2015). Thermochemical Conversion of Microalgae: Challenges and Opportunities. *Energy Procedia*, 75, 819–826. <https://doi.org/10.1016/j.egypro.2015.07.142>
- Chisti, Y. (2008). Biodiesel from microalgae beats bioethanol. *Trends in Biotechnology*, 26(3), 126–131. <https://doi.org/10.1016/j.tibtech.2007.12.002>
- Chisti, Y. (2016). Large-Scale Production of Algal Biomass: Raceway Ponds. *Green Energy and Technology*. <https://doi.org/10.1007/978-3-319-12334-9>
- Christenson, L., & Sims, R. (2011). Production and harvesting of microalgae for wastewater treatment, biofuels, and bioproducts. *Biotechnology Advances*, 29(6), 686–702. <https://doi.org/10.1016/j.biotechadv.2011.05.015>
- Clarke Energy. (n.d.). CHP efficiency for biogas. Retrieved March 9, 2018, from <https://www.clarke-energy.com/2013/chp-cogen-efficiency-biogas/>

- Converti, A., Casazza, A. A., Ortiz, E. Y., Perego, P., & Del Borghi, M. (2009). Effect of temperature and nitrogen concentration on the growth and lipid content of *Nannochloropsis oculata* and *Chlorella vulgaris* for biodiesel production. *Chemical Engineering and Processing: Process Intensification*, 48(6), 1146–1151.
<https://doi.org/10.1016/j.cep.2009.03.006>
- Cooney, M., Young, G., & Nagle, N. (2009). Extraction of bio-oils from microalgae. *Separation and Purification Reviews*, 38(4), 291–325. <https://doi.org/10.1080/15422110903327919>
- Dai, Y. M., Chen, K. T., & Chen, C. C. (2014). Study of the microwave lipid extraction from microalgae for biodiesel production. *Chemical Engineering Journal*, 250, 267–273.
<https://doi.org/10.1016/j.cej.2014.04.031>
- Dassey, A. J., & Theegala, C. S. (2013). Harvesting economics and strategies using centrifugation for cost effective separation of microalgae cells for biodiesel applications. *Bioresource Technology*, 128, 241–245. <https://doi.org/10.1016/j.biortech.2012.10.061>
- Davis, R., Aden, A., & Pienkos, P. T. (2011). Techno-economic analysis of autotrophic microalgae for fuel production. *Applied Energy*, 88(10), 3524–3531.
<https://doi.org/10.1016/j.apenergy.2011.04.018>
- Davis, R. E., Fishman, D. B., Frank, E. D., Johnson, M. C., Jones, S. B., Kinchin, C. M., ... Wigmosta, M. S. (2014). Integrated evaluation of cost, emissions, and resource potential for algal biofuels at the national scale. *Environmental Science and Technology*, 48(10), 6035–6042. <https://doi.org/10.1021/es4055719>
- de Gorter, H., & Drabik, D. (2016). Biofuel policies and the impact of developing countries' policy responses to the 2007–2008 food price boom. *Global Food Security*, 11, 64–71.
<https://doi.org/10.1016/j.gfs.2016.07.008>
- Delrue, F., Li-Beisson, Y., Setier, P. A., Sahut, C., Roubaud, A., Froment, A. K., & Peltier, G. (2013). Comparison of various microalgae liquid biofuel production pathways based on energetic, economic and environmental criteria. *Bioresource Technology*, 136, 205–212.
<https://doi.org/10.1016/j.biortech.2013.02.091>
- Delrue, F., Setier, P. A., Sahut, C., Cournac, L., Roubaud, A., Peltier, G., & Froment, A. K.

- (2012). An economic, sustainability, and energetic model of biodiesel production from microalgae. *Bioresource Technology*, 111, 191–200.
<https://doi.org/10.1016/j.biortech.2012.02.020>
- Demmig-Adams, B., & Adams, W. W. (2003). PHOTOSYNTHESIS AND PARTITIONING | Photoinhibition. *Encyclopedia of Applied Plant Sciences*, 707–714.
<https://doi.org/10.1016/B0-12-227050-9/00091-0>
- Desmorieux, H., & Decaen, N. (2005). Convective drying of spirulina in thin layer. *Journal of Food Engineering*, 66(4), 497–503. <https://doi.org/10.1016/j.jfoodeng.2004.04.021>
- DOE/EIA. (2016). International Energy Outlook 2016. Retrieved February 24, 2017, from <https://www.eia.gov/outlooks/ieo/world.cfm>
- Doshi, A., Pascoe, S., Cogle, L., & Rainey, T. J. (2016). Economic and policy issues in the production of algae-based biofuels: A review. *Renewable and Sustainable Energy Reviews*, 64, 329–337. <https://doi.org/10.1016/j.rser.2016.06.027>
- Doucha, J., & Lívanský, K. (2008). Influence of processing parameters on disintegration of Chlorella cells in various types of homogenizers. *Applied Microbiology and Biotechnology*, 81(3), 431–440. <https://doi.org/10.1007/s00253-008-1660-6>
- Ehimen, E. A., Sun, Z. F., Carrington, C. G., Birch, E. J., & Eaton-Rye, J. J. (2011). Anaerobic digestion of microalgae residues resulting from the biodiesel production process. *Applied Energy*, 88(10), 3454–3463. <https://doi.org/10.1016/j.apenergy.2010.10.020>
- Enerdata. (2016). Global Energy Statistical Yearbook 2016: Total Energy Consumption. Retrieved February 24, 2017, from <https://yearbook.enerdata.net/#energy-consumption-data.html>
- Energy Information Administration. (2018). Gasoline and Diesel Fuel Update. Retrieved July 1, 2018, from <https://www.eia.gov/petroleum/gasdiesel/>
- Enriquez, S., Duarte, C., & Sand-Jensen, K. (1993). Patterns in decomposition rates among photosynthetic organisms: the importance of detritus C: N: P content. *Oecologia*, 94(4), 457–471. <https://doi.org/10.1007/BF00566960>

- Environment and Climate Change Canada. (2016a). Historical greenhouse gas emissions and projections to 2030 with policies and measures as of November 1, 2016, Canada, 2005 to 2030. Retrieved October 5, 2017, from <https://www.ec.gc.ca/indicateurs-indicators/default.asp?lang=en&n=CCED3397-1>
- Environment and Climate Change Canada. (2016b). *Pan-Canadian Framework on Clean Growth and Climate Change*. Retrieved from <https://www.canada.ca/en/services/environment/weather/climatechange/pan-canadian-framework.html>
- Environment Canada. (2017). Average temperature map of Canada – December to February (Winter). Retrieved September 16, 2017, from https://weather.gc.ca/saisons/clim_e.html
- EPA. Renewable Fuels: Regulations & Standards, Environmental Protection Agency § (2012). Retrieved from <http://www.epa.gov/otaq/fuels/renewablefuels/regulations.htm>
- Farm Credit Canada. (2017). *Farmland Values Report 2016*. Retrieved from <https://www.fcc-fac.ca/content/dam/fcc/about-fcc/reports/farmland-values-report-2016.pdf>
- Ferrell, J., & Sarisky-Reed, V. (2010). National Algal Biofuels Technology Roadmap. *U.S. Department of Energy*, (May), 140. [https://doi.org/Public Law No. 106-554](https://doi.org/Public%20Law%20No.%20106-554)
- Fondriest Environmental, I. (2014). Water Temperature.” *Fundamentals of Environmental Measurements*. Retrieved October 20, 2017, from <http://www.fondriest.com/environmental-measurements/parameters/water-quality/water-temperature/#watertemp17>
- Fulton, L. M., Lynd, L. R., Körner, A., Greene, N., & Tonachel, L. R. (2015). The need for biofuels as part of a low carbon energy future. *Biofuels, Bioproducts and Biorefining*, 9(5), 476–483. <https://doi.org/10.1002/bbb.1559>
- Gabriel Acien Fernandez, F., González-López, C. V., Fernández Sevilla, J. M., & Molina Grima, E. (2012). Conversion of CO₂ into biomass by microalgae: How realistic a contribution may it be to significant CO₂ removal? *Applied Microbiology and Biotechnology*, 96(3), 577–586. <https://doi.org/10.1007/s00253-012-4362-z>
- García, J., Green, B. F., Lundquist, T., Mujeriego, R., Hernández-Mariné, M., & Oswald, W. J. (2006). Long term diurnal variations in contaminant removal in high rate ponds treating

- urban wastewater. *Bioresource Technology*, 97(14), 1709–1715.
<https://doi.org/10.1016/j.biortech.2005.07.019>
- Geciova, J., Bury, D., & Jelen, P. (2002). Methods for disruption of microbial cells for potential use in the dairy industry - a review. *International Dairy Journal*, 12(6), 541–553.
[https://doi.org/10.1016/S0958-6946\(02\)00038-9](https://doi.org/10.1016/S0958-6946(02)00038-9)
- Ghasemi Naghdi, F., González González, L. M., Chan, W., & Schenk, P. M. (2016). Progress on lipid extraction from wet algal biomass for biodiesel production. *Microbial Biotechnology*, 9(6), 718–726. <https://doi.org/10.1111/1751-7915.12360>
- Goyal, H. B., Seal, D., & Saxena, R. C. (2008). Bio-fuels from thermochemical conversion of renewable resources: A review. *Renewable and Sustainable Energy Reviews*, 12(2), 504–517. <https://doi.org/10.1016/j.rser.2006.07.014>
- Greenfuels. (2016). Industry map. Retrieved September 30, 2016, from <http://greenfuels.org/industry/industry-map/>
- Grima, E. M., González, M. J. I., & Giménez, A. G. (2013). *Algae for Biofuels and Energy*. *Algae for Biofuels and* <https://doi.org/10.1007/978-94-007-5479-9>
- Grobbelaar, J. U. (2010). Microalgal biomass production: challenges and realities. *Photosynthesis Research*, 106(1–2), 135–144. <https://doi.org/10.1007/s11120-010-9573-5>
- Guiry, M. D. (2012). How many species of algae are there? *Journal of Phycology*, 48(5), 1057–1063. <https://doi.org/10.1111/j.1529-8817.2012.01222.x>
- Guldhe, A., Singh, B., Rawat, I., Ramluckan, K., & Bux, F. (2014). Efficacy of drying and cell disruption techniques on lipid recovery from microalgae for biodiesel production. *Fuel*, 128, 46–52. <https://doi.org/10.1016/j.fuel.2014.02.059>
- Hallenbeck, P. C., Leite, G. B., & Abdelaziz, A. E. M. (2014). Exploring the diversity of microalgal physiology for applications in wastewater treatment and biofuel production. *Algal Research*, 6(PA), 111–118. <https://doi.org/10.1016/j.algal.2014.09.006>
- Hannon, M., Gimpel, J., Tran, M., Rasala, B., & Mayfield, S. (2010). Biofuels from algae: challenges and potential. *Biofuels*, 1(5), 763–784. <https://doi.org/10.4155/bfs.10.44>

- Hansel, P. A., Guy Riefler, R., & Stuart, B. J. (2014). Efficient flocculation of microalgae for biomass production using cationic starch. *Algal Research*, 5(1), 133–139.
<https://doi.org/10.1016/j.algal.2014.07.002>
- Harrison, S. T. L. (1991). Bacterial cell disruption: a key unit operation in the recovery of intracellular products. *Biotechnology Advances*, 9, 217–240.
- Henderson, R., Parsons, S. A., & Jefferson, B. (2008). The impact of algal properties and pre-oxidation on solid-liquid separation of algae. *Water Research*, 42(8–9), 1827–1845.
<https://doi.org/10.1016/j.watres.2007.11.039>
- Hernández-Calderón, O. M., Ponce-Ortega, J. M., Ortiz-Del-Castillo, J. R., Cervantes-Gaxiola, M. E., Milán-Carrillo, J., Serna-González, M., & Rubio-Castro, E. (2016). Optimal Design of Distributed Algae-Based Biorefineries Using CO₂ Emissions from Multiple Industrial Plants. *Industrial and Engineering Chemistry Research*, 55(8), 2345–2358.
<https://doi.org/10.1021/acs.iecr.5b01684>
- Hernando, J., Leton, P., Matia, M. P., Novella, J. L., & Alvarez-Builla, J. (2007). Biodiesel and FAME synthesis assisted by microwaves: Homogeneous batch and flow processes. *Fuel*, 86(10–11), 1641–1644. <https://doi.org/10.1016/j.fuel.2006.11.003>
- Hillebrand, H., & Sommer, U. (1999). The nutrient stoichiometry of benthic microalgal growth: Redfield proportions are optimal. *Limnology and Oceanography*, 44(2), 440–446.
<https://doi.org/10.4319/lo.1999.44.2.0440>
- Hise, A. M., Characklis, G. W., Kern, J., Gerlach, R., Viamajala, S., Gardner, R. D., & Vadlamani, A. (2016). Evaluating the relative impacts of operational and financial factors on the competitiveness of an algal biofuel production facility. *Bioresource Technology*, 220, 271–281. <https://doi.org/10.1016/j.biortech.2016.08.050>
- Investopedia. (2017a). Capital Expenditure (CAPEX). Retrieved February 18, 2018, from <https://www.investopedia.com/terms/c/capitalexpenditure.asp>
- Investopedia. (2017b). Internal Rate Of Return - IRR. Retrieved December 28, 2017, from <https://www.investopedia.com/terms/i/irr.asp>
- Investopedia. (2017c). Net Present Value - NPV. Retrieved December 26, 2017, from

<https://www.investopedia.com/terms/n/npv.asp>

Investopedia. (2017d). Operating Expense (OpEx). Retrieved February 18, 2018, from https://www.investopedia.com/terms/o/operating_expense.asp

Investopedia. (2017e). Payback Period. Retrieved April 12, 2018, from <https://www.investopedia.com/terms/p/paybackperiod.asp>

Investopedia. (2017f). Sensitivity Analysis. Retrieved December 29, 2017, from <https://www.investopedia.com/terms/s/sensitivityanalysis.asp>

Investopedia. (2017g). Time Value of Money - TVM. Retrieved December 26, 2017, from <https://www.investopedia.com/terms/t/timevalueofmoney.asp>

Johnson, M. B., & Wen, Z. (2009). Production of biodiesel fuel from the microalga *schizochytrium limacinum* by direct transesterification of algal biomass. *Energy and Fuels*, 23(10), 5179–5183. <https://doi.org/10.1021/ef900704h>

Kang, Z., Kim, B. H., Ramanan, R., Choi, J. E., Yang, J. W., Oh, H. M., & Kim, H. S. (2015). A cost analysis of microalgal biomass and biodiesel production in open raceways treating municipal wastewater and under optimum light wavelength. *Journal of Microbiology and Biotechnology*, 25(1), 109–118. <https://doi.org/10.4014/jmb.1409.09019>

Khanal, S. K., Grewell, D., Sung, S., & Van Leeuwen, J. (2007). Ultrasound applications in wastewater sludge pretreatment: A review. *Critical Reviews in Environmental Science and Technology*, 37(4), 277–313. <https://doi.org/10.1080/10643380600860249>

Khoo, H. H., Sharratt, P. N., Das, P., Balasubramanian, R. K., Naraharisetti, P. K., & Shaik, S. (2011). Life cycle energy and CO₂ analysis of microalgae-to-biodiesel: Preliminary results and comparisons. *Bioresource Technology*, 102(10), 5800–5807. <https://doi.org/10.1016/j.biortech.2011.02.055>

Kies, A., Schyska, B. U., & Von Bremen, L. (2016). The demand side management potential to balance a highly renewable European power system. *Energies*, 9(11), 1–15. <https://doi.org/10.3390/en9110955>

Klinthong, W., Yang, Y. H., Huang, C. H., & Tan, C. S. (2015). A Review: Microalgae and their

- applications in CO₂ capture and renewable energy. *Aerosol and Air Quality Research*, 15(2), 712–742. <https://doi.org/10.4209/aaqr.2014.11.0299>
- Knuckey, R. M., Brown, M. R., Robert, R., & Frampton, D. M. F. (2006). Production of microalgal concentrates by flocculation and their assessment as aquaculture feeds. *Aquacultural Engineering*, 35(3), 300–313. <https://doi.org/10.1016/j.aquaeng.2006.04.001>
- Koberg, M., Cohen, M., Ben-Amotz, A., & Gedanken, A. (2011). Bio-diesel production directly from the microalgae biomass of *Nannochloropsis* by microwave and ultrasound radiation. *Bioresource Technology*, 102(5), 4265–4269. <https://doi.org/10.1016/j.biortech.2010.12.004>
- Kontas, A., Kucuksezgin, F., Altay, O., & Uluturhan, E. (2004). Monitoring of eutrophication and nutrient limitation in the Izmir Bay (Turkey) before and after Wastewater Treatment Plant. *Environment International*, 29(8), 1057–1062. [https://doi.org/10.1016/S0160-4120\(03\)00098-9](https://doi.org/10.1016/S0160-4120(03)00098-9)
- Lardon, Laurent, Helias, A., Sialve, B., Steyer, J., & Bernard, O. (2009). Life-Cycle Assessment of Biodiesel Production from Microalgae. *Environmental Science & Technology*, 43, 6475–6481.
- Larkum, A. W. D., Ross, I. L., Kruse, O., & Hankamer, B. (2012). Selection, breeding and engineering of microalgae for bioenergy and biofuel production. *Trends in Biotechnology*, 30(4), 198–204. <https://doi.org/10.1016/j.tibtech.2011.11.003>
- Lee, A. K., Lewis, D. M., & Ashman, P. J. (2012). Disruption of microalgae cells for the extraction lipids for biofuels: process and specific energy requirements. *Biomass and Bioenergy*, 46, 89–101.
- Lee, A., Lewis, D., Kalaitzidis, T., & Ashman, P. (2016). Technical issues in the large-scale hydrothermal liquefaction of microalgal biomass to biocrude. *Current Opinion in Biotechnology*, 38, 85–89. <https://doi.org/10.1016/j.copbio.2016.01.004>
- Lee, J. Y., Yoo, C., Jun, S. Y., Ahn, C. Y., & Oh, H. M. (2010). Comparison of several methods for effective lipid extraction from microalgae. *Bioresource Technology*, 101(1 SUPPL.), 2009–2011. <https://doi.org/10.1016/j.biortech.2009.03.058>
- Lee, S. J., Yoon, B. D., & Oh, H. M. (1998). Rapid method for the determination of lipid from

- the green alga *Botryococcus braunii*. *Biotechnology Techniques*, 12(7), 553–556.
[https://doi.org/Doi 10.1023/A:1008811716448](https://doi.org/Doi%2010.1023/A:1008811716448)
- Lemões, J. S., Alves, R. C. M., Farias, S. P., Moura, R. R. De, Primel, E. G., Abreu, P. C., ... Oca, D. (2016). Sustainable production of biodiesel from microalgae by direct transesterification. *Sustainable Chemistry and Pharmacy*, 3, 33–38.
<https://doi.org/10.1016/j.scp.2016.01.002>
- Letelier-Gordo, C. O., Holdt, S. L., De Francisci, D., Karakashev, D. B., & Angelidaki, I. (2014). Effective harvesting of the microalgae *Chlorella protothecoides* via bioflocculation with cationic starch. *Bioresource Technology*, 167, 214–218.
<https://doi.org/10.1016/j.biortech.2014.06.014>
- Li, Y., Lian, S., Tong, D., Song, R., Yang, W., Fan, Y., ... Hu, C. (2011). One-step production of biodiesel from *Nannochloropsis* sp. on solid base Mg-Zr catalyst. *Applied Energy*, 88(10), 3313–3317. <https://doi.org/10.1016/j.apenergy.2010.12.057>
- Ma, Y. A., Cheng, Y. M., Huang, J. W., Jen, J. F., Huang, Y. S., & Yu, C. C. (2014). Effects of ultrasonic and microwave pretreatments on lipid extraction of microalgae. *Bioprocess and Biosystems Engineering*, 37(8), 1543–1549. <https://doi.org/10.1007/s00449-014-1126-4>
- Made-in-China.com. (2018). Cationic Starch Price. Retrieved February 28, 2018, from https://www.made-in-china.com/products-search/hot-china-products/Cationic_Starch_Price.html
- Mahesar, S. A., Sherazi, S. T. H., Abro, K., Kandhro, A., Bhanger, M. I., van de Voort, F. R., & Sedman, J. (2008). Application of microwave heating for the fast extraction of fat content from the poultry feeds. *Talanta*, 75(5), 1240–1244.
<https://doi.org/10.1016/j.talanta.2008.01.042>
- Mallick, N., Bagchi, S. K., Koley, S., & Singh, A. K. (2016). Progress and challenges in microalgal biodiesel production. *Frontiers in Microbiology*, 7(JUN), 1–11.
<https://doi.org/10.3389/fmicb.2016.01019>
- Manganaro, J. L., & Lawal, A. (2016). CO₂ Life-cycle assessment of the production of algae based liquid fuel compared to crude oil to diesel. *Energy & Fuels*, acs.energyfuels.6b00207.

<https://doi.org/10.1021/acs.energyfuels.6b00207>

- Markewitz, P., Marx, J., Schreiber, A., & Zapp, P. (2013). Ecological evaluation of coal-fired Oxyfuel power plants -Cryogenic versus membrane-based air separation. *Energy Procedia*, 37(June), 2864–2876. <https://doi.org/10.1016/j.egypro.2013.06.172>
- Marulanda, V. F. (2012). Biodiesel production by supercritical methanol transesterification: Process simulation and potential environmental impact assessment. *Journal of Cleaner Production*, 33, 109–116. <https://doi.org/10.1016/j.jclepro.2012.04.022>
- Mata, T. M., Mendes, A. M., Caetano, N. S., & Martins, A. A. (2014). Sustainability and economic evaluation of microalgae grown in brewery wastewater. *Bioresource Technology*, 168, 151–158. <https://doi.org/10.1016/j.biortech.2014.04.091>
- Mehrabadi, A., Craggs, R., & Farid, M. M. (2015). Wastewater treatment high rate algal ponds (WWT HRAP) for low-cost biofuel production. *Bioresource Technology*, 184, 202–214. <https://doi.org/10.1016/j.biortech.2014.11.004>
- Mendes, R. L., Nobre, B. P., Cardoso, M. T., Pereira, A. P., & Palavra, A. F. (2003). Supercritical carbon dioxide extraction of compounds with pharmaceutical importance from microalgae. *Inorganica Chimica Acta*, 356, 328–334. [https://doi.org/10.1016/S0020-1693\(03\)00363-3](https://doi.org/10.1016/S0020-1693(03)00363-3)
- Mercer, P., & Armenta, R. E. (2011). Developments in oil extraction from microalgae. *European Journal of Lipid Science and Technology*, 113(5), 539–547. <https://doi.org/10.1002/ejlt.201000455>
- Metzger, P., & Largeau, C. (2005). *Botryococcus braunii*: A rich source for hydrocarbons and related ether lipids. *Applied Microbiology and Biotechnology*, 66(5), 486–496. <https://doi.org/10.1007/s00253-004-1779-z>
- Milledge, J. J., & Heaven, S. (2013). A review of the harvesting of micro-algae for biofuel production. *Reviews in Environmental Science and Biotechnology*, 12(2), 165–178. <https://doi.org/10.1007/s11157-012-9301-z>
- Molina Grima, E., Belarbi, E. H., Acien Fernández, F. G., Robles Medina, A., & Chisti, Y. (2003). Recovery of microalgal biomass and metabolites: Process options and economics.

Biotechnology Advances, 20(7–8), 491–515. [https://doi.org/10.1016/S0734-9750\(02\)00050-2](https://doi.org/10.1016/S0734-9750(02)00050-2)

- Mussgnug, J. H., Klassen, V., Schlüter, A., & Kruse, O. (2010). Microalgae as substrates for fermentative biogas production in a combined biorefinery concept. *Journal of Biotechnology*, 150(1), 51–56. <https://doi.org/10.1016/j.jbiotec.2010.07.030>
- Nagarajan, S., Chou, S. K., Cao, S., Wu, C., & Zhou, Z. (2013). An updated comprehensive techno-economic analysis of algae biodiesel. *Bioresource Technology*, 145, 150–156. <https://doi.org/10.1016/j.biortech.2012.11.108>
- Najafi, G., Ghobadian, B., & Yusaf, T. F. (2011). Algae as a sustainable energy source for biofuel production in Iran: A case study. *Renewable and Sustainable Energy Reviews*, 15(8), 3870–3876. <https://doi.org/10.1016/j.rser.2011.07.010>
- National Oceanic and Atmospheric Administration. (2016). Recent Monthly mean CO₂ at Mauna Loa. Retrieved from <http://www.esrl.noaa.gov/gmd/ccgg/trends/index.html>
- Natural Resources Canada. (2017). Mean daily insolation by province per month. Retrieved May 17, 2017, from <https://www.nrcan.gc.ca/18366>
- Nicholas E. Leadbeater and Lauren M. Stencel. (2006). Fast, easy preparation of biodiesel using microwave heating. *Energy Fuels*, 20(5), 2281–2283.
- Nikolaou, A., Hartmann, P., Sciandra, A., Chachuat, B., & Bernard, O. (2016). Dynamic coupling of photoacclimation and photoinhibition in a model of microalgae growth. *Journal of Theoretical Biology*, 390, 61–72. <https://doi.org/10.1016/j.jtbi.2015.11.004>
- Norsker, N. H., Barbosa, M. J., Vermuë, M. H., & Wijffels, R. H. (2011). Microalgal production - A close look at the economics. *Biotechnology Advances*, 29(1), 24–27. <https://doi.org/10.1016/j.biotechadv.2010.08.005>
- OFX. (2018). Yearly Average Rates. Retrieved February 28, 2018, from <https://www.ofx.com/en-ca/forex-news/historical-exchange-rates/yearly-average-rates/>
- Oilgae. (2010). Photobioreactors. Retrieved October 23, 2017, from <http://www.oilgae.com/ref/glos/photobioreactor.html>

- Oliveira, E. G., Rosa, G. S., Moraes, M. A., & Pinto, L. A. A. (2009). Characterization of thin layer drying of *Spirulina platensis* utilizing perpendicular air flow. *Bioresource Technology*, 100(3), 1297–1303. <https://doi.org/10.1016/j.biortech.2008.05.052>
- Ontario Energy Board. (n.d.). Historical electricity rates. Retrieved March 9, 2018, from <https://www.oeb.ca/rates-and-your-bill/electricity-rates/historical-electricity-rates>
- Onwudili, J. A., Lea-Langton, A. R., Ross, A. B., & Williams, P. T. (2013). Catalytic hydrothermal gasification of algae for hydrogen production: Composition of reaction products and potential for nutrient recycling. *Bioresource Technology*, 127, 72–80. <https://doi.org/10.1016/j.biortech.2012.10.020>
- ÖQUIST, G. (1983). Effects of low temperature on photosynthesis. *Plant, Cell & Environment*, 6(4), 281–300. <https://doi.org/10.1111/1365-3040.ep11612087>
- Orfield, N. D., Keoleian, G. A., & Love, N. G. (2014). A GIS based national assessment of algal bio-oil production potential through flue gas and wastewater co-utilization. *Biomass Bioenerg*, 63, 0–9. <https://doi.org/10.1016/j.biombioe.2014.01.047>
- Oswald, W. (1988). Micro-algae and waste-water treatment. *Micoalgal Biotechnology*, 305–328.
- Oswald, W., Gotaas, H., Golueke, C., & Kellen, W. (1957). Algae in waste treatment. *Sewage Ind. Wastes*, 29, 437–455.
- Pachauri, R., Meyer, L., & Team, T. C. W. (2014). *Climate Change 2014 Synthesis Report. IPCC* (Vol. 1). IPCC. <https://doi.org/10.1017/CBO9781107415324.004>
- Patil, P. D., Gude, V. G., Camacho, L. M., & Deng, S. (2010). Microwave-assisted catalytic transesterification of camelina sativa oil. *Energy and Fuels*, 24(2), 1298–1304. <https://doi.org/10.1021/ef9010065>
- Patil, P. D., Gude, V. G., Mannarswamy, A., Deng, S., Cooke, P., Munson-McGee, S., ... Nirmalakhandan, N. (2011). Optimization of direct conversion of wet algae to biodiesel under supercritical methanol conditions. *Bioresource Technology*, 102(1), 118–122. <https://doi.org/10.1016/j.biortech.2010.06.031>
- Peng, C., Li, S., Zheng, J., Huang, S., & Li, D. (2017). Harvesting Microalgae with Different

- Sources of Starch-Based Cationic Flocculants. *Applied Biochemistry and Biotechnology*, 181(1), 112–124. <https://doi.org/10.1007/s12010-016-2202-9>
- Peng, G. (2015). Methane Production from Microalgae via Continuous Catalytic Supercritical Water Gasification: Development of Catalysts and Sulfur Removal Techniques, 6740, 239.
- Pittman, J. K., Dean, A. P., & Osundeko, O. (2011). The potential of sustainable algal biofuel production using wastewater resources. *Bioresource Technology*, 102(1), 17–25. <https://doi.org/10.1016/j.biortech.2010.06.035>
- Power Plant CCS. (2010). Costs of PBRs. Retrieved October 26, 2017, from http://www.powerplantccs.com/ccs/cap/fut/alg/costs_of_pbr.html
- Prabakaran, P., & Ravindran, A. D. (2011). A comparative study on effective cell disruption methods for lipid extraction from microalgae. *Letters in Applied Microbiology*, 53(2), 150–154. <https://doi.org/10.1111/j.1472-765X.2011.03082.x>
- Prakash, J., Pushparaj, B., Carlozzi, P., Torzillo, G., Montaini, E., & Materassi, R. (1997). Microalgal biomass drying by a simple solar device. *International Journal of Solar Energy*, 18(4), 303–311. <https://doi.org/10.1080/01425919708914325>
- Quinn, J. C., & Davis, R. (2015). The potentials and challenges of algae based biofuels: A review of the techno-economic, life cycle, and resource assessment modeling. *Bioresource Technology*, 184, 444–452. <https://doi.org/10.1016/j.biortech.2014.10.075>
- Radakovits, R., Jinkerson, R. E., Darzins, A., & Posewitz, M. C. (2010). Genetic engineering of algae for enhanced biofuel production. *Eukaryotic Cell*, 9(4), 486–501. <https://doi.org/10.1128/EC.00364-09>
- Ramadhass, A. S., Jayaraj, S., & Muraleedharan, C. (2005). Biodiesel production from high FFA rubber seed oil. *Fuel*, 84(4), 335–340. <https://doi.org/10.1016/j.fuel.2004.09.016>
- Ramos Tercero, E. A., Domenicali, G., & Bertucco, A. (2014). Autotrophic production of biodiesel from microalgae: An updated process and economic analysis. *Energy*, 76, 807–815. <https://doi.org/10.1016/J.ENERGY.2014.08.077>
- Ranjan, A., Patil, C., & Moholkar, V. S. (2010). Mechanistic assessment of microalgal lipid

- extraction. *Industrial and Engineering Chemistry Research*, 49(6), 2979–2985.
<https://doi.org/10.1021/ie9016557>
- Refaat, A. A. (2010). Different techniques for the production of biodiesel from waste vegetable oil. *Int. J. Environ. Sci. Tech.*, 7(1), 183–213. <https://doi.org/10.1007/BF03326130>
- Ren, H., Tuo, J., Addy, M. M., Zhang, R., Lu, Q., Anderson, E., ... Ruan, R. (2017). Cultivation of *Chlorella vulgaris* in a pilot-scale photobioreactor using real centrate wastewater with waste glycerol for improving microalgae biomass production and wastewater nutrients removal. *Bioresource Technology*, 245(July), 1130–1138.
<https://doi.org/10.1016/j.biortech.2017.09.040>
- Renaud, S. M., Thinh, L.-V., Lambrinidis, G., & Parry, D. L. (2002). Effect of temperature on growth, chemical composition and fatty acid composition of tropical Australian microalgae grown in batch cultures. *Aquaculture*, 211(1–4), 195–214. [https://doi.org/10.1016/S0044-8486\(01\)00875-4](https://doi.org/10.1016/S0044-8486(01)00875-4)
- Resa, J. M., Goenaga, J. M., Iglesias, M., Gonzalez-Olmos, R., & Pozuelo, D. (2006). Liquid-liquid equilibrium diagrams of ethanol plus water plus (ethyl acetate or 1-pentanol) at several temperatures. *Journal of Chemical & Engineering Data*, 51(4), 1300–1305.
<https://doi.org/10.1021/je060054+>
- Rezvani, S., Moheimani, N. R., & Bahri, P. A. (2016). Techno-economic assessment of CO₂ bio-fixation using microalgae in connection with three different state-of-the-art power plants. *Computers and Chemical Engineering*, 84, 290–301.
<https://doi.org/10.1016/j.compchemeng.2015.09.001>
- Richardson, J. W., Johnson, M. D., & Outlaw, J. L. (2012). Economic comparison of open pond raceways to photo bio-reactors for profitable production of algae for transportation fuels in the Southwest. *Algal Research*, 1(1), 93–100. <https://doi.org/10.1016/j.algal.2012.04.001>
- Richardson, J. W., Johnson, M. D., Zhang, X., Zemke, P., Chen, W., & Hu, Q. (2014). A financial assessment of two alternative cultivation systems and their contributions to algae biofuel economic viability. *Algal Research*, 4(1), 96–104.
<https://doi.org/10.1016/j.algal.2013.12.003>

- Rickman, M., Pellegrino, J., Hock, J., Shaw, S., & Freeman, B. (2013). Life-cycle and techno-economic analysis of utility-connected algae systems. *Algal Research*, 2(1), 59–65.
<https://doi.org/10.1016/j.algal.2012.11.003>
- Rizwan, M., Lee, J. H., & Gani, R. (2015). Optimal design of microalgae-based biorefinery: Economics, opportunities and challenges. *Applied Energy*, 150, 69–79.
<https://doi.org/10.1016/j.apenergy.2015.04.018>
- Ryckebosch, E., Muylaert, K., Eeckhout, M., Ruysen, T., & Foubert, I. (2011). Influence of drying and storage on lipid and carotenoid stability of the Microalga *Phaeodactylum tricornutum*. *Journal of Agricultural and Food Chemistry*, 59(20), 11063–11069.
<https://doi.org/10.1021/jf2025456>
- Sager, J., & McFarlane, J. C. (1997). *Growth Chamber Handbook*. (R.W. Langhans & T.W. Tibbitts, Eds.) (1st ed.). Iowa: North Central Regional Research. Retrieved from
<https://www.controlledenvironments.org/growth-chamber-handbook/>
- Sayre, R. (2010). Microalgae: The Potential for Carbon Capture. *BioScience*, 60(9), 722–727.
<https://doi.org/10.1525/bio.2010.60.9.9>
- Sharma, Y. C., & Singh, V. (2017). Microalgal biodiesel: A possible solution for India's energy security. *Renewable and Sustainable Energy Reviews*, 67, 72–88.
<https://doi.org/10.1016/j.rser.2016.08.031>
- Shelef, G., & Sukenik, A. (1984). Microalgae Harvesting and Processing : A Literature Review. *Technion Research and Development Foundation Ltd*, (0), 65.
<https://doi.org/http://dx.doi.org/10.2172/6204677>
- Shift Project Data Portal. (2015). Carbon Intensity of Fuel. Retrieved February 24, 2017, from
<http://www.tsp-data-portal.org/Carbon-Intensity-of-GDP#tspQvChart>
- Shirzad-Semzar, M., Kulicke, W., & Lotz, M. (1993). Cationic Starches as Substitutes for Synthetic Cationic Flocculants in Waste Water Treatment. *Agfdt.De*, (February), 1–9.
Retrieved from <http://www.agfdt.de/loads/st08/kulicke.pdf>
- Singh, A., Nigam, P. S., & Murphy, J. D. (2011). Renewable fuels from algae: An answer to debatable land based fuels. *Bioresource Technology*, 102(1), 10–16.

<https://doi.org/10.1016/j.biortech.2010.06.032>

Singh, R. P., Tripathy, T., Karmakar, G. P., Rath, S. K., Karmakar, N. C., Pandey, S. R., ... Lan, N. T. (2000). Novel biodegradable flocculants based on polysaccharides. *Current Science*, 78(7), 798–803.

Slade, R., & Bauen, A. (2013). Micro-algae cultivation for biofuels: Cost, energy balance, environmental impacts and future prospects. *Biomass and Bioenergy*, 53(1), 29–38.
<https://doi.org/10.1016/j.biombioe.2012.12.019>

Sukahara, K. T., & Awayama, S. S. (2005). Liquid Fuel Production Using Microalgae. *Journal of the Japan Petroleum Institute*, 48(5), 251–259. <https://doi.org/10.1627/jpi.48.251>

Sun, A., Davis, R., Starbuck, M., Ben-Amotz, A., Pate, R., & Pienkos, P. T. (2011). Comparative cost analysis of algal oil production for biofuels. *Energy*, 36(8), 5169–5179.
<https://doi.org/10.1016/j.energy.2011.06.020>

Suslick, K. S., & Flannigan, D. J. (2008). Inside a Collapsing Bubble: Sonoluminescence and the Conditions During Cavitation. *Annual Review of Physical Chemistry*, 59(1), 659–683.
<https://doi.org/10.1146/annurev.physchem.59.032607.093739>

Sustainable america. (2016). wastewater treatment plant capabilities. Retrieved October 2, 2016, from http://www.sustainableamerica.org/wordpress/wp-content/uploads/2013/03/us_wastewater_treatment_plant_capabilities.pdf

Swanson, R. M., Platon, A., Satrio, J. A., & Brown, R. C. (2010). Techno-economic analysis of biomass-to-liquids production based on gasification. *Fuel*, 89(SUPPL. 1).
<https://doi.org/10.1016/j.fuel.2010.07.027>

Talbot, P., Thébault, J. M., Dauta, A., & de la Noüe, J. (1991). A comparative study and mathematical modeling of temperature, light and growth of three microalgae potentially useful for wastewater treatment. *Water Research*, 25(4), 465–472.
[https://doi.org/10.1016/0043-1354\(91\)90083-3](https://doi.org/10.1016/0043-1354(91)90083-3)

Technology, E. (2008). Biofuel generations. Retrieved from <http://energyfromwasteandwood.weebly.com/generations-of-biofuels.html>

- Today in Energy. (2013). Few transportation fuels surpass the energy densities of gasoline and diesel - Today in Energy - U.S. Energy Information Administration (EIA). Retrieved February 25, 2016, from <http://www.eia.gov/todayinenergy/detail.php?id=9991>
- Tolieng, V., Prasirtsak, B., Sitdhipol, J., Thongchul, N., & Tanasupawat, S. (2017). EVALUATION OF MICROALGAE FOR SECONDARY AND TERTIARY WASTEWATER TREATMENT. *Malaysian Journal of Microbiology*, 13(2), 100–108. <https://doi.org/10.1017/CBO9781107415324.004>
- Topare, N. S., Raut, S. J., Renge, V. C., Khedkar, S. V., Chavan, Y. P., & Bhagat, S. L. (2011). Extraction of oil from algae by solvent extraction and oil expeller method. *International Journal of Chemical Sciences*, 9(4), 1746–1750.
- Udom, I., Zaribaf, B. H., Halfhide, T., Gillie, B., Dalrymple, O., Zhang, Q., & Ergas, S. J. (2013). Harvesting microalgae grown on wastewater. *Bioresource Technology*, 139, 101–106. <https://doi.org/10.1016/j.biortech.2013.04.002>
- Uduman, N., Qi, Y., Danquah, M. K., Forde, G. M., & Hoadley, A. (2010). Dewatering of microalgal cultures: A major bottleneck to algae-based fuels. *Journal of Renewable and Sustainable Energy*, 2(1). <https://doi.org/10.1063/1.3294480>
- Unit Juggler. (n.d.). Energy Converter. Retrieved April 12, 2018, from <https://www.unitjuggler.com/convert-energy-from-GJ-to-kWh.html>
- Van Den Hende, S., Vervaeren, H., & Boon, N. (2012). Flue gas compounds and microalgae: (Bio-)chemical interactions leading to biotechnological opportunities. *Biotechnology Advances*, 30(6), 1405–1424. <https://doi.org/10.1016/j.biotechadv.2012.02.015>
- Vandamme, D., Foubert, I., Meesschaert, B., & Muylaert, K. (2010). Flocculation of microalgae using cationic starch. *Journal of Applied Phycology*, 22(4), 525–530. <https://doi.org/10.1007/s10811-009-9488-8>
- Ventura, J. S., Yang, B., Lee, Y., Lee, K., & Jahng, D. (2013). Bioresource Technology Life cycle analyses of CO₂, energy, and cost for four different routes of microalgal bioenergy conversion. *Bioresource Technology*, 137, 302–310. <https://doi.org/10.1016/j.biortech.2013.02.104>

- Verbyla, M., von Sperling, M., & Maiga, Y. (2017). *Management of risk from excreta and wastewater - Part 4*. Retrieved from <http://www.waterpathogens.org/book/waste-stabilization-ponds>
- Wahlen, B. D., Willis, R. M., & Seefeldt, L. C. (2011). Biodiesel production by simultaneous extraction and conversion of total lipids from microalgae, cyanobacteria, and wild mixed-cultures. *Bioresource Technology*, 102(3), 2724–2730.
<https://doi.org/10.1016/j.biortech.2010.11.026>
- Walmsley, D. C., Bailis, R., & Klein, A.-M. (2016). A Global Synthesis of Jatropha Cultivation: Insights into Land Use Change and Management Practices. *Environmental Science & Technology*, 50(17), 8993–9002. <https://doi.org/10.1021/acs.est.6b01274>
- Ward, A. J., Lewis, D. M., & Green, F. B. (2014). Anaerobic digestion of algae biomass: A review. *Algal Research*, 5(1), 204–214. <https://doi.org/10.1016/j.algal.2014.02.001>
- Weschler, M. K., Barr, W. J., Harper, W. F., & Landis, A. E. (2014). Process energy comparison for the production and harvesting of algal biomass as a biofuel feedstock. *Bioresource Technology*, 153, 108–115. <https://doi.org/10.1016/j.biortech.2013.11.008>
- Wicaksana, F., Fane, A. G., Pongpaiboj, P., & Field, R. (2012). Microfiltration of algae (*Chlorella sorokiniana*): Critical flux, fouling and transmission. *Journal of Membrane Science*, 387–388(1), 83–92. <https://doi.org/10.1016/j.memsci.2011.10.013>
- Wiley, P., Campbell, J. E., & McKuin, B. (2011). Production of biodiesel and biogas from algae: a review of process train options. *Water Environment Research*, 83(5), 326–338. Retrieved from https://www.academia.edu/1913585/Production_of_biodiesel_and_biogas_from_algae_a_review_of_process_train_options?auto=download
- World Nuclear Association. (2018). Heat values of various fuels. Retrieved March 9, 2018, from <http://www.world-nuclear.org/information-library/facts-and-figures/heat-values-of-various-fuels.aspx>
- Xin, C., Addy, M. M., Zhao, J., Cheng, Y., Cheng, S., Mu, D., ... Ruan, R. (2016). Comprehensive techno-economic analysis of wastewater-based algal biofuel production: A

case study. *Bioresource Technology*, 211, 584–593.

<https://doi.org/10.1016/j.biortech.2016.03.102>

Zaimes, G. G., & Khanna, V. (2013). Supporting Information for : Microalgal biomass production pathways : Evaluation of life cycle environmental impacts. *Biotechnology for Biofuels*, 1–45. <https://doi.org/10.1186/1754-6834-6-88>

Zamalloa, C., Vulsteke, E., Albrecht, J., & Verstraete, W. (2011). The techno-economic potential of renewable energy through the anaerobic digestion of microalgae. *Bioresource Technology*, 102(2), 1149–1158. <https://doi.org/10.1016/j.biortech.2010.09.017>

Zhang, Y., Li, Y., Zhang, X., & Tan, T. (2015). Biodiesel production by direct transesterification of microalgal biomass with co-solvent. *Bioresource Technology*, 196, 712–715. <https://doi.org/10.1016/j.biortech.2015.07.052>

Zhou, W., Chen, P., Min, M., Ma, X., Wang, J., Griffith, R., ... Ruan, R. (2014). Environment-enhancing algal biofuel production using wastewaters. *Renewable and Sustainable Energy Reviews*, 36, 256–269. <https://doi.org/10.1016/j.rser.2014.04.073>