Sustainable production pathways for added-value products from microalgae

A superstructure optimization approach

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SUSTAINABLE PRODUCTION PATHWAYS FOR ADDED-VALUE PRODUCTS FROM MICROALGAE

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A SUPERSTRUCTURE OPTIMIZATION APPROACH

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Summary

Among various types of biomasses, microalgae are considered valuable due to their high growth rate and photosynthesis efficiency. Microalgae are marine microorganisms that grow in wastewater using carbon dioxide and sun or artificial light. The capability to capture carbon dioxide and treat wastewater makes this microorganism an attractive alternative to solve environmental issues. In addition, microalgae have a high potential to serve as a source for various added-value products.

Several processes are at hand to extract these added-value products from the microalgae. The amount and final price of these bioproducts are determined by the character and efficiency level of each technology. Finding suitable technologies for each type of microalga biorefinery is a challenge that needs to be overcome. Additionally, there are variations in the composition and amount of the feedstock and the amount and purity of the bioproducts produced. The profitability of a microalgae biorefinery is also affected by these parameters. Thus, considering microalgal biorefinery under different types of uncertainties is another challenge. This challenge should be tackled to extend the industrial application of microalgae.

In chapter 1 of this thesis, the general developments and challenges that are encountered in extracting added-value products from microalgae are discussed. Also, the main objectives and scope of this thesis are introduced.

In chapter 2, an introduction to the various technologies that can be used at each stage of the biorefinery process to produce added-value products and biofuels from microalgae is presented. Energy and efficiency are discussed for each of the available technologies. A discussion of the role of process systems engineering in enhancing the microalgae economy follows. Process systems engineering can play an important role in the biobased economy, especially by applying sustainability and economic concepts in the decision-making process for selecting the best feedstock, processing pathways, and desired products. Tools such as uncertainty analysis, techno-economic analysis, and life cycle assessment (LCA) can be applied to design sustainable microalgae biorefinery.

In chapter 3, the techno-economic analysis tool is used to compare three types of microalgae biorefinery. A superstructure of a microalgae biorefinery is developed to find cost-effective pathways to produce added-value products. This superstructure is then transformed into a mixed-integer nonlinear programming (MINLP) model to optimize with BARON/AOA in AIMMS software. A new block integration approach reduces the number of variables, parameters, and constraints to drastically reduce CPU time. For all three biorefineries (cultivating different microalgae), the cost-effective production pathways are the same: an open pond, sedimentation and flotation, flocculation without any dryer, sonication, organic solvent pigment extraction, n-butanol solvent lipid extraction, lipid production, and anaerobic digestion. Due to the high amount of pigment produced by Haematococcus Pluvialis, it is the

most profitable biorefinery. The profits of this biorefinery are 22 and 47 times higher than Chlorella Vulgaris and Nannochloropsis spp. biorefineries, respectively.

In chapter 4, a sensitivity analysis is executed to evaluate how uncertainty in feedstock characteristics and availability affects the economies and the quantities of bioproducts. There are two types of uncertainty associated with feedstock: the composition of the influent wastewater and the duration of the sunshine. Dutch data are used to determine the statistical parameters (average composition and associated standard deviations) for the Monte Carlo simulation. The probability of reaching an average profit and revenue of bioproducts are then calculated. During uncertainty of composition of wastewater studies, Haematococcus Pluvialis biorefinery profit margin per kg of microalgae varies between 58.59 ($/kg) and 62.94 ($/kg), with an average of 62.86 and 0.912 standard deviation. A Haematococcus Pluvialis biorefinery has a higher probability of achieving a profit margin (62 $/kg of microalgae). Second, the amount of sunshine duration varies across seasons, affecting profit margins and bioproduct amounts. Microalgae biorefineries have a 50% reduction in annual profit margin when actual sunshine duration is used.

Chapter 5 introduces a life cycle analysis to compare three microalgae biorefineries from an environmental point of view. A cradle-to-gate LCA of these biorefineries, using Dutch influent wastewater as the functional unit, shows Nannochloropsis spp. biorefinery has the highest environmental impacts due to its high productivity and biomass production. In addition, it is compared how different steps of microalgae biorefinery will impact the environment when producing one kilogram of pigment. Microalgae is converted to lipid, pigment, and other components in the cell disruption step and most of the cell components are burned in the remnant treatment step to produce biogas and fertilizers. The remnant treatment is the most effective stage of biorefinery in terms of global warming, human toxicity, ecotoxicity, and acidification impacts followed by the cell disruption step. A comparison of the economic and environmental points of view of two types of biomasses (microalga and carrot) shows that some efforts are needed to decrease the environmental impact and cost of microalgae biorefineries when producing 1 kg β-carotene.

In chapters 6 and 7 conclusions and outlook of this thesis are presented, respectively.

Samenvatting

Van diverse biomassabronnen worden microalgen als waardevol beschouwd vanwege hun hoge groeisnelheid en fotosynthese-efficiëntie. Microalgen zijn mariene micro-organismen die in afvalwater kunnen groeien met behulp van kooldioxide en zon-of kunstlicht. Het vermogen om kooldioxide af te vangen en afvalwater te behandelen, maakt dit micro-organisme een aantrekkelijke optie om milieuproblemen op te lossen. Bovendien hebben microalgen een groot potentieel om als bron te dienen voor verschillende producten met toegevoegde waarde.

Er zijn verschillende processen beschikbaar om deze producten met toegevoegde waarde van microalgen te scheiden. De opbrengst en de uiteindelijke verkoopprijs van deze bioproducten worden bepaald door het karakter en de efficiëntie van elke technologie. Het vinden van geschikte technologieën voor elk type bioraffinage van de toegevoegde waarde producten uit de microalgen is een uitdaging die moet worden overwonnen. De samenstelling en hoeveelheid van de grondstof (afvalwater) en de hoeveelheid en zuiverheid van de geproduceerde bioproducten kunnen erg variëren. De winstgevendheid van een microalgenbioraffinaderij wordt dan ook beïnvloed door de variatie in deze parameters. Het analyseren van microalgenbioraffinage onder verschillende soorten onzekerheden is dus een andere belangrijke uitdaging. Deze uitdaging moet worden aangepakt om de industriële toepassing van microalgen uit te breiden.

In hoofdstuk 1 worden de huidige ontwikkelingen en voornaamste uitdagingen die er bestaan bij het scheiden van toegevoegde waarde producten uit microalgen besproken. Daarnaast worden de onderzoeksdoelstellingen en de scope van dit proefschrift geintroduceerd.

In hoofdstuk 2 wordt een inleiding gegeven over de verschillende technologieën die in elke fase van het bioraffinageproces kunnen worden gebruikt om producten met toegevoegde waarde en biobrandstoffen te produceren uit microalgen. Voor elk van de beschikbare technologieën wordt de energievraag en operationele efficiëntie besproken. Daarop volgt een discussie over de rol van process systems engineering (PSE) bij het verbeteren van de microalgeneconomie. PSE kan een belangrijke rol spelen in de biobased economy, vooral door duurzaamheids- en economische concepten toe te passen in het besluitvormingsproces voor het selecteren van de beste grondstof, verwerkingsroutes en gewenste producten. Instrumenten zoals onzekerheidsanalyse, techno-economische analyse en levenscyclusanalyse (LCA) kunnen worden toegepast om duurzame bioraffinage van microalgen te ontwerpen.

In hoofdstuk 3 wordt eentechno-economische analyse uitgevoerd om drie soorten bioraffinage van microalgen te vergelijken. Een superstructuur van een bioraffinaderij voor microalgen is ontwikkeld om kosteneffectieve routes te vinden om producten met toegevoegde waarde te produceren. Deze superstructuur wordt vervolgens getransformeerd

in een mixed-integer non-linear programming (MINLP)-model om te optimaliseren met BARON/AOA in AIMMS-software. Een nieuwe aanpak voor blokintegratie vermindert het aantal variabelen, parameters en beperkingen om de CPU-tijd drastisch te verminderen. Voor alle drie de bioraffinaderijen (het kweken van verschillende microalgen) zijn de kosteneffectieve productieroutes hetzelfde: een open vijver, sedimentatie en flotatie, flocculatie zonder droger, sonicatie, extractie van organisch oplosmiddelpigment, extractie van n-butanol-oplosmiddellipiden, lipideproductie, en anaërobe vergisting. Vanwege de hoge hoeveelheid pigment die door Haematococcus Pluvialis wordt geproduceerd, is het de meest winstgevende bioraffinaderij. De winst van deze bioraffinaderij is 22 tot 47 keer hoger dan van respectievelijk de Chlorella Vulgaris en Nannochloropsis spp. bioraffinaderijen.

In hoofdstuk 4 wordt een gevoeligheidsanalyse uitgevoerd om te evalueren hoe onzekerheid in grondstofkenmerken en beschikbaarheid van de grodstof de economische aspecten en de hoeveelheden bioproducten beïnvloedt. Er zijn twee soorten onzekerheid die samenhangen met grondstof: de samenstelling van het influent afvalwater en de duur van het zonlicht. Voor het bepalen van de statistische parameters (gemiddelde samenstelling en bijbehorende standaarddeviaties) voor de Monte Carlo-simulatie worden Nederlandse gegevens gebruikt. Vervolgens wordt de kans berekend op het behalen van een gemiddelde winst en omzet van bioproducten. Tijdens de onzekerheidsanalyse over de samenstelling van afvalwater varieert de winstmarge van de bioraffinaderij van Haematococcus Pluvialis per kg microalgen tussen 58,59 ($) en 62,94 ($), met een gemiddelde van $62,86 en een standaarddeviatie van 0,912. Een bioraffinaderij van Haematococcus Pluvialis heeft een grotere kans om een winstmarge te behalen (62 $/kg microalg). Ten tweede varieert de hoeveelheid zonneschijn per seizoen, wat van invloed is op de winstmarges en hoeveelheden bioproducten. Bioraffinaderijen voor microalgen hebben een afname van 50% van de jaarlijkse winstmarge wanneer de werkelijke zonneschijnduur wordt gebruikt.

Hoofdstuk 5 introduceert een levenscyclusanalyse om drie microalgenbioraffinaderijen te vergelijken vanuit milieuoogpunt. Een cradle-to-gate LCA van deze bioraffinaderijen, met Nederlands influent afvalwater als functionele eenheid, toont aan dat bioraffinage van Nannochloropsis spp. de grootste milieu-impact heeft vanwege de hoge productiviteit en biomassaproductie. Daarnaast wordt vergeleken hoe verschillende stappen van bioraffinage van microalgen het milieu zullen beïnvloeden bij het produceren van één kilogram pigment. Microalgen worden omgezet in lipiden, pigmenten en andere componenten in de celvernietigingsstap en de meeste celcomponenten worden verbrand in de restbehandelingsstap om biogas en meststoffen te produceren. De restbehandeling is de meest invloedrijke fase van bioraffinage in termen van opwarming van de aarde, menselijke toxiciteit, ecotoxiciteit en verzuringseffecten, gevolgd door de stap van celverstoring. Vergelijking van economisch en milieuoogpunt van twee soorten biomassa (microalgen en

wortel) toont aan dat er enkele inspanningen nodig zijn om de milieu-impact en kosten van bioraffinaderijen voor microalgen te verminderen bij de productie van 1 kg β -caroteen.

In hoofdstuk 6 en 7 worden respectievelijk de conclusies en de vooruitzichten van dit proefschrift gepresenteerd.

Table of Contents

Chapter One

Introduction

Abstract

The purpose of this chapter is to present the main topics discussed in the thesis entitled: "Sustainable production pathways for added value products from microalgae -A superstructure optimization approach". It begins by explaining the need for renewable energy, the benefits of using biomass, the different categories of biomass, and the potential usage of microalgae. Then, the overall goal of this research is described with a description of the four tasks to improve the economy of three types of microalgae biorefineries which include: understanding the role of process engineering in the bioeconomy, the development and use of superstructures to find promising production pathways, uncertainty in feedstock characteristics on biorefinery economics, and examining the environmental impacts of the promising pathways. Finally, the outline of the thesis explains how each chapter contributes to the overall goal.

1.1 Background

During the transition from the 20th century to the 21st century, pollution has been increasing as a result of the increasing use of fossil fuels for industrial production. Due to this factor and high energy demand, society is looking for alternative energy sources. Accordingly, major institutions in several countries have been working to develop a unified regulatory framework for promoting renewable energies which would diversify energy sources by replacing conventional ones.

Among many alternative energy sources, biomass has played a vital role in policy debates within the EU's policy context, confirming that providing energy efficiency measures and environmental protection can result in comprehensive development plans and guidelines [1]. There is no doubt that biomass could play an important role in renewable sources, not only with enormous potential in the production of biofuels for transportation, electricity, and heat [2], but also with the ability to produce various types of biocomponents.

Apart from these, the problem of climate change has brought nations to higher levels of commitment and new groups have been formed by the scientific community, such as the International Panel on Climate Change (IPCC) in 1988, to limit the emissions of greenhouse gases (GHGs) into the atmosphere primarily caused by fossil fuel utilization [3,4]. The debate has been paralleled with the importance of renewable energies, with an increasing focus on biomass, as a contributor to energy security and sustainable development. Due to recent scientific advancements in biomass exploitation, various promising techniques have been developed to produce multiple products, including biofuels, biocomponents, electricity, heat, etc. by optimizing these abundant and inexpensive natural resources.

Furthermore, it is estimated that 8% of the EU's workforce is employed in the bioeconomy. By 2030, there could be more than 1 million green jobs that are created as a result of bio-based industries, particularly in rural and coastal areas. Biobased innovation will modernize and renew industries and restoring healthy ecosystems and enhancing biodiversity. For instance, 12 million tonnes of plastic waste are dumped into our oceans every year, but if the bioeconomy is used to reduce this amount by 90% by 2025, this waste will be disposed in a far more sustainable way [5].

In general, biomass is a renewable energy resource since the carbon dioxide released during its combustion and utilization does not contribute to increased atmospheric carbon dioxide. The plants use the carbon dioxide released into the environment during the degradation of other plants for growth and metabolism. Using biomass will only increase atmospheric CO_2, which plants will re-use for biomass production (Figure 1-1) [4,6].

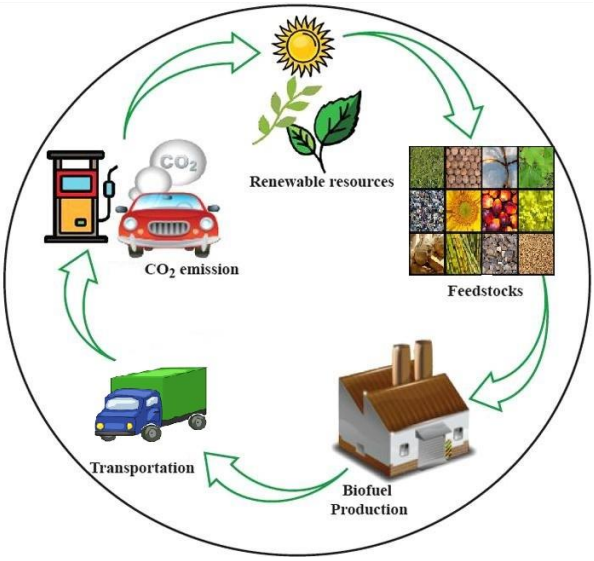


Figure 1-1: Biomass carbon cycle.

Biomass refers to any organic substance produced directly or indirectly by photosynthesis [7]. Various definitions of biomass exist because of their heterogeneity, their use, and their origin. Overall, biomass refers to all-natural components that are derived from plants, such as shrubs, trees, algae, and crops, as well as other materials based on organic materials except for plastic derived from petrochemical and fossil sources [8]. Figure 1-2 shows the most important biomass sources, including agricultural and forestry residues, industrial and animal waste, sewage, algae, and municipal solid waste (if they cannot be recycled) [4].

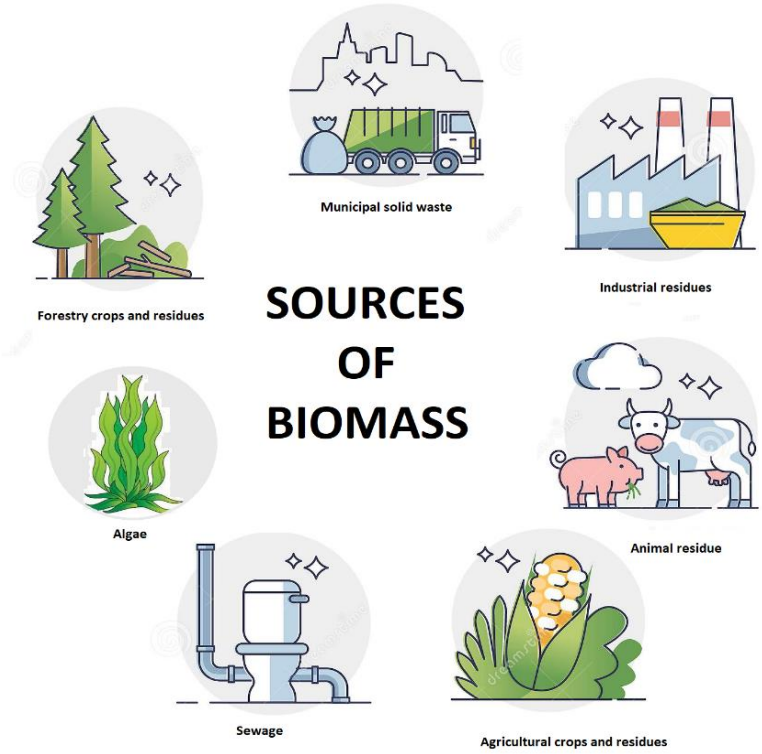


Figure 1-2: Sources of biomass

1.2 Biomass categories

Depending on the types of biomasses in nature, biomass is categorized into four groups: 1-wood and woody biomass, 2-Herbaceous biomass, 3-animal and human waste biomass, and 4-aquatic biomass.

Wood and woody biomass: Trees and roots residues, bark, and leaves of woody shrubs in both the upper and lower ground are usually included in this category and can either be burned directly (or gasified) or converted through a variety of processes to produce energy[9]. There are many components in woody biomass, but lignin and carbohydrates are the dominant components. In terms of renewable energy sources, woody biomass is currently the most important [1].

Herbaceous biomass: It consists of plants with non-woody stems that die at the end of their growing phase seasons. In addition, it includes grains and seeds crops from food processing industries, as well as their by-products, such as cereal straw [3].

Animal and human waste biomass: Human dung, animal manure, and bones are some of the most common biomasses in this category. In the past, these wastes were reclaimed for use as fertilizer or used on agricultural land, but stricter regulations on emission levels, health concerns, and an unpleasant odor have led to proper waste management. Among the available methods for converting these wastes into products, anaerobic digestion offers the best efficiency for producing biogas.

Aquatic biomass: Macroalgae, microalgae, and emerging plants are included in aquatic biomass [10]. Newly emerging plants grow partially immersed in marshes and swamps. A macroalga is a multicellular organism with a rapid growth rate. They can reach lengths of 60 m in a short time. They are used mainly in the food industry and hydrocolloid extraction [1].

A microalga is a microscopic organism that consists of diatoms and green/golden algae. Diatoms are brown and unicellular algae that are usually only a few millimeters in size; they are one of the main components of aquatic flora and represent one of the greatest sources of biomass on earth. Green/golden algae are especially abundant in freshwater resources. Algae biomass comprises pigments, lipids, proteins, and carbohydrates that can be converted into various products [11]. There is a growing industrial interest in using microalgae for an extensive range of applications, including biofuels and bioenergy, biofertilizers, vitamins, and chemical compounds for food production, nutraceutical dietary supplements, cosmetics, and pharmaceutical products, etc.[12].

Microalgae are recognized as one of the most valuable biomass types due to source for various added-value products and their high growth rate and high efficiency of photosynthesis [13]. Microalgae are oceanic microorganisms that grow in wastewater using carbon dioxide and sun or artificial light. The capability to capture carbon dioxide and treat wastewater makes this microorganism one alternative to solve environmental issues. This type of biomass can also grow in fresh water enriched with nutrients[14].

Despite the vast potential to use microalgae as a feedstock for various industries, a technical challenge must be addressed to commercially extend the use of biochemicals and biofuels from algal biomass. The optimization of a superstructure is the approach followed in this thesis to enhance the application of microalgae on a large scale by finding a cost-effective pathway. Rizwan et al. (2015) formulated a superstructure as a mixed integer non-linear program (MINLP), optimizing the net present value (NPV) of an algae biorefinery. Although biodiesel, bio-oil, and biogas are produced in this biorefinery, the capital costs are not considered [127]. Galanopoulos et al. (2019) proposed a superstructure for an integrated algae biorefinery to minimize the price of biodiesel. The total biodiesel costs can be decreased with 20 % by producing bioethanol, glycerol, and levulinic acid [131]. Their study showed that the price of biodiesel could be decreased by producing added-value products. Still, the profits of this algae biorefinery are not high enough to scale it up to a commercial level. Furthermore, they considered only a Chlorella Vulgaris biorefinery. Including different types of microalgae with different compositions and investigating various bioproducts will increase the prospect of commercializing the algae biorefinery. For this reason, a superstructure that includes three types of microalgae is developed to optimize the production pathway of added value products such as pigments, biodiesel, biogas, glycerol, omega-3, fertilizers.

1.3 Challenge of microalgae biorefinery

Different species and strains of microalgae show a great variety of growth rates, productivity, and composition. The type and amounts of bioproducts that can be extracted from them are varied. Microalgae have potential as biofuel resources; however, large-scale production of biofuels from microalgae is still in the early stages due to capital and energy-intensive production processes. One way to overcome this problem is to produce other bioproducts simultaneously with biofuel production. Therefore, an integration of extracting different bioproducts from microalgae is one option to increase the usage of this biomass. Discovering and identifying all the possible bioproducts and deciding on an integrated group of products for each microalga biorefinery is a challenge.

Each of these bioproducts is produced by passing some processes. In each process, different technologies are utilized. Because each technology has a different level of efficiency, it determines the amount and final price of bioproducts. Finding appropriate technologies for each type of microalga biorefinery is another challenge.

Furthermore, there is uncertainty on the composition and amount of feedstock, amount and purity of bioproducts, etc. These parameters also affect the profits of microalgae biorefinery. Considering microalgae biorefinery under a different type of uncertainties is another challenge.

1.4 Thesis objectives

The main objective of this work is to provide a modeling tool for optimizing the superstructure is that assist in identifying promising pathways (from an economic as well as from an environmental viewpoint). The superstructure can be used for various refineries, however, in this research three types of microalgae biorefineries to produce added-value products besides biofuels are studied. Four steps are therefore defined to reach this objective. The first step is to describe different aspects how process systems engineering can assist in improving the biobased economy. The second step is to develop a superstructure to identify promising pathways (economically and environmentally). The third step is to quantify the effect of uncertainty conditions of wastewater (composition and amounts) and sunshine duration on the amount of bioproducts and profits of these microalgae biorefineries. Lastly, the life cycle assessment (LCA) evaluates the environmental impacts of the promising pathways.

1.5 Thesis outline

An outline of this thesis is presented schematically in figure 1-3.

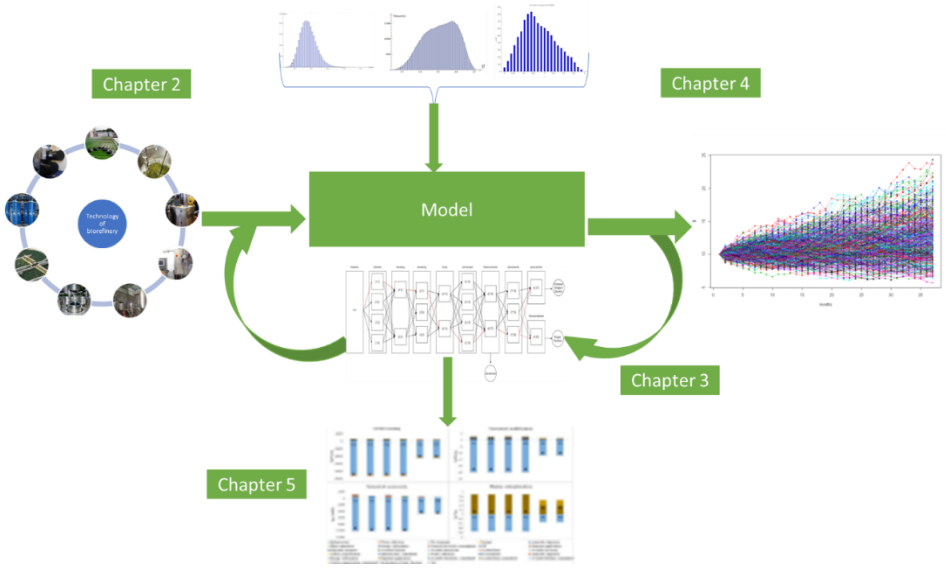


Figure 1-3: An outline of the thesis in schematic form

Chapter 2 presents an overview of the various technologies available in each step of a biorefinery for producing added-value products and biofuels from microalgae. In addition, these technologies are compared in terms of energy requirements and efficiency. Next, the role of process systems engineering in enhancing algae economies is discussed.

In chapter 3, a superstructure to produce added-value products (pigment, omega-3, glycerol, biodiesel, biogas, and fertilizers) from three species of microalgae (Chlorella Vulgaris, Haematococcus Pluvialis, Nannochloropsis spp.) is presented. The superstructure is optimized to find cost-effective production pathways for each microalga biorefinery. The operating and investment costs of these three microalgae biorefineries are compared. The annual profits margin of each of them are reported. Furthermore, the effect of extracting each bioproduct on annual profits are highlighted.

In chapter 4, the uncertainty conditions of wastewater (composition and amounts) and sunshine duration on the amount of bioproducts and profit margin of three types of microalgae biorefineries have been studied in this chapter. The Monte Carlo simulation study with real data of influent wastewater and duration of sunshine in the Netherlands is used. The probability of profits and bioproducts in different conditions of feedstocks are predicted.

Chapter 5 conducts life cycle assessments to investigate the environmental impacts of producing bioproducts from microalgae. The promising pathway of these three microalgal biorefineries is studied to evaluate them from different environmental impact aspects such as ozone depletion, acidification, global warming, etc. Furthermore, the ecological impact of different stages of the microalgae biorefinery is investigated.

Finally, conclusions and outlook of this thesis are presented in chapter 6 and chapter7, respectively.

Chapter Two

Microalgae Biorefinery

Abstract

In this chapter, basic characteristics of the various strains of microalgae are presented. In addition, the beneficial extracted bioproducts and their applications are reviewed. Then, an overview of the various technologies available in each step of biorefinery to produce added-value products and biofuels from microalgae is provided. These technologies are compared in terms of required energy and efficiency. Then, the role of process systems engineering in enhancing the algae economy is highlighted. Different perspectives of the algae industry, from molecule to enterprise scale, where process systems engineering can have a role, are addressed. Subsequently, the roles of process systems engineering in process and product design, process control, and supply chain of the algae biorefinery are discussed. It is found that process systems engineering can play an important role in the biobased economy, especially by applying sustainability and economic concepts in the decision-making process for selecting the best feedstock, processing pathways, and desired products. Tools such as market analysis, techno-economic analysis and life cycle assessment (LCA) can be applied to design sustainable algae biorefinery. There are, however, several challenges such as the lack of data, the complexity of optimization, and validation that should be addressed before using these tools.

2.1 Introduction

During World War II, medical emergencies prompted microalgae as a possible supply of protein and antibiotics. In the 1950s, the use of microalgae for the generation of hydrogen and methane began [15]. Later, the energy crises of the 1970s encouraged the use of this microorganism to generate renewable energy.

The Aquatic Species Initiative (ASP) was the first research and development program focused on producing biodiesel by microalgae. It was started in 1978 by the US National Renewable Energy Laboratory (NREL) and continued until 1996 [16]. However, Nihon pioneered the first large-scale cultivation of Chlorella species in the 1960s [17].

More recently, research has been conducted to use algae for treating wastewater and capturing carbon dioxide [18]. In addition, different studies have been done to design microalgae to produce essential products which have a crucial role in the global economy.

Although some progress has been made towards the medium to large-scale application of microalgae, the technical and economic feasibility is not comparable to the use of conventional raw materials and processes that are often fossil based.

This chapter addresses the role of process systems engineering in different aspects of the algae industry. First, the essential factors of growth and the potential applications are discussed. Second, the potential bioproducts are described. The possible technologies for each step of algae biorefinery are listed and compared. Then, the procedures of process systems engineering to design and operate algae biorefineries efficiently are explained. The current process systems engineering methodologies implemented to enhance the algae industry are addressed. Finally, the challenges and limitations of these tools are highlighted.

2.2 Microalgae

Algae are marine creatures utilized as primary feedstocks of the third generation of biorefineries (first and second-generation biorefineries use other types of biomasses such as sugar cane and animal fats, and lignocellulose, respectively). They grow with the help of sunlight, water, carbon dioxide (CO_2), and nutrients like nitrogen and phosphorus in wastewater or seawater. They are either single-cell or multicellular. Some of them do not have any roots, stems, or leaves and no sterile protection around reproductive cells [19]. Depending on the size, they divide into microalgae and macroalgae [20]. Macroalgae are made up of several cells; conversely, microalgae are a diverse community of microscopic photosynthetic species, several of which are found in unicellular form and various environments. Since ancient times, coastal populations have used macroalgae for multiple purposes, including food, feed, medical treatment, and fertilizer.

Microalgae can be classified into prokaryotic cyanobacteria, which lack nuclear structures, and eukaryotic algae, which have nuclear structures [21]. Green algae, red algae, and diatoms are the three main types of eukaryotic alga [19]. With over 200,000 species of algae described, including 50,000 microalgae, these organisms have a greater diversity than all terrestrial plants [22].

Microalgae biomass has several advantages over plant-derived biomass [23–25]. Most significantly, microalgae do not need (fertile) land and can be grown in (waste) water. The most significant benefit of utilizing (waste) water is that it is readily available throughout the year and inexpensive [26]. In addition, microalgae can be used to capture CO_2. Typically, 1.83 kg of CO_2 is needed to produce 1 kg of microalgae biomass [27]. They transfer around 3.8% of absorbed solar energy during cellular metabolism, compared to 0.5% for terrestrial crops. Microalgae have higher photosynthetic productivity than plants in general. Microalgae can grow at a much higher rate and reproduce more consistently compared to plants. As compared to soybean (0.4 tonnes $ha^{-1}y^{-1}$(tonnes per hectare per year)), rapeseed (0.7 tonnes ha^{-1} y^{-1} [27]), microalgae-derived biomass is usually higher in lipids (4.5-7.5 tonnes ha^{-1} y^{-1} [28]). Another benefit of this microorganism is the short harvesting time (between 1 and 10 days depending on the process), which allows for multiple or continuous harvests [29].

2.2.1 Microalgae Characteristics

Microalgae can develop in various environments, including autotrophic, heterotrophic, and mixotrophic. Depending on these, they use CO_2/ organic culture medium and light/ organic medium as carbon and energy sources, respectively [30,31]. The autotrophic microalgae absorb energy from natural or artificial light and utilize photosynthesis to generate organic matter while bio-fixing CO_2. In heterotrophic cultivation, microalgae grow in a dark environment by using an external source of organic matter. Eventually, mixotrophic means that the microalgae use photosynthesis and other carbon sources during their lifetime [32]. Many microalgae strains with different characteristics are available in each category.

Since the cellular contents (compositions of lipids, proteins, pigments, etc.) and growth behaviours of different microalgae strains are diverse, it is critical to choose the right one depending on the application. For instance, carotenoids, free fatty acids, and proteins are abundant in Chlorella [22,33]. Due to their high lipid content (14-63 percent of dry weight), they are also a promising source for biofuel processing. Therefore, specific microalgae characteristics such as the concentration of lipid and other different added-value components, growth rate, optimal growth conditions, and scale-up capacity should be considered when applying them at industrial scales [34]. Figure 2-1 compares the composition of 5 kinds of microalgae.

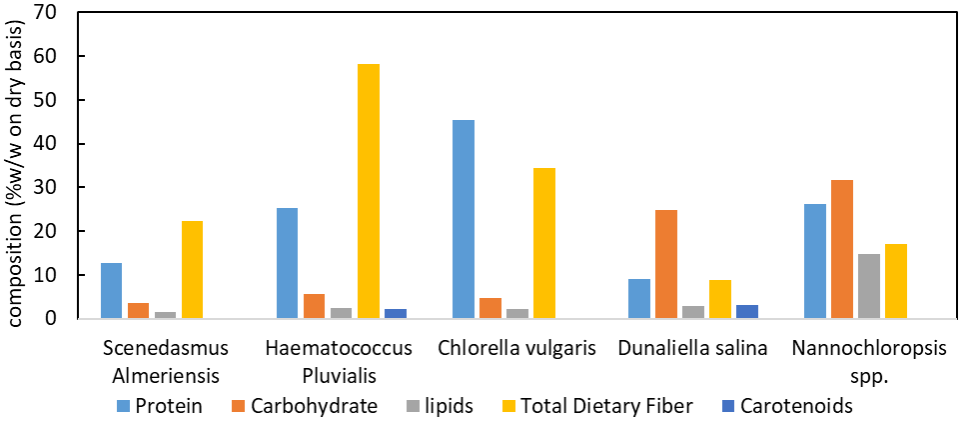


Figure 2-1: Compositions of different kinds of microalgae (Scenedasmus Almeriensis, Haematococcus Pluvialis, Chlorella Vulgaris, Dunaliella Salina, Nannochloropsis Spp. [35]

2.2.2 Environmental conditions for microalgal biomass production

Some nutrients are essential for growing microalgae. They should be provided in smaller or larger quantities to the culture medium to ensure species development and higher biomolecule synthesis. Carbon, phosphorus, nitrogen, and micronutrients are essential nutrients for guaranteeing a minimum of microalgae growth conditions [31,36]. One study

estimated the required nutrients for 29 species. It was claimed that the ratio C:N:P:S should be 132:18:1:0.99 [37].

Carbon is an essential nutrient for growing microalgae since it is a necessary component for forming all organic substances produced by the cell, such as proteins and carbohydrates. It can be delivered in salts (bicarbonate) or by infusing carbon dioxide. CO_2 must be solubilized before microalgae can use it for photosynthesis [38]. Beside carbon dioxide, nitrogen is an important building block for structural and operating proteins. Concentrations of carotenoids, proteins, and chlorophyll are related to these components. In addition, phosphorus plays an essential role in cellular metabolism. Pigment accumulation in certain microalgae may be caused by phosphorus deficiency, but the effect is less than that of nitrogen deficiency. Because of the role of iron in the transport of electrons during photosynthesis, this component is an essential trace element for microalgae growth. Besides those, potassium, silica, sulphur, metals, and vitamins are needed to cultivate microalgae [39].

One of the most important physical factors affecting microalgae growth is temperature. It affects cell composition. As the temperature drops, the degree of lipid unsaturation increases, and as the temperature rises, the pigment concentrations rise, but the concentration of oxygen radicals rises as well. If microalgae do not grow at their optimal temperature, the need for carbon and nutrients to maintain the same pace of growth becomes more critical [39,40]. Furthermore, the cellular composition of microalgae is affected by light. Generally, the best temperature (between 20- 30 centigrade) and the light irradiance (between 33-400 μ mol/m^2 /s) are reported for different algae species [41–43]. Light intensity, temperature, nitrogen sources, minerals, pH, and salinity, in addition to the intrinsic ability of algal species, influence the concentration of lipid and proteins in the microalgae.

2.2.3 Significant compounds produced by microalgae

2.2.3.1 Carotenoids

Although more than 750 types of carotenoids have been already discovered, only a small number have been economically marketed, and the most prevalent are β-carotene, astaxanthin, and lycopene [44]. The primary benefit of employing microalgae as a source of carotenoids is their good influence on human health, as many other antioxidant chemicals are found in algal cells.

Astaxanthin, as one of the non-provitamin A carotenoids, has lately attracted interest due to its antioxidant properties. This component can scavenge free radicals, protects against cancer, and has been linked to the healing of inflammatory processes and diabetes [45]. The Chlorella, Chlamydomonas, Dunaliella, and Haematococcus spp. can produce more astaxanthin in comparison to other types of microalgae. For instance, Haematococcus spp. can collect xanthophylls outside the plastids in the cytoplasm [46,47].

One of the non-provitamin A carotenoids that can be converted to retinol is β-carotene. This vitamin has an important role in reducing the risk of macular degeneration [48]. Chlorella spp., Chlorella Ellipsoidea, Coccomyxa Acidophila, Dunaliella Salina, and Scenedesmus Almeriensis are well-known microalgae that can produce these types of carotenoids [49].

Lycopene is a non-provitamin A carotenoid with a wide range of biological functions. Prevention of oxidative DNA damage, probable creation of carcinogen-metabolizing enzymes, decreasing risk for some malignancies, cancer prevention with inhibiting cancer growth, and certain cardiovascular events are all recognized some benefits of these added-value components [50].

2.2.3.2 Sterols

Some microalgae species have been utilized to promote the growth of oysters due to their high sterol content. Sterol levels are high in microalgae like Thalassiosira and Pavlova. Unusual sterols such as sitosterol, brassicasterol, campesterol, and stigmasterol are available in this microorganism. High cholesterol (e.g. LDL (low-density lipoprotein)) levels are well known to increase the risk of heart and coronary illnesses, which can be decreased by sterol [49].

2.2.3.3 Proteins and enzymes

Proteins are biopolymers of amino acids that cannot be made by the human body and are provided from external sources such as food. Proteins (smaller peptides and amino acids) have roles related to health and have nutritional advantages. Arthrospira and Chlorella are rich in protein and amino acid content, and they can be utilized as nutraceuticals or added to functional meals to help prevent tissue damage and disease [49].

2.2.3.4 Vitamins

The Haslea / Navicula ostrearia is exceptionally high in vitamin E in addition to marennine, a blue pigment that causes oysters to become green. The Porphyridium cruentum, another microalga, is high in vitamins C, E (tocopherols), and provitamin A (β-carotene). Dunaliella salina also generates tocopherol, thiamine, nicotinic acid, pyridoxine, biotin, and riboflavin, in addition to β-carotene (provitamin A) [49].

2.2.3.5 Polyunsaturated fatty acids (PUFA)

The two most common polyunsaturated fatty acids are omega-3 and omega-6. These are designated essential fatty acids because they participate in building and maintaining cell membranes. Omega-3 (such as EPA (Eicosapentaenoic acid) and DHA(Docosapentaenoic acid)) has been shown to lessen the risk of cardiovascular strokes and arthritis and decreases blood pressure. Omega-3 also helps reduce cholesterol levels by lowering triglycerides,

increasing HDL (high-density lipoprotein) levels, and acting as an anti-inflammatory agent [51].

Even though marine fish oil is the conventional source of both EPA and DHA, research suggests that algae can provide higher quantities of omega-3. The DHA is found in various species, including Schizochytrium, Crypthecodinium, and Thraustochytrium, and the EPA is discovered in Phaeodactylum, Chlorella, and Monodus [52].

2.2.3.6 Fertilizer

Fertilizer is another bioproduct from algae that can have an important role in the agricultural industries. Algae contain the required nutrients for growing plants [53]. For example, dry biomass derived from Acutodesmus Dimorphus can be used as a biofertilizer and increased tomato plant growth [54]. In another study, Chlorella Vulgaris was used as a biofertilizer for growing lettuce plants. The results show that dry powder of microalgae might be employed as plant nutrients for optimal growth since they can increase soil fertility [55]. Furthermore, the effect of two microalgae (Spirulina Platensis and Chlorella Vulgaris) on the production of the maize crop was studied by Dineshkumar et al. According to their findings, maize production and growth can be increased up to 51.1 percent 60 days after planting [56].

2.2.3.7 Fuels

Three fundamental fuel types can be delivered by microalgae: biomethane, biodiesel, and bioethanol. The simultaneous generation of different biofuels may decrease the cost of a single fuel [57]. In addition, to extract the lipids in biodiesel production, the residual algae biomass can be transferred to an anaerobic digestion plant to produce biomethane. Furthermore, carbohydrates and proteins that are not removed during the biodiesel production process might be utilized to make bioethanol.

Table 2-1 lists several added-value products from microalgae, their application, and the predicted annual global market value. As can be seen, the market of these added values is high enough to attract investors.

Table 2-1: Global market and application of some added value products extracted from microalgae

Added value products	Application	Global market size (billion USD)
Astaxanthin	• Aging skin • Muscle soreness from exercise and athletic performance • Alzheimer disease	0.2 [58]
Lutein	• Eye diseases	0.23 [59]
β-carotene	• Source of vitamin A	0.2 [58]
Chlorophyll	• Food industry (color confectionery, gelatine, and drinks)	1 (natural food collars markets)[60]
Phycobiliproteins	• Food industry • Pharmaceuticals and cosmetic industry [61]	0.05 [58]
Vitamins, minerals, and nutrition	• Food industry • Pharmaceuticals, cosmetic industry	82 [62]

2.3 Algae biorefinery

The term biorefinery has been utilized in the literature since the 1980s and alludes to the coproduction of a range of added-value products and bioenergy from biomass [63]. Many budgets are specified for various research projects to enhance algae biorefinery. For instance, the US Department of Energy issued a $100 million grant for three organizations to investigate algae biorefineries in December 2009 [64]. The University of Greenwich in the UK with 14 European partners, started one project named ‘D-factory’ in December 2013, which the European commission funded. Approximately 10 million euros were specified for this project to improve Dunaliella microalgae biorefinery [65].

The algal biorefinery approach offers a comprehensive methodology to various products, with the added benefit of using all algae components and creating numerous revenues [66]. Upstream and downstream processing are the two main phases of the microalgae biorefinery. The upstream process consists mainly of microalgae cultivation. Harvesting, extraction, and purification of bioproducts are considered downstream processes [67]. Figure 2-2 shows the overall concept of the algae biorefinery and the possible bioproducts. As can be seen, depending on the type of algae, various products can be extracted simultaneously.

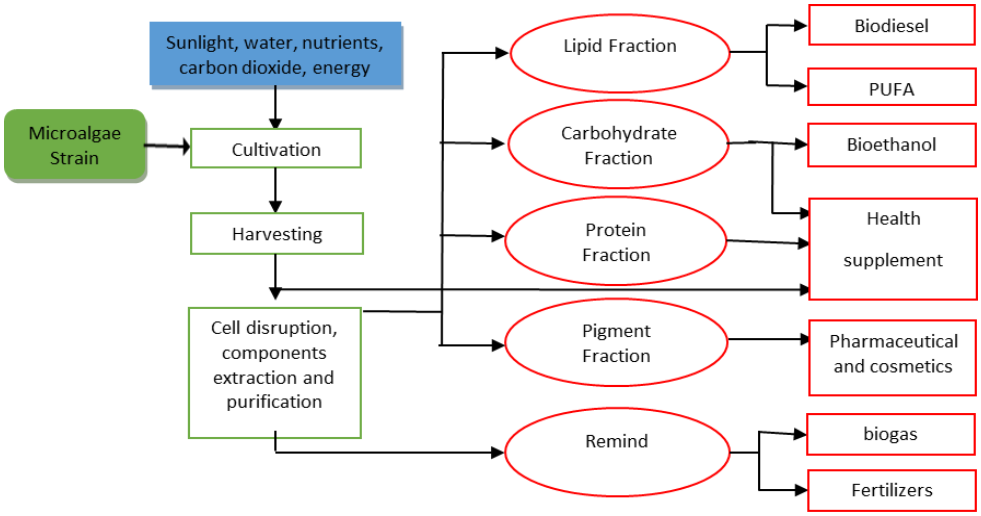


Figure 2-2: The microalgae biorefinery with the possible bioproducts [67]

Cultivating microalgae for biofuels may not be economical, and the microalgal industry must take advantage of markets for added value products such as nutraceuticals and vitamins. The combined extraction of numerous added-value components from a single microalgal slurry with biodiesel moves the general sustainability of the system forward [68,69].

2.4 Steps in the biorefinery

Various techniques can be employed at each step of the algae biorefinery (cultivation, harvesting, dewatering, cell disruption, drying, extraction, transesterification, and treatment) to produce added-value products and biofuels.

After growing microalgae in the cultivation step, they are concentrated in the following steps: harvesting, dewatering, and drying. The concentrated microalgae substance enters a cell disruption step to break the cell walls and facilitate the valuable components' extraction. Different techniques can be used to extract lipids, pigments, and proteins. The extracted components require further treatment to produce final products. These steps and current techniques of each of them are explained below.

2.4.1 Methods of algae cultivation

Microalgae can be grown at industrial scales in an outdoor or indoor system [27]. The raceway is an outdoor system, while the photobioreactor is an outdoor or indoor system. Although the investment cost of an outdoor system such as a raceway is not high, controlling operational conditions is difficult. Growing efficiency depends on the location and environmental conditions, including temperature and light. Another disadvantage of this type of cultivation system is the large area requirement with a low microalgae concentration [70]. The major advantages and disadvantages of the raceway and the photobioreactor are listed in table 2-2 [71].

Table 2-2: Comparison of raceway and photobioreactor [71]

Type of cultivation method	**Advantages**	**Disadvantages**
Raceway ponds	Low energy requirements and capital costs, Dissipation of heat with evaporation	High risk of contamination, Dependency of productivity on the location/ environment of the pond, High water evaporation, Large land requirements, High upstream costs
Photobioreactor	High surface ratio, Controlling growing conditions by adjusting temperature, pH, etc., Implementing artificial light instead of sunlight, Low upstream cost	High energy requirements and capital costs, Increase shear stress (flat plate photobioreactor), Risk of carbon dioxide depletion and oxygen accumulation (tubular photobioreactor), variation of pH (tubular photobioreactor)

The flat plate photobioreactor, tubular photobioreactor, and bubble column photobioreactor are the three most common types of photobioreactor [72]. Due to high production rates and short harvest periods, the tubular photobioreactor is the most promising technology. It comprises a series of straight clear tubes, often composed of plastic or glass. To catch the most solar light energy, the tube arrays are positioned in various configurations (vertically, inclined, horizontally, or as a helix). The diameter of the solar collector tubes is less than 0.1 m to enable deep light penetration with the presence of thick cultured broth inside the tubes [73].

Although the flat photobioreactor is one of the earliest types of a closed system, it is widely used at a laboratory scale. It is made up of thin glass panels arranged horizontally or occasionally in an inclined orientation. This configuration makes it easier to quantify irradiance at the culture surface [72], but it is challenging to construct at industrial scales [74].

The bubble column photobioreactor is a hollow cylinder reactor made from glass. Due to the structure of the column (long optical paths), the dependence of yield of production on light penetration is very low. Furthermore, this structure increases surface area, allowing for the most algae growth compared to other types of photobioreactors [29].

2.4.2 Methods of separating microalgae biomass

Experimental results show that the algae concentration in the output flow of the cultivation step is typically between 0.1 and 3.0 g L^{-1} [75]. Separating water from the culture medium is necessary, but it accounts for 20–30% of the total production costs. For this reason, the types of technologies used for this purpose have significant effects on the economics of the process [76]. Harvesting/dewatering methods, e.g., gravity sedimentation, filtration, coagulation/flocculation, flotation, and centrifugation, are applied to increase the density of microalgae slurry. Drying methods such as spray drying, solar drying, greenhouse drying, and lyophilization are needed to obtain dried microalgae biomass.

Gravity sedimentation is a straightforward, cheap, and common technology for harvesting microalgae. However, this method also has disadvantages: it is time-consuming, has a high environmental temperature requirement [77], and only used for large algae cells such as spirulina [19].

In flocculation, microalgae cells are aggregated to form flocs, which causes them to settle faster. Although flocculants are needed, the energy requirement for this harvesting method is very low. A flocculating agent can be categorized into chemical flocculants, physical flocculants, and bio-flocculants [71]. A chemical flocculant consists of metal salts, polymers, and/or biopolymers. In electrocoagulation–flocculation, magnetic nanoparticles can be used as physical flocculation methods. In the bio-flocculation method, microorganisms such as

bacteria are assisting the flocculating of microalgae. The flocculation bacteria can proliferate by using the wastewater as a carbon source. Bio-flocculation is a safe and eco-friendly approach with more than 90% harvesting efficiency [78,79].

Flotation is a process of utilizing air or gas bubbles to transfer the liquid-solid suspension to the surface. These bubbles are generated to aggregate microalgae cells on the surface to separate them from water [80]. Dissolved air flotation, electrolytic flotation, and dispersed flotation are the most common flotation technologies [79,81]. Generally, the flotation method is very energy intensive.

Centrifugation is another technique to dewater the algae biomass. A centrifugal force separates microalgae from the culture medium based on the density and size of the components [82]. Although the efficiency of this method is very high (more than 95 %), it requires high operational costs and is very time-consuming to achieve desired separation efficiency [25,83]. Two types of centrifugation are available: fixed wall (e.g., hydrocyclone) and rotary wall (e.g., tubular centrifuges, centrifugal decanters, and disc centrifuges) [81].

Large microalgae cells (> 70 μm) can be separated from the cultivation medium by (semi) permeable membrane filtration[19]. The filtration method is prone to fouling and clogging. Thus, it has a high operating cost, and there is a need to replace and clean the filters frequently [76].

Drying is an expensive and energy-intensive part of the biorefinery. Solar and wind dryers are the most economical approaches. However, they require large spaces, and their efficiency depends on environmental conditions such as airflow, temperature, and humidity. Besides natural dryers, many artificial dryers have been proposed for microalgae drying [84,85]. The spray dryer is the most common one. Although it is costly, its energy and operational efficiency are high. Several advantages and disadvantages of the drying techniques are listed in table 2-3. The appropriate type of dryer is chosen based on the scale of operation, energy and cost requirements, other downstream processes, and the kind of microalgae [84].

Table 2-3: Advantages and disadvantages of various types of dryers [21,84]

Type of dryer	Advantages	Disadvantages
Freeze dryer	Gentle process	Cost intensive, Slow process
Sun dryer	Low running costs and very low capital cost	Weather dependent, Slow process
Drum dryer	Fast and efficient	Cost intensive
Spray dryer	Fast and efficient	Cost intensive
Crossflow dryer	Faster than sun dryer and cheaper than drum dryer	High electricity requirements
Vacuum shelf dryer	Gentle process	Cost intensive

2.4.3 Methods of algae cell disruption

Cell disruption is another crucial step in the biorefinery before extracting pigment, lipid, or other components. An appropriate cell disruption method can reduce the costs for extraction significantly [86]. There are two methods for breaking down algae cell membranes: mechanical and non-mechanical [87] methods. The non-mechanical methods use enzymes, chemical breakdown, and osmotic shock. The most common mechanical methods are bead mill, ultrasonic, high-pressure homogenization, and microwave. Although the energy consumption for the mechanical methods is higher than the non-mechanical ones, the non-mechanical methods have several other advantages, such as being fast and monitorable for use at industrial scales [87,88].

In bead milling, a rotating cylinder made of quarts or metal damages microalgae cells within less than minutes without any preparation. However, it must be noted that several factors such as the shape and size of the cylinder and the stirring speed affect this method's efficiency and energy consumption. Because this is a high-speed technique with a simple setup, it is often used to extract lipids from microalgae at a large scale [81].

Ultrasonication combines two mechanisms to break the cell walls: cavitation and wave propagation. High energies are needed in this method due to the cooling and power of ultrasound. To have effective cell disruption, the frequency should be in the range of 18-40 kHz or 400-800kHz. Due to heat dissipation, the temperature should be controlled [81,87].

High-pressure homogenization is a scalable and straightforward method to disrupt microalgae cells. In this method, the cell is driven through a tiny orifice where mechanical forces such as turbulence, cavitation, and shear stress induce cell lysis. This method’s

efficiency varies and is directly linked to the type of microalgae [89,90]. The downsides of this method include long processing times, a substantial amount of cell debris that might create a problem for extraction, and the high energy requirement [71].

In the microwave method, electromagnetic radiation waves (between 0,3- 300 GHz) break microalgae cells by inducing the interaction of heat and molecules [91]. However, the high temperature could create problems for the biodegradable components in the algae, such as lipid and fatty acids. Another disadvantage of this method is the very high energy requirement on a large scale. Although the microwave method has short extraction times, it might require solvents [92,93].

Furthermore, solvents, salts, surfactants, nanoparticles, and acids may break the microalgae cell chemically. The efficiency, suitability, and selectivity of these chemical materials strongly depend on the cell wall structure and types of microalgae [87]. Although chemical materials must be used continuously to disrupt the cell, the heat and energy requirements are low compared to mechanical methods.

The osmotic shock is considered a non-mechanical method to disrupt the microalgae cell. In this method, the osmotic pressure balance between the interior and exterior of the cell is disturbed by changing the suspension salt concentration. To damage the cell, two technologies, hyperosmotic or hypoosmotic stress, increasing or decreasing the concentration of salt outside of the cell, respectively, can be utilized. Although low-cost chemical salts, e.g., sodium chloride and sorbitol, are used in these approaches, a large amount of water is needed for dilution [81].

In addition, biological techniques incorporating suitable lysis enzymes (such as lipase, protease, and cellulase) or algicidal treatment, including bacteria, viruses, and cyanobacteria, can be used to disrupt microalgae cell walls. The biological selectivity, mild operating conditions, and low energy requirements are the main advantages of this technique. However, several challenges exist and prevent large-scale implementation, such as long processing times and the high cost of enzymes [94].

2.4.4 Methods of extraction

An appropriate extraction method would be more selective in extracting certain microalgae components while minimizing contaminants in co-extraction. Extraction of biochemicals has been done in a variety of ways. To extract bio components, two approaches are common: organic solvent extraction and supercritical solvent extraction (e.g., supercritical CO_2).

2.4.4.1 Supercritical solvent extraction

Supercritical fluid (such as CO_2) extraction is a new green technology that has received considerable interest and increased popularity in recent years due to the possibility of

replacing highly toxic organic solvents. A supercritical fluid appears to be a suitable extraction solvent for extraction bio components due to high selectivity while it has both gas and liquid properties. In addition, further treatment is not needed after extraction[92]. Carbon dioxide waste from industry might be utilized under supercritical circumstances, saving money and being better for the environment [95]. The main drawback of this technique (compared to organic solvent methods) is that it requires a lot of energy [96].

Lipids of microalgae can be extracted by supercritical carbon dioxide. After extraction, CO_2 evaporates into the atmosphere, and the extracted lipids are precipitated. The performance of this technique depends on the type of microalgae, duration of extraction times, temperature, and pressure of the process. For instance, one study showed that increasing the pressures of supercritical fluids enhances the amount of lipids extracted from Chlorella Vulgaris[97].

Goto et al. considered extraction of astaxanthin from Haematococcus Pluvialis with supercritical CO_2 and ethanol as entrainer under different pressure conditions. The addition of ethanol, which increased the solubility of components in supercritical CO_2, resulted in a considerable improvement in extraction efficiency. Approximately 80% of astaxanthin can be extracted using 5% (v/v) of ethanol under 40 MPa and 40 °C. Although acetone can also be used as an entrainer, ethanol is preferable due to its lower toxicity[98].

2.4.4.2 Organic solvent extraction

Organic solvents can also extract bicomponent such as β-carotene, astaxanthin, and lipids. Hexane, butanol, chloroform, acetone, and methanol are the most common solvents to extract different lipids. The polarity of the solvents defines the types of extracted lipid. For instance, hexane can extract hydrocarbons and triacylglycerols. Due to that, a mixture of solvents (for example, a combination of chloroform, methanol, and water or a mixture of chloroform and methanol) is proposed to extract all lipids classes [81].

The extraction of carotenoids from microalgae has recently been studied using many green solvents such as ethanol and a mixture of organic solvents [99]. Damergi et al. considered carotenoid extraction with ethanol and 2-methyl tetrahydrofuran (MTHF) solvents. 45% and 66% of the total carotenoids can be extracted using pure MTHF and a mixture of ethanol and MTHF (1:1), respectively [100].

The resultant extraction process is a combination of solvents, residual water, biocomponents (lipid, pigment, etc.), and cell debris. A solid-liquid or liquid-liquid separation process is implemented to separate the components in this mixture from each other. Subsequently, some lipids such as triacylglycerol, free fatty acids, and phospholipids can be converted into biodiesel by transesterification reactions [95]. Furthermore, other processes such as liquid-liquid extraction can be utilized to separate Omega-3 (methyl eicosapentaenoic acid and

methyl docosahexaenoic acid) from biodiesel [101]. Finally, the cell debris is sent to anaerobic digestion to produce biomethane and fertilizers.

2.5 Role of process system engineering

Algae biorefinery as a burgeoning industry must be designed and operated effectively and efficiently to survive in today's highly competitive environment. Process systems engineering combines science with engineering to provide the required methods that allow this industry to prosper [102]. It offers a molecule to enterprise systems view for the decision-making over the entire algae value chain [103]. In this field, molecular and microscopic (e.g., microalgae cell) discoveries are tied to strategies and logistics for manufacturing and production [102]. Optimizing each element of this chemical supply chain (molecule, process units, plant, supply chain, etc.) is essential. Process systems engineering methodologies and technologies provide the necessary means to produce integrated solutions near the global optimum [104]. Different aspects of the algae industry, where process systems engineering can play a role in, are shown in Figure2-3.

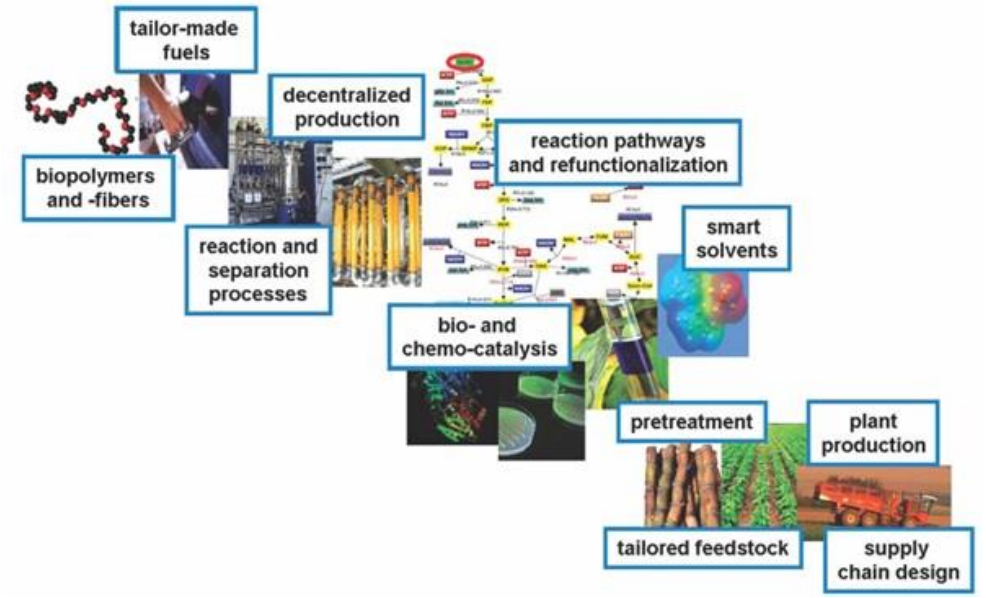


Figure 2-3: Set of interacting systems problems: from biorenewables production to novel generated molecular products [104].

The extensive dependencies between cell composition of microalgae, its pretreatment, processing pathway into target bioproduct, and end bioproduct structure-property relationships necessitate a comprehensive approach [104]. Microalgae cell compositions depend on the selected microalgae strain and cultivation condition (e.g., pH, temperature,

etc.). The composition of the cell also depends on the duration of cultivation. It is possible to manage the quality of algae biomass within the ecological restrictions to perfectly fit the valorization process. Optimizing cultivation land area and operation conditions and maximizing usage of carbon dioxide are some issues that can be solved with process systems engineering.

Process systems engineering interlinks decision-making at the molecular level to the process and/or product synthesis and design level. Knowledge-based or heuristics approach can be used as a technique for this purpose [105,106]. The process synthesis and design approach are to develop computationally efficient models based on mathematical programming for each unit operation to improve efficiency. In product synthesis and design, the current focus is on selecting suitable solvents for various chemical reactions [107].

In addition, process systems engineering supports identifying optimal process configurations, thus contributing to the development of novel process intensification technologies. Process intensification dramatically reduces the energy consumption and processing costs of chemical processes by taking advantage of multiple multifunctional phenomena at different spatial and temporal scales and improving transfer (momentum, mass, and heat) rates. Thus, the configuration and size of different equipment of the process can be optimized by this approach [108].

Process control, as an integral part of process system engineering, traditionally has been limited to regulating key variables (around certain predetermined operating conditions or trajectories) to ensure that products of the required specifications are produced in the presence of disruptions and design errors. With the expansion of its scope, it now deals with economic issues, such as reducing energy consumption and enhancing productivity. It is important to simultaneously think about regulation and economic optimization since this tends to drive the process operation towards an intersection of constraints where precise control is essential for success [102,107].

Optimization of supply chain and planning is another area where process systems engineering has an important role. To remain competitive, algae biorefinery must now optimize operations across the entire supply chain. Supply chain performance has been improved greatly due to globalization and modern telecommunication technology [109]. Furthermore, supply chain optimization provides various academic research opportunities through industrial needs and fosters and strengthens many industry-academia collaborations since it has strong industrial relevance [110]. For example, the supply chain of algae biorefinery under uncertainty can be studied with the Best-Worst Method (BWM) and a mathematical model. In addition, robust optimization is a field of interest that can be used to map uncertainty in demand. In addition, multi-objective optimization aids decision-makers in realizing the trade-off between benefits and investment [91].

Different process systems engineering tools can be applied to design a sustainable algae biorefinery. These tools include market analysis, techno-economic analysis and life cycle assessment (LCA), and supply chain (SC) analysis. They are used to analyze the performance of algae biorefinery from economic, social, and environmental aspects [111].

The market analysis consists of two steps. First, depending on the company's competitive position (e.g., financial status), the possible markets in its area, access to feedstock, current processes, and SC assets, lists of potential bioproducts, processes, and partners are developed. In the second step, these products are ranked with SWOT (strengths, weaknesses, opportunities, and threats) approaches. The ranked products are the result of an analysis study that can be evaluated with other tools such as techno-economic analysis and LCA [111,112].

The techno-economic analysis considers all possible technologies that can be implemented in algae biorefinery steps to produce targeted bioproducts. Different production pathways are presented by defining various technology options for each step of the biorefinery. These technologies are gathered in a superstructure. The superstructure is optimized based on multiple objectives to propose an appropriate production pathway. The superstructure is a graphical representation of all possible processing pathways and is converted to a mathematical model and solved with the appropriate software (often as a mixed-integer (non)linear programming problem). Pinch analysis can be used to investigate possible ways to integrate the biorefinery into the current company process and consider the effect of process options [111–113].

An LCA can be conducted on the production pathways and related products to assess their environmental impacts. First, functional units and system boundaries of the case study are defined based on the goal and scope. Then, the mass and energy quantities of the case study convert to different impact categories such as local, regional, global, etc., named life cycle impact assessment (LCIA). It is performed by translating the life cycle inventory to environmental impacts. Finally, the results of the environmental impacts on the case study for different criteria such as greenhouse gas emissions are determined for further multi-criteria decision-making [111,112].

SC analysis considers the price and demand volatility of feedstocks and products and evaluates its profitability under various market scenarios. The SC indicators have been created to quantify the SC's resilience and adaptability in the face of market volatility in a dynamic market. A robustness metric is calculated to measure each case's resilience under uncertain market conditions. The parameter determines how far the downside profits deviate from the base case profit. Furthermore, a flexibility metric was employed to demonstrate how much the production volume deviates from the nominal production rate [111].

Process systems engineering will face different challenges when using these tools for developing and designing commercial-scale algae biorefinery. First, the sustainable design

depends on choosing an appropriate feedstock, a product portfolio, a location, and scale, which are all affected by government policies, environmental conditions, and the current market situation. Second, many physical and chemical data are needed to design and simulate the process of algae to bioproduct/biofuel. Due to the varying composition of algae and the complex structure, these data are not available completely and sufficiently. Some of these data should be reported by researchers in other fields (e.g., agriculture); this makes process systems engineering a multi-disciplinary field. Third, actual operation data of algae biorefinery are not available due to few available industrial scales of algae biorefinery. Thus, validating the algae biorefinery model/simulation encounters many barriers. Furthermore, numerical methods are needed to optimize algae biorefinery superstructure. Solving these complex models is another critical challenge [112,113].

Conclusion

Microalgae are a rich source of added-value components such as pigments and lipids (e.g., omega-3 polyunsaturated fatty acids), with significant health, energy, food advantages. To extract these bio components, microalgae should pass various steps of algae biorefinery. Different technologies, including raceway, photobioreactor (cultivation), flocculation, centrifugation (harvesting/dewatering), spray dryer (drying), cell mechanical and non-mechanical methods of cell disruption, and green and organic solvent extraction can be implemented. These technologies are compared from different perspectives such as economics and energy.

Various process systems engineering tools such as market analysis, techno-economic analysis, LCA, and SC analysis can be applied to improve the microalgae industry. These tools consider a molecule to enterprise systems view for the decision-making over the entire algae value chain.

Chapter Three

Development of a superstructure-mathematical model

Abstract

A superstructure to produce added-value products (pigment, omega-3, glycerol, biodiesel, biogas, and fertilizers) from three species of microalgae (Chlorella Vulgaris, Haematococcus Pluvialis, Nannochloropsis spp.) is developed in this study. The superstructure is converted into a mixed-integer nonlinear programming (MINLP) model. A block integration approach is used to drastically decrease the CPU times by reducing the number of variables, parameters, and constraints. The model is solved with Baron/AOA in AIMMS software, and the most promising production pathway is identified. For all three biorefineries (cultivating different microalgae), the most promising production pathways (in terms of cost-effectiveness) are the same and consist of an open pond, sedimentation and flotation, flocculation without any dryer, sonication, organic solvent pigment extraction, n-butanol solvent lipid extraction, lipid production, and anaerobic digestion. Changing technologies of dewatering stages (flocculation to centrifugation and filter press) proposes the second and third cost-effective production pathways. The most profitable biorefinery cultivates Haematococcus Pluvialis, with annual profits of 62 $/kg of microalgae. A high amount of valuable pigment produced by Haematococcus pluvialis leads to 22 times higher profits than Chlorella Vulgaris and 47 times higher than Nannochloropsis spp. This biorefinery produces approximately 500 t of pigment bioproducts from 24 Kt biomass by using 200 Kt of wastewater and 164 Kt of carbon dioxide, annually. Ultimately, a sensitivity analysis is executed to confirm how the production of pigment and the price of this bioproduct, and day/ night ratio affect the profitability of microalgae biorefineries.

3.1. Introduction

A growing interest has been directed toward microalgae as a promising sustainable feedstock [114]. Microalgae can be used to produce various types of biofuels such as biodiesel and biogas only by using carbon dioxide from the atmosphere/flue gasses and (waste) water [115]. Thus, microalgae growth is accompanied by reductions in carbon dioxide emissions. Furthermore, microalgae have more advantages as compared to other biomass sources. For example, they can grow in nonarable land, and the duration of cultivation is very short [27]. However, their applications at industrial scales are not yet recommended due to high bioproduct costs. To overcome this problem, the search for producing appropriate bioproducts from microalgae and to improve the efficiency of such biorefining process is in progress [53,116].

A microalgae biorefinery consists of various processing stages, including cultivation, dewatering/ harvesting, drying, cell disruption, and bioproduct extraction. Microalgal biorefinery can be constructed in a variety of ways by considering different options for each processing stages and different bioproducts [117].

Hierarchical decomposition [105,106] and superstructure optimization [118,119] are two approaches used for the conceptual design of a process. In hierarchical decomposition, the design of the process is gradually defined at different levels and stages. A hierarchy of decisions can be made at each level based on defined factors. A disadvantage of this approach is the complexity of interaction between various decisions at different levels, slowly adapt to changing conditions, which decreases decision-making speed [120,121]. Superstructure optimization, on the other hand, addresses the simultaneous design challenge as a mathematical programming problem. Superstructure optimization is preferred for systematically evaluating a large space of structural alternatives [122–124].

Superstructure optimization consists of three main steps. The first step is to develop a superstructure with all the potential alternative processes over the different processing stages. The second step is to transfer the superstructure to a mathematical model containing mass and energy balances. The third step is to solve the mathematical model to find the optimal structure for a given objective, for example an economic or environmental measure [118]. Gebreslassie et al. propose a superstructure for algae-based hydrocarbon biorefinery. The superstructure is optimized as a mixed-integer nonlinear programming (MINLP) model to simultaneously minimize global warming potential (GWP) and maximize net present value (NPV). The superstructure consists of 5 major stages. Although an optimal pathway for biodiesel production is proposed, alternative energy-efficient technologies with an expanded superstructure can also be considered [125]. Gong et al. consider microalgal biorefineries with various production pathways. With the optimized environmental pathway, biodiesel costs are 9.712 $/GGE (gasoline gallon equivalent), whereas the price with the optimized economic pathway is 7.017 $/GGE. To increase the efficiency of the computational model, a linear approximation with partition points is defined. This approach is not very useful for large problems. Further improvements to decrease the number of partition points are needed [126]. Rizwan et al. optimize the superstructure of a microalgae biorefinery to produce biofuel from Chlorella Vulgaris. 1440 production pathways are evaluated in the GAMS software. Although an optimized route has been found, the profits of this microalgae biorefinery are below the breakeven point. Hence, biofuel production in this biorefinery is not economically viable [127].

Cheali et al. propose an optimal processing pathway for producing protein, ethanol, and biodiesel from microalgae. In this superstructure, 1920 production pathways are evaluated to maximize profit from only producing biodiesel [128]. Prieto et al. formulate an MINLP to optimize the superstructure for producing added-value bioproducts (biodiesel, poly-hydroxybutyrate (PHB), and astaxanthin) from microalgae. Maximizing the NPV is the objective of this study. The results show that simultaneous bioproduct production increases biodiesel's economic feasibility [129]. Fasahati et al. investigate the economic and technical feasibility of producing biochemicals in cyanobacteria biorefineries using a superstructure-

based approach. The results show that using wastewater to grow microalgae can enhance the economics of the process [130]. Galanopoulos et al. propose an MINLP model to optimize superstructure to minimize the total biodiesel production costs of an integrated algae biorefinery. Producing bioethanol, glycerol, and levulinic acid reduces 20% of the cost of biodiesel [131]. The cost of biodiesel decreases significantly, but the algae biorefinery's profitability is not high enough to be commercially viable.

In either of these studies, no production pathway has been proposed to simultaneously produce economically viable biofuels and bioproducts. The low profits from algae biorefineries keep the conventional refineries from switching to new versions (algae biorefineries). Using biochemicals that can be extracted during biodiesel production can effectively reduce the cost of biofuels/bioproducts. Depending on the type of microalgae, these biochemicals are different. Consequently, each microalga biorefinery's profit potential and optimal production pathway are distinct. However, two major obstacles exist when considering algae biorefineries. Firstly, the suggested microalgae biorefinery superstructures are restricted to a few options for each stage. They are not focused to produce different bioproducts simultaneously. Second, it is challenging to develop a mathematical model that fully describes the process. Additionally, solving the model is complicated and time-consuming and requires professional software.

3.2. Methodology

In this work, a superstructure of a microalgae biorefinery with all current alternatives for each processing stage is developed to find cost-effective pathways for producing various bioproducts (such as pigment, omega-3, glycerol, biodiesel, biogases and fertilizers) from three common microalgae (Chlorella Vulgaris, Haematococcus Pluvialis, and Nannochloropsis spp.). The superstructure is converted to a mathematical program that is optimized for a cost-objective function.

First, the problem statement of this study with all given data, condition/ assumption, decision, and objective variable is explained. Secondly, the production processes of various added-value products are described, and a superstructure is proposed. The mathematical program with all required parameters is given. Thirdly, a new block integration approach is explained, which reduces the program size.

3.2.1. Problem statement

Given is:

- composition and quantities of feedstock (wastewater and carbon dioxide gas),
- composition of bioproducts extracted from each microalga,

- composition and growth condition of each microalga,
- processes of extracting different bioproducts from microalgae,
- current technologies in each stage of microalgae biorefinery,
- superstructure with all current alternatives and production pathways,
- equipment performance parameters (split factors and yields),
- economic specification of each alternative (CAPEX/OPEX, Lang factors, interest rates).

Under the following conditions and assumptions:

- each alternative is associated with binary decision variables;
- mass and energy balance equations are applied for each block;
- investment costs are calculated based on economy of scale;
- other components of wastewater do not have any positive/negative effect on microalgae growth conditions;
- pure carbon dioxide gas is used;
- the rate of growing microalgae at different temperatures and PH of the cultivation area is fixed;
- the cost of transporting materials has not been considered, and the cultivation plant is located near the algae biorefinery;
- steady-state, and values of parameters/ variables are constant over time;
- the mass flow rate and energy consumption have a linear relationship.
- procedures of lipid and pigment extraction are the same for different types of microalgae

The following decisions are made:

- processing alternative for each stage;
- quantities of produced bioproduct;
- utility consumption;

The objective is:

- maximize the total profit margin of the algae biorefinery

3.2.2. Process description and superstructure development

Carbon dioxide and wastewater are needed as feedstock for cultivating microalgae. It is assumed carbon dioxide is supplied by external supplier and injected into the cultivation area to prepare the carbon required for growing microalgae. Furthermore, wastewater that contains water and nutrients enters the microalgae biorefinery to produce microalgal biomass. For this study a hypothetical biorefinery that can process 10% of the total mass flow of Dutch influent wastewater for 2018[132]. Average quantities and compositions of influent wastewater are shown in table C-1.

Microalgae grow under autotrophic conditions and are unaffected by elements such as arsenic and mercury. The influent wastewater contains nitrogen and phosphorus compounds. It is presumed all nitrogen and phosphor solve completely as NH_4^+ and PO_4^{3-} ions, respectively. Another required ion, sulphate, is added externally as a pure component. In addition, it is assumed that all the essential ions are consumed completely, and unusable nutrients leave the microalgae biorefinery as waste flow.

Three types of microalgae are considered in this study (Chlorella Vulgaris [133], Haematococcus Pluvialis[134], and Nannochloropsis spp.[135]). Data from biological studies are utilized to define the chemical structural formula of each microalga. These formulas can be found in table 3-1. The average mass percentages of lipids, pigment, and other cell components of each type of microalgae are shown in table 3-1.

Table 3-1: Chemical formula and composition of microalgae cell[131,134–139]

Type of microalgae	**Chemical formula**	**Percent of total cell weight**		
		Lipid	**pigment**	**Another cell components**
Chlorella Vulgaris	$CO_{0.48}H_{1.82}N_{0.11}P_{0.01}S_{.001}$	12%	2.53%	85.47%
Haematococcus Pluvialis	$CO_{0.38}H_{1.65}N_{0.12}P_{0.005}S_{.007}$	15%	3.18%	81.82%
Nannochloropsis spp.	$CO_{0.54}H_{1.77}N_{0.11}S_{.006}$	18.36%	0.16%	81.48%

Phosphate, and ammonia ions of wastewater are used for regenerating and growing microalgae based on the reactions (3.1), (3.2) and (3.3). These reactions show the mass stoichiometric coefficient of each type of microalgae.

$$0.28\ H_2o + CO_2 + 0.04NH_4^+ + 0.02PO_4^{3-} + 0.002\ SO_4^{2-} \rightarrow 0.82O_2 + 0.53\ Chlorella\ Vulgaris \quad (3.1)$$

$$0.24\ H_2O + CO_2 + 0.05NH_4^+ + 0.01PO_4^{3-} + 0.02\ SO_4^{2-} \rightarrow 0.82O_2 + 0.50\ Haematococcus\ Pluvialis \quad (3.2)$$

$$0.28\ H_2O + CO_2 + 0.045NH_4^+ + 0.01\ SO_4^{2-} \rightarrow 0.78O_2 + 0.55\ Nannochloropsis\ spp. \quad (3.3)$$

In addition, sunlight is another requirement for growing microalgae. A fixed day/night ratio of 0.5 is assumed. i.e., microalgae biomass can be produced only during 12h when there is daylight. Based on the environmental/ feedstock conditions of cultivating and type of microalgae, different microalgae cell densities will be left cultivation area (A maximum of 286 g/L[140] and a minimum of 0.05 g/L [141]have been reported for microalgae cell densities).

Microalgae can be grown in open ponds (OP), turbo photobioreactors (TPBR), bubble column photobioreactors (BPBR), or flat plate photobioreactors (FPBR). Then, to separate water from the microalgae slurry, sedimentation with filtration or flotation can be used. Next,

centrifugation, a filter press, or flocculation can be implemented to increase the concentration of algae biomass. A dryer is optional to dry microalgae before cell disruption takes place. It is assumed that there is no recycling stream and that all dissolved nitrogen and phosphate are used completely in the cultivation stage.

To break the microalgae cells and release lipids, pigments, and other components, four alternatives are available: bead beating, high-pressure homogenization, microwaving, and sonication. The extraction of pigments as one of the bioproducts can be done using two types of solvent extraction (organic solvent and supercritical carbon dioxide). Microalgae consist of complex lipids (for instance 56 mass percentage of total lipids of Chlorella vulgaris has 18 carbon in fatty acid chain[142]). Acetone (as organic solvent) cannot extracted this type of lipids without helping of catalyst [143] or cosolvent[144] . Thus, it is assumed that the lipids have not been extracted in pigment extraction. They are extracted from the remaining cell compositions in lipid extraction step. N-butanol, hexane, and supercritical carbon dioxide can be used to extract lipid and send it to a further process for lipid production to produce omega-3, biodiesel, and glycerol. An earlier study demonstrated how to extract omega-3 from biodiesel and glycerol [101]. Finally, the remainder of microalgae cells goes to remnant treatment to produce biogas and fertilizers. More information for each of these alternatives can be found in the previous chapter (chapter2).

The algae biorefinery consists of nine stages and 22 alternatives. Each of these alternatives is depicted as a block in the superstructure. These blocks and all possible pathways to produce six added-value products (pigment, omega-3, biodiesel, biogas, glycerol, and fertilizers) from microalgae are shown in figure 3-1.

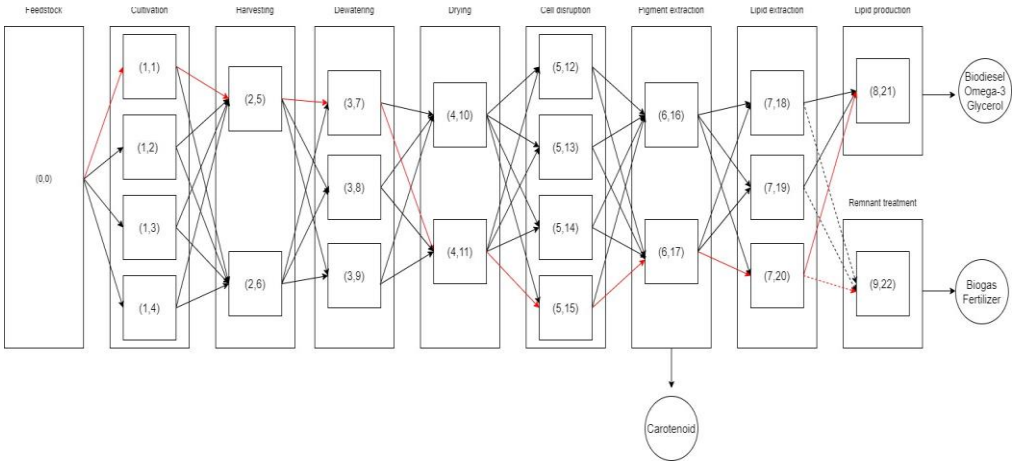


Figure 3-1: Superstructure with chosen production pathway of microalgae biorefinery.

3.2.3. Mathematical program

The mathematical program consists of four types of constraints: mass balance constraints, energy balance constraints, economic evaluation constraints, and logical constraints. These constraints and variables are defined over several indices. In the program, the index for the stages is h, and the index for alternatives is j, whereas the index of components in the program is k.

3.2.3.1 Mass balance constraints

Figure 3-2 shows the generic mass balance. There is a mixing process in the first stap of each alternative. The input flow of component k for alternative j ($m_{k.j}^{IN}$) is the summation of two streams: the upstream mass flow ($m_{k,j}^{U}$) from the previous stage and the reactant stream mass flow ($m_{k,j}^{R}$) from external resources. The upstream streams for the first four alternatives of the cultivation stage are the feedstock mass flow (F_k), whereas the other upstream mass flows are equal to the downstream mass flows from the previous stage. The reactant stream mass flow can be calculated with a concentration factor $x_{k,j}$, which is a weight fraction based on component k in the upstream flow for alternative j. Eq. (3.4) - (3.5) are the constraints related to the input stream mass flows.

$$m_{k,j}^{IN} = m_{k,j}^{U} + m_{k,j}^{R} = m_{k,j}^{U} + x_{k,j} \cdot m_{k,j}^{U} \tag{3.4}$$

$$m_{k,j}^{U} = \begin{cases} F_k & (j < 5) \\ m_{k,j-n}^{D} & (j \geq 5) \end{cases} \tag{3.5}$$

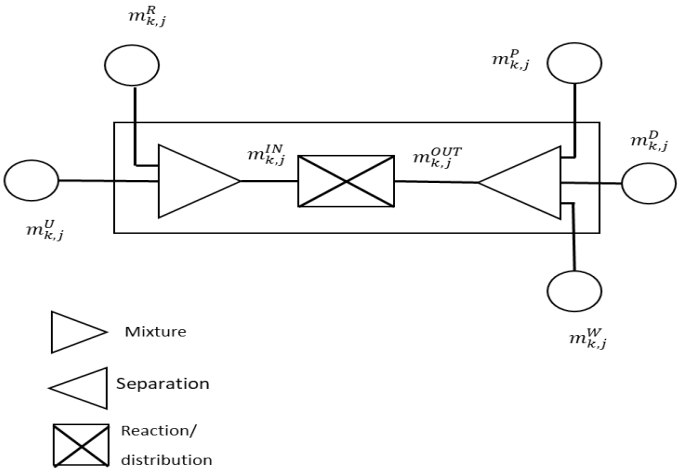


Figure 3-2: Schematic superstructure block *k* and associated mass flows.

The value of the concentration factor for all stages except the lipid production stage can be found in table C-2. Required solvents for lipid production stages are dependent on the type of microalgae due to the different types and compositions of lipid components. These values can be found in table C-3.

When a reaction takes place inside the alternative j, the output steam mass flow ($m_{k,j}^{OUT}$) can be calculated by the stoichiometric coefficient ($S_{k,j}$). Required parameters of the mass stoichiometric coefficient of reactions can be found in Table C-4 in addition to table 3-1 and Eq. (3.1) - (3.3). The conversion factor $CF_{k,j}$ based on component k is used to calculate the amount of reactant components. These values are shown in Table C-5. Instead of a reaction, distribution occurs for the remnant treatment stage, and the stoichiometric coefficient is replaced with the distribution coefficients ($D_{k,j}$) (as seen in Table C-6). If no reaction or distribution takes place inside the alternative j, the outlet flow should equal the inlet flow. The constraint is shown in Eq. (3.6).

$$m_{k,j}^{OUT} = m_{k,j}^{IN} + S_{k,j} \cdot m_{k,j}^{IN} \cdot CF_{k,j} + D_{k,j} \cdot m_{k,j}^{IN} \quad (3.6)$$

Then the output mass flow of the component (k) for alternative (j) $\left(m_{k,j}^{OUT}\right)$ can be divided into three types of streams: the downstream flow ($m_{k,j}^{D}$) going to the next stage, the waste flow $\left(m_{k,j}^{W}\right)$, and the product flow $\left(m_{k,j}^{P}\right)$. These streams can be calculated using split factors ($SF_{k,j}$) of the component (k) for alternative (j) as shown in Eq. (3.7). The values for split factors for different alternatives and flows can be found in Table C-7 (a-c).

$$m_{k,j}^{OUT} = m_{k,j}^{D} + m_{k,j}^{W} + m_{k,j}^{P} = SF_{k,j}^{D} \cdot m_{k,j}^{OUT} + SF_{k,j}^{W} \cdot m_{k,j}^{OUT} + SF_{k,j}^{P} \cdot m_{k,j}^{OUT} \quad (3.7)$$

3.2.3.2 Energy balance constraints

For the energy balances, three types of utility (U_j) are considered in this model: (U_j^E) for electricity,(U_j^H) for heating, and (U_j^C) for cooling. Assumed is that the utility consumption is linear to the total input stream mass flow (m_j^{IN}) going through the alternative (j). The energy constraint is shown in Eq. (3.8). (SUC_j) is the specific utility consumption factor for alternative (j). Data of this parameter for each type of utility can be found in Table C-8.

$$U_j = \sum_k m_{k,j}^{IN} \cdot SUC_j \quad (3.8)$$

3.2.3.3 Economic evaluation constraints

The profit margin (PM) is calculated based on the annualized investment cost (AIC), annualized operating cost (AOC), and product sales (PS) are shown in Eq. (3.9).

$$PM = PS - (AIC + AOC) \quad (3.9)$$

The AIC, can be determined using Eq. (3.10) with total installation plant cost ($TIPC$), interest rate (IR), and lifetime (LT). The $TIPC$ can be calculated based on the equipment cost (EC_j) for alternative with an engineering coefficient (K^{ENG}) and the land cost (LC_j) for cultivation stages, which is shown in Eq. (3.11). In this study, amounts of interest rate, lifetime, and engineering coefficient are 0.1, 20 (year), and 3.30, respectively [145].

$$AIC = TIPC \cdot \frac{IR \cdot (IR+1)^{LT}}{(IR+1)^{LT}-1} \tag{3.10}$$

$$TIPC = K^{ENG} \cdot \sum_j EC_j + LC_j \tag{3.11}$$

The EC_j is calculated with the equipment reference cost (EC_j^{ref}), the reference mass flow (m_j^{ref}), the cost index in 2020 (IDX_j^{2020}), the reference cost index (IDX_j^{ref}), and sizing factor (f_j) by Eq. (3.12). It is assumed that the cost index for 2020 for all the required equipment alternatives is 596.2. List and number of equipment needed for each alternative can be found in Table C-9. Required parameters of Eq. (3.12) are extracted from literature and shown in Table C-10.

$$EC_j = EC_j^{ref} \cdot \left(\frac{\sum_k m_{j,k}^{IN}}{m_j^{ref}}\right)^{f_j} \cdot \left(\frac{IDX_j^{2020}}{IDX_j^{ref}}\right) \tag{3.12}$$

The LC_j is calculated with the land price P^{Land} (3000 ($/ha)) and the productivity of algae by Eq. (3.13). (Table C-11 shows the productivities of microalgae in a different type of cultivation area).

$$LC_j = P^{Land} \cdot \frac{m_{Algae,1-4}^{OUT}}{Productivity_{Algae,1-4}} \tag{3.13}$$

The AOC consist of raw material cost (RMC), utility cost (UC), operating and maintenance cost (OMC), and waste treatment cost (WTC), which is presented in Eq. (3.14).

$$AOC = RMC + UC + OMC + WTC \tag{3.14}$$

The raw material cost and utility cost are calculated with operating hours per year H ($7920(h)$), material prices ($P^{Material}$), and utility price ($P^{Utilitiy}$) as shown in Eq. (15) and Eq. (16). (These data can be found in Tables C-12-13and 3.

$$RMC = H \cdot \sum_k (P_k^{Material} \cdot \sum_j m_{k,j}^R) \tag{3.15}$$

$$UTC = H \cdot \sum_u (P_u^{Utility} \cdot \sum_j U_j) \tag{3.16}$$

The OMC is calculated with the operating and maintenance factor (K^{OM}) which is 0.02 [125] by Eq. (3.17).

$$OMC = K^{OM} \cdot AIC \quad (3.17)$$

The WTC is linear to the waste stream mass flow with a price for waste treatment (P^{Waste}) which is 0.58 ($/t) [146]by Eq. (3.18).

$$WTC = H \cdot P^{Waste} \cdot \sum_j \sum_k m_{k,j}^{W} \quad (3.18)$$

The product sales are calculated with product prices (P^{p}) and total product mass flow as shown in Eq. (3.19). Pigment contains various components such as carotenoid, astaxanthin, chlorophyll, etc. Based on the type of microalgae, the composition and price of pigment products are varied. The average price of pigment products for Chlorella Vulgaris, Haematococcus Pluvialis, and Nannochloropsis spp. are 566, 3608.5, and 2913.5 ($/kg), respectively [12,138,147]. The price of Omega-3, biodiesel, glycerol, biogas, and fertilizer are 31.8, 1.73, 0.225,0.435, and 0.4($/kg), respectively.

$$PS = H \cdot \sum_p P^{p} \cdot \sum_j \sum_k m_{k,j}^{P} \quad (3.19)$$

3.2.3.4 Logical constraints

The selection of alternatives can be defined by a binary decision variable $y_{(h,j)}$. The logical constraint can guarantee that only one alternative can be chosen in each stage. This logical constraint is presented in Eq. (3.20).

$$\sum_j y_{(h,j)} = 1 \quad (3.20)$$

3.2.3.5 Objective function

The objective of this project is to maximize annualized profits, and the objective function is shown in Eq. (3.21).

$$maximize \quad PS - (AIC + AOC) \quad (3.21)$$

3.2.4. Block integration approach

The required time for optimizing superstructure varies, depending on the size of constraints, variables, and input data. Although more constraints and variables and fewer assumptions help improve the model's accuracy, time consumption is one obstacle to optimizing the developed superstructure. In this study, to overcome this problem, the number of variables and constraints are decreased by the block integration approach.

There are some technical data that indicate how alternatives perform in terms of mass balances and energy balances, and there is also some economic data that is needed for the cost estimation. The technical data mainly includes feedstock composition, split factors, stochiometric or distribution coefficients, conversion factors of reactant in mass balance, and

specific utility consumption in energy balances. The economic data includes reference equipment costs, sizing factors, reference mass flows, cost factors, and prices of materials and utilities. Utilizing these data in the model to illustrate each alternative's efficiency and character is necessary. However, the characteristics and performance of a block as a whole influence optimization.

A block integration is generated for different blocks that require many reactors, mixers, and separators. By merging operations with the overall reaction and separation, a complicated alternative can also be considered as one integrated process with the same properties inside. Only the inlet flow and outlet flow of this integrated process should be determined. The overall parameters for the alternative are calculated based on the parameters for each individual operation. Mass and energy balance are applied for each unit operation and block to define these data. As an example, the data calculation for the lipid production interval is presented in the appendix. Thus, in this approach, instead of using all required data for each of the unit operations of each block, a new set of data for each block is defined, presenting the performance and character of these unit operations.

The block integration approach is proposed for a model with thousands of constraints and variables (in this study, the model has approximately 50000 variables and constraints before using this approach). Block integration can not only make the alternatives in the superstructure simplified and uniformly but also eliminate the influence among the processes in an alternative.

3.2.5. Model characteristics

This is a mixed-integer non-linear programming (MINLP) model which is solved with Advanced Interactive Multidimensional Modelling (AIMMS) software version 4.82.3.29 64-bit. This MINLP model contains 6710 variables, 6161 constraints, and 22 integers variables. Two solvers (Outer Approximation Algorithm (AOA) and BARON) are used to solve the model. The AOA consists of the CONOPT 4.1 and the CPLEX 20.1 solvers for solving the non-linear and mixed-integer parts, respectively. BARON is a global optimization solver that uses a branch-and-reduce algorithm to solve MINLP.

3.3-Results and discussions

First, cost-effective production pathways are proposed for each microalga biorefinery. Second, these three microalgae biorefineries are compared from an economic and environmental aspect. Third, a sensitivity analysis is conducted to investigate the impact of uncertainty in some parameters on the profit margin of microalgae biorefinery. Finally, validation of the model is done to guarantee the accuracy of our studies.

3.3.1. Microalgae biorefinery production pathway selection

Three kinds of microalgae biorefineries (Chlorella Vulgaris biorefinery, Haematococcus Pluvialis biorefinery, Nannochloropsis spp. biorefinery) are considered in this study. More than 1100 possible production pathways to produce pigment, omega-3, biofuel, biogas, glycerol, and fertilizers from microalgae are optimized with the superstructure. The selected technologies for each microalgae species are the same, however, parameters such as productivity, the stoichiometric coefficients, the split factors, the required solvents, the composition of microalgae, the price and composition of products vary for each microalga biorefinery.

Optimized superstructure chooses the same production pathway for all these three microalgae biorefinery. The optimized superstructure uses an open pond for cultivation, sedimentation and flotation for harvesting, flocculation without any dryer for dewatering and drying, sonication for cell disruption, organic solvent (acetone) for pigment extraction, n-butanol solvent for lipid extraction, lipid production for various lipid production, and anaerobic digestion for remnant treatment. This pathway is shown in Figure 3-1.

If it is impossible to operate the proposed production pathway, other optimized process alternatives should be chosen. To find other substitutes, the superstructure is optimized to propose two alternatives for a cost-effective production pathway. These alternative pathways have only differences in choosing the technology of dewatering stages. Instead of flocculation, centrifugation and filter press are selected as the second and third best cost-effective production pathways. The investment and operating costs of these three technologies are very similar. Also, their efficiency of them is not very distinguished from each other. In conclusion, although the first cost-effective pathway has some economic benefits, the types of technology used in the dewatering stages do not considerably affect biorefinery's overall cost and profits.

The model consists of 6760 constraints and 6710 variables when using the block integration approach. The maximum time is required to optimize the superstructure are 276 seconds with a CPU Intel(R) Core(TM) i5-8265U CPU @ 1.80 GHz and 8.00 RAM. Concerning the size of the superstructure, this is the shortest time to optimize the production pathway. The block integration approach has improved the speed of optimization significantly. The number of variables and constraints without using block integration approach are 23493 and 23006, respectively. Approximately 1004 s are needed to optimize the superstructure without block integration. The model was solved with two solvers (BARON and AOA). Both solvers give the same optimized production pathways for each type of microalgae biorefinery.

3.3.2. Economic and environmental comparison of three microalgae biorefineries

10% of the mass flow of Dutch influent wastewater enters each microalga biorefinery for treatment. Approximately 200 kt of wastewater is treated to separate 9 kt Ammonia and 1.3 kt phosphate, annually. Chlorella Vulgaris biorefinery and Haematococcus Pluvialis biorefinery need 0.2 kt sulfate annually. A Sankey diagram of all the mass flow that enters and leaves the microalgae biorefinery is shown in figure 3-3.

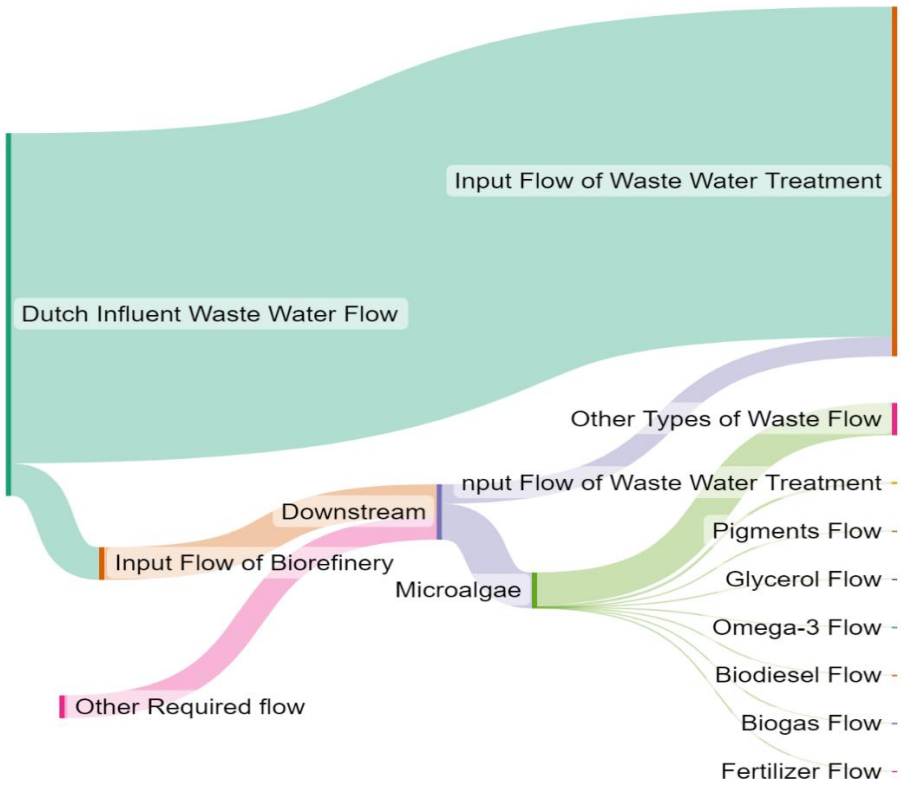


Figure 3-3: Sankey diagram of mass flow of microalgae biorefinery

In addition, these three biorefineries have an important role in capturing carbon dioxide. While Chlorella Vulgaris biorefinery can capture 82 kt carbon dioxide, Haematococcus Pluvialis biorefinery, and Nannochloropsis spp. biorefinery consume 164 kt and 281kt, respectively, to grow microalga. Thus, Nannochloropsis spp. biorefinery can capture a large amount of carbon dioxide compared to two other microalgae biorefinery.

With this feedstock characteristic, 12Kt, 24Kt, and 42 Kt Chlorella Vulgaris, Haematococcus Pluvialis, Nannochloropsis spp. can be produced, annually. Various amounts of added-value

products can be extracted from each of them. As seen in figure 3-4, the amount of pigment that can be extracted from Haematococcus Pluvialis is two times higher than Chlorella Vulgaris. Haematococcus Pluvialis biorefinery produces 0.5 kt piment. The amount of omega-3 in all these three biorefinery is very low. A maximum of 30t omega-3 can be produced from Chlorella Vulgaris. Furthermore, Nannochloropsis spp. biorefinery has an important role in the production of biodiesel, biogas, and fertilizers. While Nannochloropsis spp. biorefinery produces 6 kt biodiesel, 7kt biogas, and 5 kt fertilizer, low productions of biodiesel, biogas, and fertilizer have had in Haematococcus Pluvialis biorefinery which is more than Chlorella Vulgaris biorefinery.

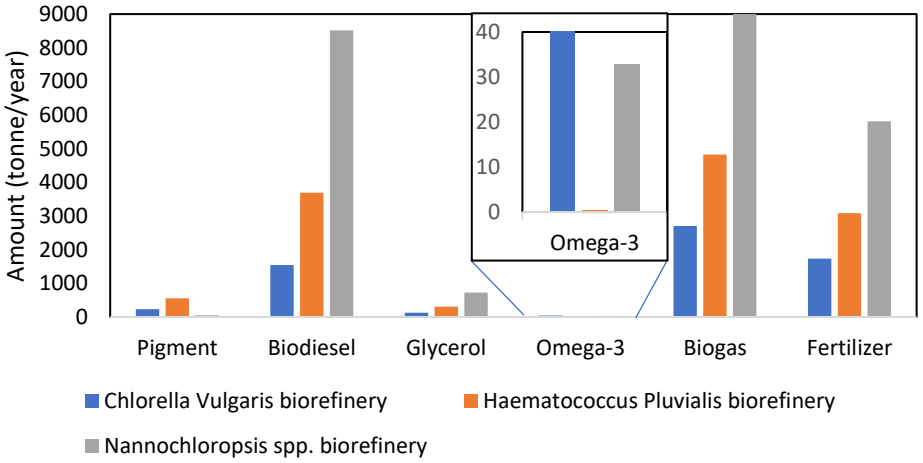


Figure 3-4: Annual amounts of added-value products produced in each of the three biorefineries

The aims of these biorefineries are to be able to meet the future demand of added-value bioproducts. It is expected that between 2020 and 2028, the global omega-3, pigment market size will reach 2.10 and 8.29 billion USD, respectively [148,149]. The Haematococcus Pluvialis biorefinery is not able to cover a high percentage of the global market for omega-3 (less than 1%), but it is provided 20% demands of the pigment market.

Selling these added-value products (pigment, biodiesel, omega-3, glycerol, biogas, fertilizer) leads to revenues of 102 M$/year, 1.6 B$/year, 128 M$/year for Chlorella Vulgaris biorefinery, Haematococcus Pluvialis biorefinery, Nannochloropsis spp. biorefinery, respectively. However, Haematococcus Pluvialis biorefinery can produce a low amount of biodiesel, glycerol, biogas, omega-3, fertilizer compared with Nannochloropsis spp. biorefinery, the high price of pigment (3608500 ($/t)) significantly increases annual production revenue. Furthermore, the price of the pigment product of Haematococcus

Pluvialis biorefinery is highest due to a large amount of astaxanthin [12]. Thus, pigment and its composition have an important role in annual production revenue and, consequently, profit margin of biorefinery. The Haematococcus Pluvialis biorefinery has the largest profit margin (62 $/kg of microalgae).

The total investment cost of these three microalgae biorefinery is shown in Figure 3-5. Depending on the type of microalgae, the investment cost is varied between 6-9 M$. The percentage of the investment cost of each stage is approximately the same for each of these microalgae biorefineries. Although the volume of water flow is reduced by ignoring recycled water of harvesting, and dewatering stages, a high percentage of investment costs is specified to separate water from microalgae substances. Furthermore, the investment cost of remnant treatment stages is significant because all other microalgae cell composition is transferred to this stage. Totally, less than 14 % of total cost specified to investment cost. Operating costs of Chlorella Vulgaris biorefinery, Haematococcus Pluvialis biorefinery, Nannochloropsis spp. biorefinery are 27, 58, and 86 M$, respectively. Utilities and raw materials cost mainly define the operating costs. 80%, 77%, and 87% of the total operating cost are allocated to utility in Chlorella Vulgaris biorefinery, Haematococcus Pluvialis biorefinery, Nannochloropsis spp. biorefinery, respectively. A total of 21, 44, and 75 M$ are needed annually to prepare required utilities for the Chlorella Vulgaris, Haematococcus Pluvialis, and Nannochloropsis spp. biorefineries.

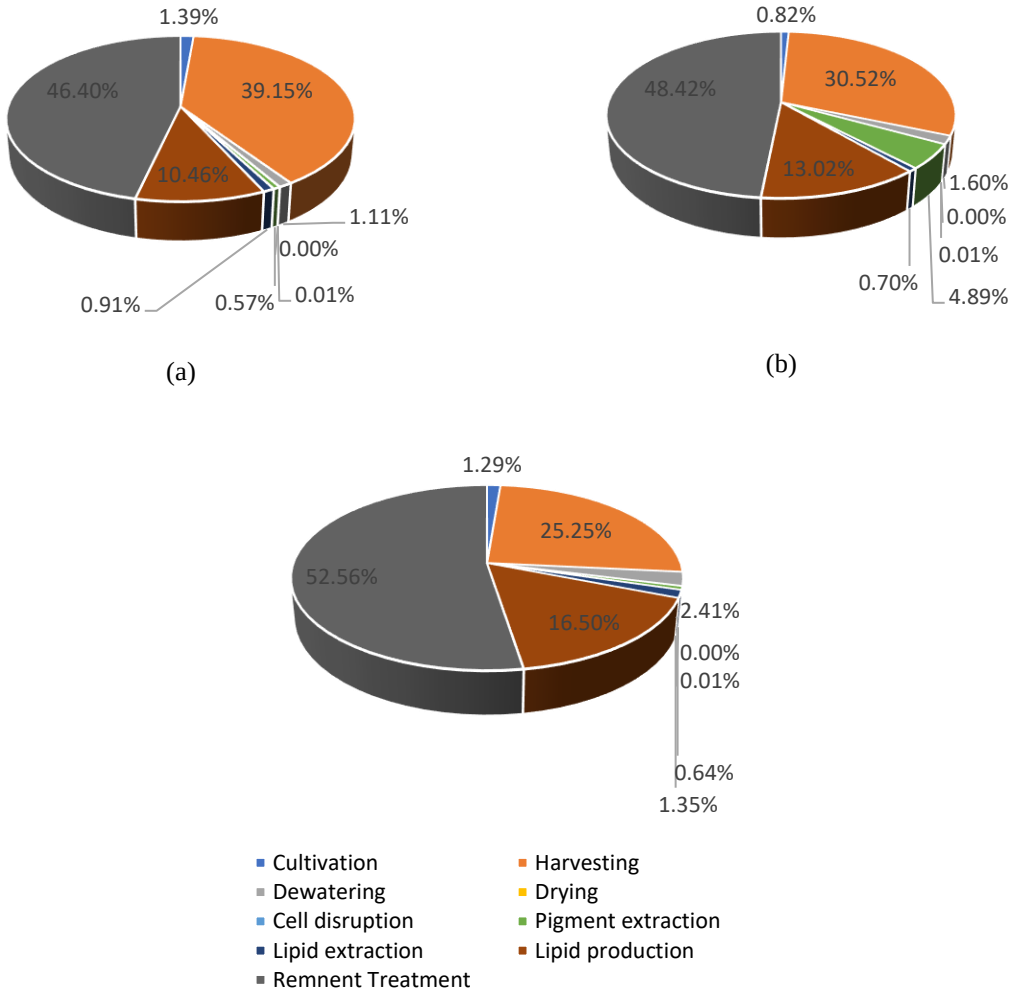


Figure 3-5: Investment costs of (a) Chlorella Vulgaris biorefinery, (b) Haematococcus Pluvialis biorefinery, (c) Nannochloropsis spp. biorefinery

3.3.3. Sensitivity analysis

A preliminary sensitivity analysis is performed to consider the impact of uncertainty in some of the parameters on the profit margin of microalgae biorefinery. Pigment is one of the

bioproducts that significantly determines the microalgae biorefinery's profit margin. The price and content of pigment are two parameters considered in this sensitivity analysis. Furthermore, ratio day/night, lipid content, and biodiesel price are other parameters of this study.

The pigment consists of various components such as astaxanthin, chlorophyll, beta-carotene, etc. The compositions of these components are varied in different microalgae. Table 3-2 shows mass percentages of these components in three microalgae. The prices of them are also not the same, as you can see in Table C-12. Based on these two uncertainties, the lower bound and upper bound of the price of pigment are defined and shown in table 3-3.

Table 3-2: Mass percentage of different types of pigments in Chlorella Vulgaris, Haematococcus Pluvialis, and Nannochloropsis spp. [12,138]

	Chlorella Vulgaris	**Haematococcus Pluvialis**	**Nannochloropsis spp.**
Chlorophyll	89%	73%	2.84%
Astaxanthin	11%	8.8%	27.05%
Beta-carotene	-	18.2%	11.82%
Vaucheriaxanthin	-	-	30.05%
Lutein/Zeaxanthin	-	-	12.81%
Canthaxanthin	-	-	13.49%
Others	-	-	1.84%

Previous studies showed that the ratio of day/night is between 0.35 – 0.63 in the Netherlands [150]. The Upper and lower bounds of other parameters are assumed ±25% of the average value. Table 3-3 shows the value of the upper and the lower bounds of all the parameters.

Table 3-3: Parameters of sensitivity analysis

	Low value	Average value	High value
Day/ night ratio	0.35	0.5	0.63
Biodiesel price ($/t)	1384	1730	2076
	Chlorella Vulgaris		
Lipid content (weight ratio)	0.09	0.12	0.15
Pigment content (weight ratio)	0.02	0.025	0.03
Pigment price ($/kg)	318	566	813
	Haematococcus Pluvialis		
Lipid content (weight ratio)	0.112	0.15	0.187
Pigment content (weight ratio)	0.020	0.032	0.038
Pigment price ($/kg)	1966	3608.5	5251
	Nannochloropsis spp.		
Lipid content (weight ratio)	0.138	0.184	0.23
Pigment content (weight ratio)	0.0012	0.0016	0.002
Pigment price ($/kg)	1793	2913.5	4032

The results of sensitivity analysis for Chlorella Vulgaris biorefinery, Haematococcus Pluvialis biorefinery, Nannochloropsis spp. biorefinery are shown in Figure 3-6. Regardless of the type of microalgae biorefinery, the profits are very sensitive to the price of pigment and pigment content of microalgae cells. The day/ night ratio is another parameter that should be considered to calculate the exact profit margin of the microalgae biorefinery. In total, the amount of microalgae produced, and pigments extracted determines annual profit margin,

and biodiesel price and lipid content have a very low impact. Biodiesel revenue has a meagre percentage of the total annual profits of microalgae biorefinery.

The annual profits of Chlorella Vulgaris biorefinery and Nannochloropsis spp. biorefinery are very sensitive to pigment price. Instead of selling pigment of Chlorella Vulgaris 566 ($/kg) to 813 ($/kg), the profit increase 63%. This scenario happens for Nannochloropsis spp. biorefinery with a 125% increase in annual profits. In these two biorefineries, the quantity of pigment products is smaller than in Haematococcus Pluvialis biorefineries. Due to that, these are very sensitive to the price of pigment.

The pigment content of each microalga is investigated in order to determine the point at which it is profitable to use microalgae biorefinery. The break-even pigment contents points of Chlorella Vulgaris biorefinery, Haematococcus Pluvialis biorefinery, and Nannochloropsis spp. biorefinery are 0.36 %, 0.027%, and 0.051%, respectively. Extraction of pigments of microalgae is in primary steps. More efforts are needed to increase the yield of pigment extraction.

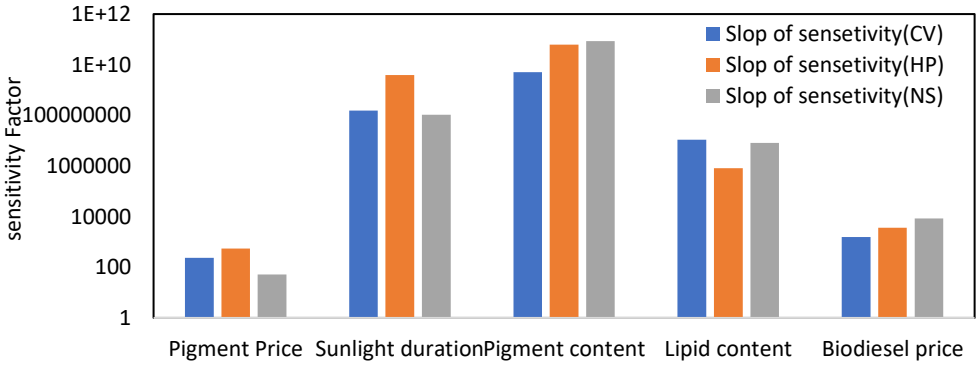


Figure 3-6: Sensitivity analysis of different parameters on profits margin of (CV) Chlorella Vulgaris biorefinery, (HP) Haematococcus Pluvialis biorefinery, (NS) Nannochloropsis spp. biorefinery

3.3.5. Validation of model

To validate the model and results, the process is simplified to produce only common products (biodiesel) from Chlorella vulgaris. Due to the availability of literature, this microalga (Chlorella Vulgaris) with 25% lipid is chosen [126,131,151]. Thus, two alternatives of pigment extraction are removed from the superstructure. Except for this change, the superstructure is the same as before. The pigment is transferred with another cell component to remnant treatment. The superstructure is optimized to estimate the price of biodiesel based

on Eq. (3.22). In this equation, by-products consist of glycerol, omega-3, fertilizers, and biogas.

$$Biodiesel\ cost\ (\$/tone) = \frac{Total\ annualized\ cost\ (\$/year) - Products\ income(\$/year)}{Biodiesel\ amount\ per\ year(tone/year)} \quad (3.22)$$

With this assumption, the cost of biodiesel is 6.18 $/l which is in good agreement with previous studies. The prices of biodiesel are reported between 2.6 ($/l)- 9.2 ($/l) range when cultivating in the open pond [152,153]. This cost can be reduced when using a photobioreactor or ignoring the production of omega-3. Optimizing the cost of biodiesel is out of the scope of this validation. Other studies have been done in this regard [154].

3.4. Conclusions

A block integration approach is developed and tested to optimize a superstructure for a microalgae biorefinery. This approach significantly decreases the number of variables and constraints of the MINLP model and increases the speed of solving it with both Baron and AOA solvers in AIMMS software. In this study, the number of variables, and constraints 4 times decreased. The superstructure is optimized for each microalga biorefinery (Chlorella Vulgaris biorefinery, Haematococcus Pluvialis biorefinery, Nannochloropsis spp. biorefinery). The type of Microalgae biorefinery does not have any effect on a chosen pathway. The identified pathway is: 1) an open pond, 2) sedimentation and flotation, 3) flocculation without a dryer, 4) sonication, 5) organic solvent pigment extraction, 6) n-butanol solvent lipid extraction, 7) lipid production, and 8) anaerobic digestion. stage

Among the various added-value products (pigment, omega-3, glycerol, biodiesel, biogas, fertilizers), pigment yields a higher profit for the microalgae biorefinery. The haematococcus pluvialis biorefinery has a more valuable pigment composition which leads to the highest profit (62 $ annual profit margin per kg of microalgae) only by using 200 Kt wastewater and 164 Kt carbon dioxide.

For such quantities of wastewater and carbon dioxide, 35 out of 350 sewage treatment plants in the Netherlands are required. Due to limitation in land availability, it is recommended to have only one Haematococcus Pluvialis biorefinery. For this biorefinery 45 M$ annual profit margin is expected.

Chapter Four

The effect of uncertainty on the economics and byproducts from microalgae in a biorefinery

Abstract

The chapter discusses how uncertainty in feedstock characteristics and availability affects economy in terms of profit margin and quantities of bioproducts in a microalgae biorefinery. Three types of uncertainties are considered: 1- the composition of influent wastewater and 2- the sunshine duration, 3- simultaneous effect of two previous uncertainty parameters (composition of wastewater and sunshine duration). To quantify the variation in influent composition, data of Dutch influent wastewater composition and sunshine duration during 1981-2019 have been collected.

The obtained probability density functions and cumulative distribution function for the influent composition and sunshine duration are subsequently used to assess the possible variations in profit margin. For this analysis the optimized production pathway obtained from the superstructure optimization in the previous chapter is used.

When composition of wastewater is varied in different years, it turns out that the probability of reaching an average profit margin (62 $/kg of microalgae) in Haematococcus Pluvialis biorefinery is more than two others (Chlorella Vulgaris and Nannochloropsis spp.) biorefineries. Its profit margin per kg of microalgae varies between 58.59 ($) and 62.94 ($), with an average of 62.86 and 0.912 standard deviation

The uncertainty in sunshine duration significantly affects the profit margin of microalgae biorefineries. Results show that annual profit margin of microalgae biorefinery are reduced by approximately 50% when using actual sunshine duration. The amount and revenue of different bioproducts in each month can be estimated. Based on this information and market demand, an appropriate type of microalgae can be chosen.

Under these two uncertainties (composition of wastewater and sunshine duration), standard deviation of profit margin of Haematococcus Pluvialis biorefinery is 3.65, which is the highest in comparison with Nannochloropsis spp. biorefineries (0.05) and Chlorella vulgaris biorefineries (0.2); due to highest profit. A range of profit margins per kg of microalgae in Chlorella Vulgaris, Haematococcus Pluvialis, and Nannochloropsis spp. biorefineries are 1.3-1.9, 27-37, and 0.27-0.42, respectively.

4.1 Introduction

Along with the increasing demands for energy, food and cosmetics, caused by population growth, waste production and pollution have skyrocketed along with agriculture intensification, industrialization, and urbanization [155]. One of modern society's problems is the need for the effective and sustainable management of urban wastewater. Aquatic ecosystems can be eutrophicated by untreated wastewater, posing a serious threat to water bodies. Therefore, it is important to apply appropriate treatment plans for removing

ammonium (NH_4^+), nitrate (NO_3^-), and phosphate (PO_4^{3-}) [155,156]. In the European Union, nitrogen pollution is estimated to cost between 70 and 320 billion euros annually [157]. Thus, nutrients and water should be recycled: if recycled, they can be considered resources rather than waste [158].

The global water consumption is 450 billion m^3 per year for industrial and domestic purposes. Domestic use accounts for 70% of this consumption, and if wastewater were used as a substrate for microalgae growth, about 23.5 billion tonnes of lipid could be produced that can provide 50% demands (global demand for crude oil is 91 million barrels per day in 2020 [159]). As well as being used as a source of energy, algal biomass could also be used in human or animal nutrition and cosmetics because of its high molecule content [160].

Efforts to improve wastewater management began in Europe with Directive 91/271/EEC, which outlines processes for wastewater treatment to prevent eutrophication [161]. This Directive states that the maximum amount of total phosphorus released into the environment should not exceed 1 mg/L for over 100,000 population equivalents and 10 mg/L for over 10,000 population equivalents [162].

The Netherlands has a combined sewer system, which collects runoff from rain and wastewater from households, businesses, and industries. The collected water is then pumped towards the different wastewater plants for treatment. After arriving at a plant, the water undergoes several treatment processes before it is returned to the surface. In the wastewater plants, the water that enters the treatment is called influent, while the water that leaves the treatment is called effluent. Influent compositions and flow rate vary throughout the year [163]. For instance, the volumes of Dutch influent wastewater in different years are shown in Figure 4-1[132].

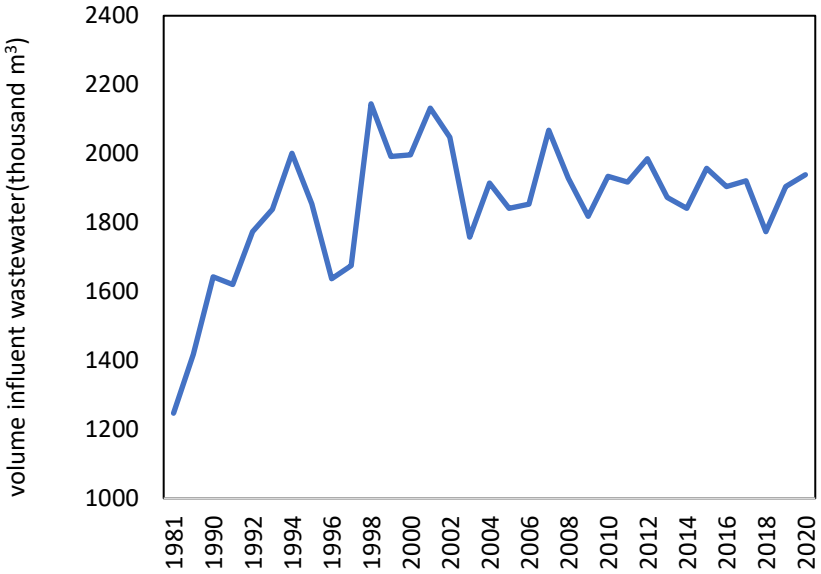


Figure 4-1: The volume of Dutch influent wastewater in different years [132]

To process the wastewater and reach the Directive, the methods of treating urban wastewater have improved throughout most of Europe over the past 30–40 years. Secondary wastewater treatment today involves the biological purification of sewage with activated sludge produced by microorganisms such as bacteria. However, this method has some disadvantages, including high energy usage (due to nitrification-denitrification), high operating costs, and sludge disposal [164–166]. A viable alternative to conventional wastewater treatment (WWT) is the cultivation of microalgae [167–169].

Waste water is one of the substrates on which microalgae grow due to their ability to grow in many different environments. Microalgae absorb nutrients (nitrogen and phosphor) needed for growth when growing on wastewater. Additionally, they can absorb heavy metals and pharmaceutical products from wastewater and capture carbon dioxide (CO_2). Apart from the fact that this can facilitate bioremediation of wastewater and protect the environment from the risk of eutrophication, it can also facilitate the removal of dangerous contaminants from wastewater and mitigate the negative effects of greenhouse gases (CO_2). Besides recycling water, this type of treatment produces microalgae biomass that can be used for food, energy, and other products at lower costs [170,171].

To grow microalgae light is necessary. There is no better light source than the sun. It is the most cost-effective, and eco-friendly light source. However, light availability from this source is one of the key factors limiting microalgae cultivation by photosynthesis. The available sunlight varies with geography and with weather conditions. In low latitude regions

(close to the equator), the sun rises and sets quite rapidly during midday compared to higher latitude regions (closer to the poles), where the sun is at a lower angle and days are longer [172].

In this chapter, the effect of uncertainty of feedstock on the amount of bioproducts and profit margin of the microalgae biorefinery is quantified. Uncertainties in the composition of Dutch influent wastewater are selected to study the role of nutrients and water on the bioeconomy of microalgae biorefinery. Furthermore, due to the variation in sunshine duration in the Netherlands, profit margin per kg of microalgae for three types of microalgae biorefineries in different seasons are estimated.

4.2 Methodology

Estimating the amount of bioproducts extracted from microalgae and the overall profitability of algae biorefineries is complicated because of the stochastic nature of the physical and chemical characteristics of the required feedstock used for cultivating microalgae.

Stochastic character refers to variable properties of feedstock (e.g., composition of wastewater) which lead to uncertain outcomes. It is important to take into consideration the uncertainties associated with feedstocks, quantities of materials needed/bioproducts, and prices when issuing stochastic process studies [173].

This chapter evaluates how the profit margin and amount of biomass and bioproducts changes for various conditions of feedstock (named uncertainty variables in this study). Uncertainty of concentration and purity of required carbon dioxide gas have not considered in this study.

Figure 4-2 shows how the standard deviation of the stochastic input variables (composition and sunlight duration) propagates up to the amount of bioproducts and the associated profit margins.

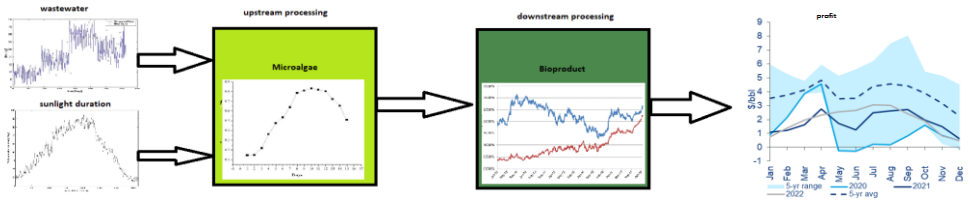


Figure 4-2: Effects of feedstock uncertainty on the probability of algae, products, and profitability.

The approach described in this work is shown in Figure 4-3. The optimized production pathway proposed in the previous chapter is used as the case for the uncertainty assessment. This pathway is 1) an open pond, 2) sedimentation and 3) flotation, 4) flocculation without

any dryer, 5) sonication, 6) organic solvent for pigment extraction, 7) n-butanol solvent for lipid extraction, 8) lipid production, and 9) anaerobic digestion. The mass and energy balance and economy of scale are defined for each of these technologies. If we want to consider:

1-uncertainty of composition of waste water: The quantities and variation of phosphate and nitrogen in influent wastewater of Netherlands for the period 1981-2019 are extracted [132]. Based on amount of the nutrients (phosphate and nitrogen) in the wastewater for each year, equations (4-1) - (4-3) can be used to calculate amount of required carbon dioxide and sulphate.

$$0.28\ H_2O + CO_2 + 0.04NH_4^+ + 0.02PO_4^{3-} + 0.002\ SO_4^{2-} \rightarrow 0.82O_2 + 0.53\ Chlorella\ Vulgaris \quad (4\text{-}1)$$

$$0.24\ H_2O + CO_2 + 0.05NH_4^+ + 0.01PO_4^{3-} + 0.02\ SO_4^{2-} \rightarrow 0.82O_2 + 0.50\ \ Haematococcus\ Pluvialis \quad (4\text{-}2)$$

$$0.28\ H_2O + CO_2 + 0.045NH_4^+ + 0.01\ SO_4^{2-} \rightarrow 0.78O_2 + 0.55\ Nannochloropsis\ spp. \quad (4\text{-}3)$$

To calculate mass balance and energy balance, the equations in sections 3.3.1 (mass balance constraints) and 3.3.2 (energy constraints) of chapter 3 are used, respectively. Equations (3-9)- (3-19) of chapter 3 are used in this study to calculate the economy of scale. The required parameters are found in tables ((C-2)- (C-13)). By using these equations, the amount of microalgae, bioproducts, profit margin and revenue can be estimated.

2-uncertainty of sunshine duration: The amount of sunshine reaching the pond is affected by weather conditions such as clouds and rain. Figure 4-4 shows sunshine duration on different days in 2018 in the Netherlands. Each day of the year, sunshine duration is extracted from literature [174]. To consider uncertainty of sunshine duration, amount of microalgae biomass and consequently amount of bioproducts and profit margin of biorefinery are calculated with actual sunshine duration data. For the calculations of the mass balance and energy balance, the equations presented in sections 3.3.1 (mass balance constraints) and 3.3.2 (energy constraints) of chapter 3 are used. To calculate economy of scale, we use equations (3-9)- (3-19) of chapter 3. Parameters required for this chapter are available in the tables ((C-2) to (C-13)). These equations (mass, energy, and economy) and equations (4-1) - (4-3) can be used to estimate the amount of microalgae, bioproducts, profit margin, and revenue. This approach is repeated for each sunshine duration set.

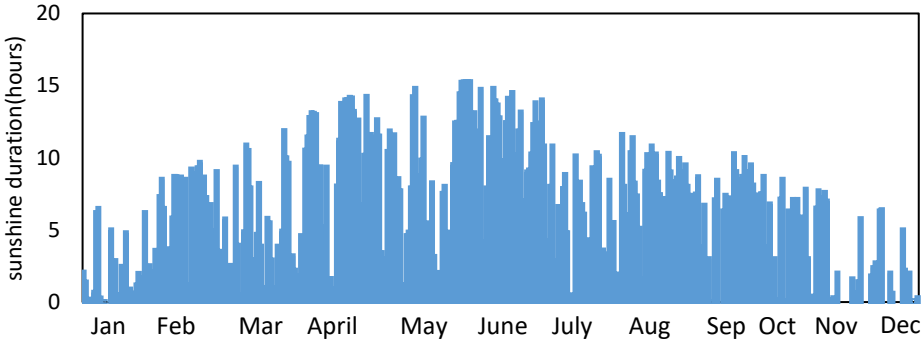


Figure 4-4: Sunshine duration in the Netherlands in 2018

Uncertainty of sunlight duration and wastewater: literature data was used to determine each year's sunshine duration and nutrient amount in influent wastewater [132,174]. Based on the amount of nutrients and equations (4-1) -(4-3), the required amount of carbon dioxide and sulphate is calculated. Mass balance and energy balance calculations are performed using equations of sections 3.3.1 and 3.3.2 of chapter 3. To calculate economy of scale, we use equations (3-9) - (3-19) of chapter 3. In this chapter, the parameters needed are found in the tables ((C-2) to (C-13)). It is possible to estimate the amount of microalgae, bioproducts, profit margin, and revenue from these equations (mass, energy, and economy, and equations (4-1) - (4-3)).

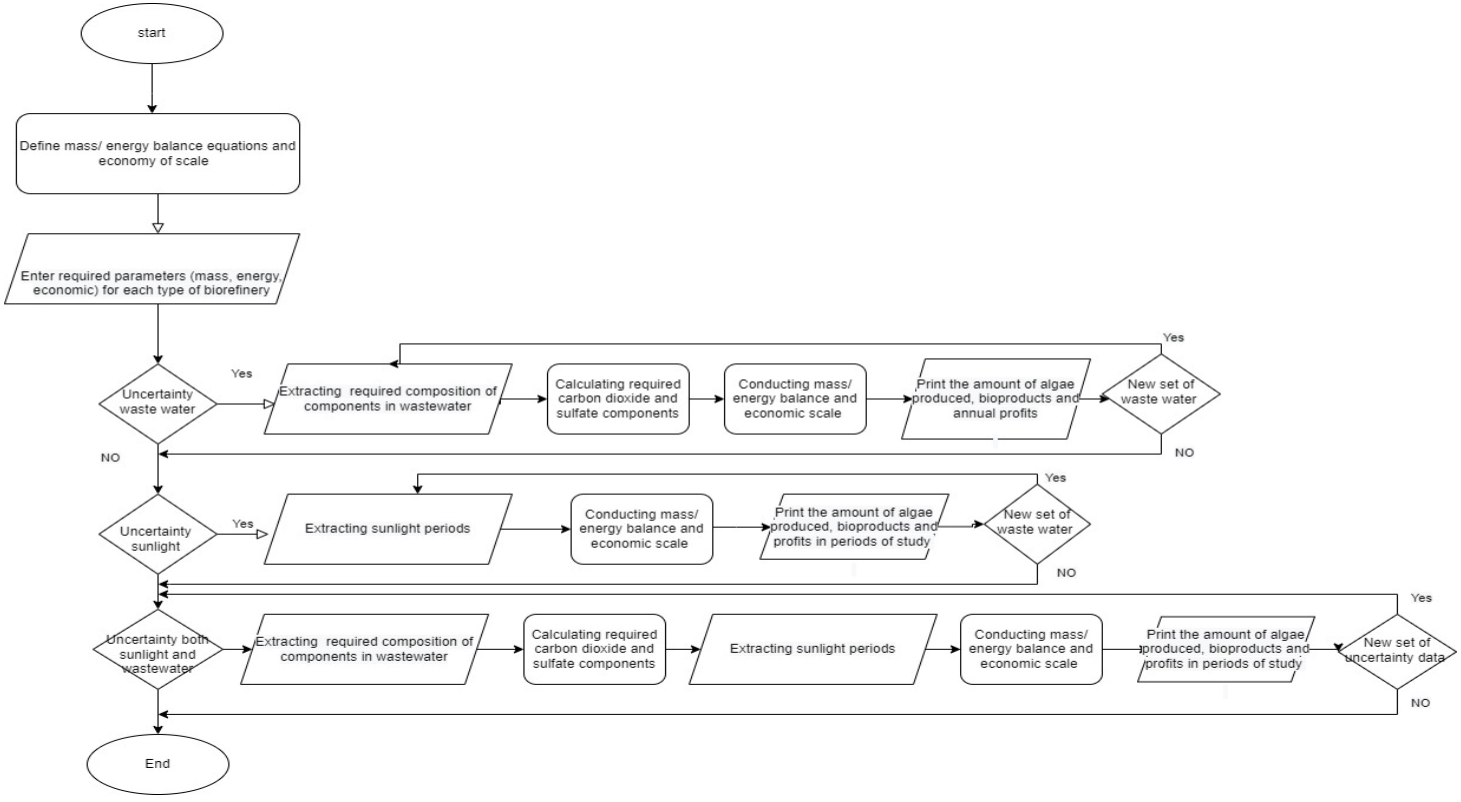


Figure 4-3: Flowchart for considering the effects of uncertainty conditions of feedstock on annual profit and the amount of bioproducts extracted from microalgae

The probability density function (PDF) can be used to determine the likelihood that a value of a random variable will occur within a specified range of values. For instance, in this study, the probability density function can estimate an expected outcome of the profit for a microalgae biorefinery.

In the cumulative distribution function (CDF), the cumulative probability is calculated for a given value of the random variable (say x). The CDF measures the probability that a random variable will take a value less than or equal to x, while PDF measures how likely it is to take a value exactly equal to x. Thus, calculating the cumulative probability of a profit based on the probability density function. The cumulative distribution function is between 0 and 1. Thus, calculating the cumulative distribution is a useful way to evaluate the probability for certain outcomes [175].

Calculating the average value and standard deviation are the first step in setting up the probability distributions. The results (e.g., amount of profit margin, revenue for each uncertainty variable) of previous part (uncertainty of composition of wastewater, uncertainty of sunshine duration and uncertainty of sunlight duration and wastewater) are used to calculate average value. An average is obtained by adding up the result of all the trials and dividing them by the number of trials. The standard deviation measures how dispersed a dataset is in relation to its average and is calculated as the square root of the variance. A normal distribution is a symmetrical plot of data around its mean value, where the standard deviation defines the width of the curve. In this study, the probability density functions, cumulative distribution function, average and standard deviation are calculated in MATLAB using default functions (pdf, cdf, mean, and std, respectively).

4.3 Results and discussion

Amounts of water, nitrate, and phosphate as required components for growing microalgae in different years are shown in Figure 4-5 [132].

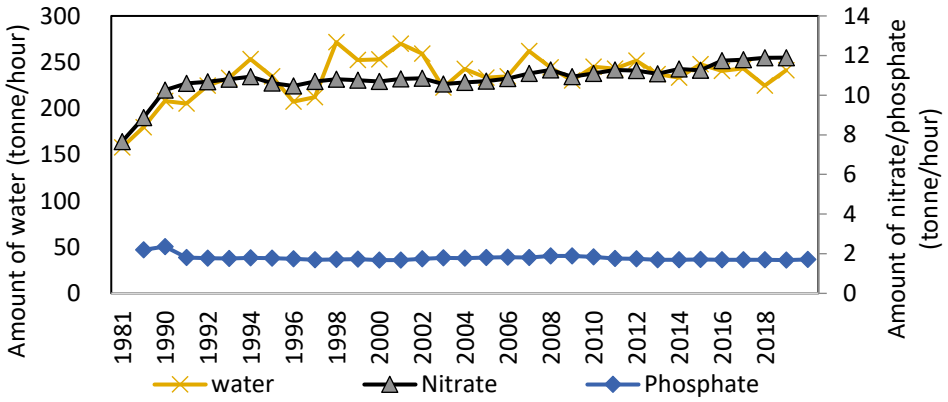


Figure 4-5:Amount of various components of wastewater in different years[132]

The variation in the amount of water is greater than the variation in the amount of phosphate and nitrate. Between 1981 and 1990, nitrate levels increased dramatically. After this period, the rate of the increasing amount of this component is fixed. Phosphate, however, decreased in 1992 compared to previous years.

The amount of three microalgae that can be produced each year is shown in Figure 4-6. Nannochloropsis spp. can be produced more than two others due to high productivity and stochiometric coefficient. Furthermore, the increasing microalgae production is related to increased nutrient levels.

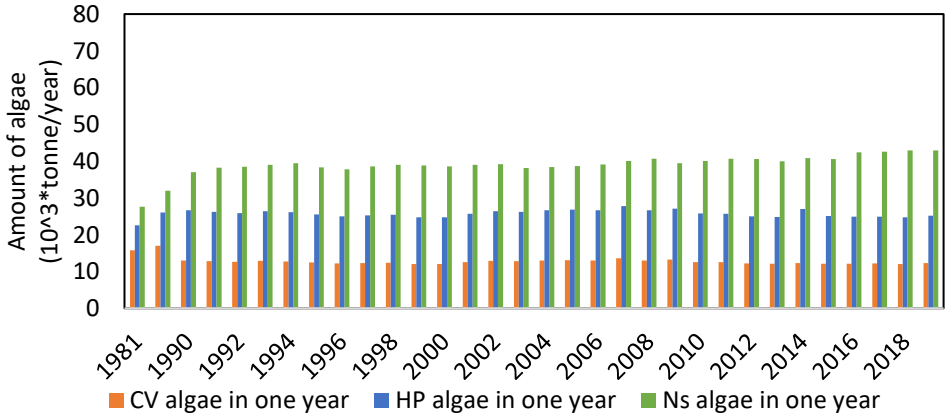
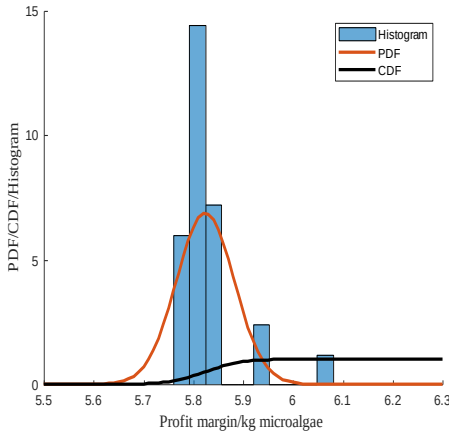


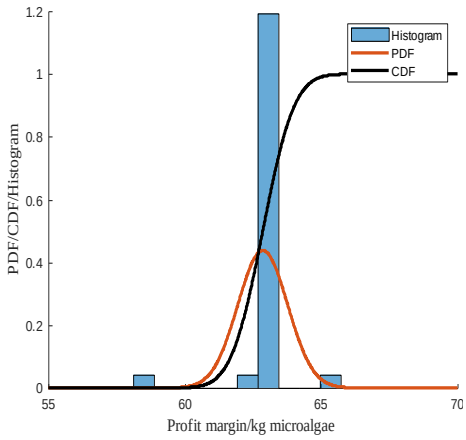
Figure 4-6: Three types of microalgae (CV: Chlorella Vulgaris, HP: Haematococcus Pluvialis, NS: Nannochloropsis spp.) production in different years

4.3.1 Economic analysis of biorefinery under uncertainty of composition of wastewater

Based on Monte Carlo simulations the probability densities of profit margin of these three microalgae biorefineries under uncertainty of wastewater composition are obtained (as seen in Figure 4-7). Although probability density is more than one, but cumulative distribution is less than one that sure accuracy of the results. The histograms bar of profits of these biorefineries show that varies of profit margin in Haematococcus Pluvialis biorefinery is less than others. The standard deviation for the profit margin per kg of Haematococcus Pluvialis is low and is about 0.912. Its profit margin per kg of microalgae varies between 58.59 ($) and 62.94 ($), with an average of 62.86 ($). Its cumulative curve is shown that with a high percentage of assurance (80%), the profit margin of the Haematococcus Pluvialis biorefinery are between 62.55 ($/kg microalgae) and 62.98 ($/ kg microalgae). The standard deviations and average profit margin for the other two microalgae biorefineries can be found in Table 4-1. In comparison with Nannochloropsis spp. biorefinery, standard deviation of profit margin of Chlorella Vulgaris biorefinery is low. Thus, with more chance can get the average value. In total, each of the three microalgae biorefineries has a reasonable probability of profit margin despite the uncertainty of feedstock.



(a)



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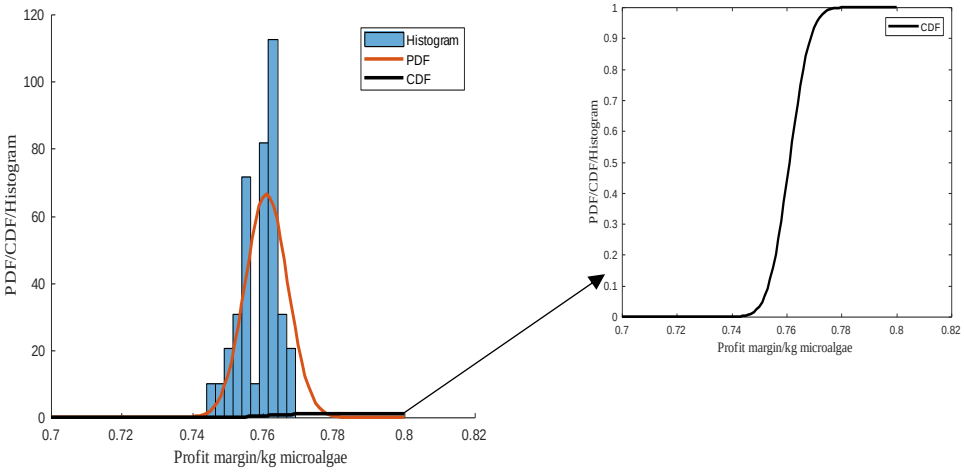


(c)

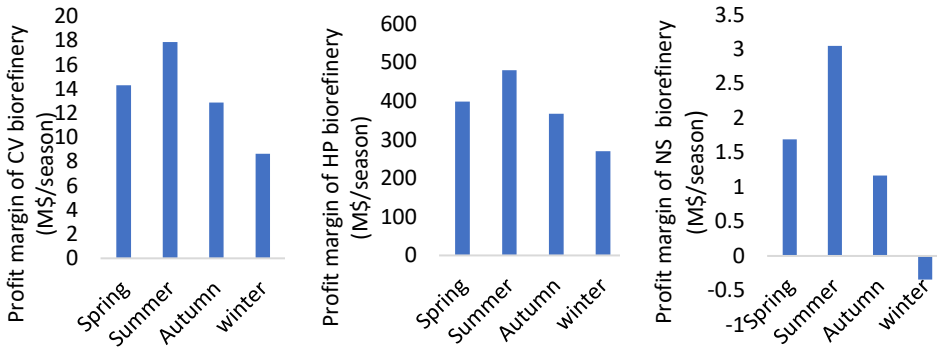
Figure 4-7: Probability density (PDF), histogram, and cumulative distribution (CDF) of profit margin of (a)Chlorella Vulgaris (b) Haematococcus Pluvialis (c) Nannochloropsis spp. biorefineries

Table 4-1: Standard deviations and the average profits of microalgae (CV: Chlorella Vulgaris, HP: Haematococcus Pluvialis, NS: Nannochloropsis spp.) biorefinery.

	Average profit margin ($)/kg microalgae	**Standard deviations**
CV biorefinery	5.82	0.058
HP biorefinery	62.86	0.912
NS biorefinery	0.76	0.761

4.3.2 Economic analysis of biorefinery under uncertainty of sunshine duration

Season profit margins are varied due to sunshine duration. Figure 4-8 shows profit margin in Chlorella Vulgaris biorefinery (a), Haematococcus Pluvialis biorefinery (b), and Nannochloropsis spp. biorefinery (c) in different seasons. In all three microalgae biorefineries, the summer season with the highest sunshine duration leads to high profit margin as result from elevated levels of photosynthesis. Furthermore, Nannochloropsis spp. biorefinery has negative profit in winter. Therefore, it would be better to temporarily close this biorefinery in this season. In conclusion, the profit margin for different seasons may be utilized for further decisions (for instance temporary close or change type of microalgae biorefinery).



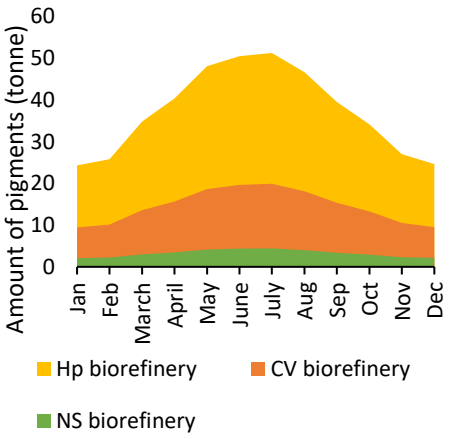
(a) (b) (c)

Figure 4-8: Amounts of profit margin of three microalgae (CV: Chlorella Vulgaris (a), HP: Haematococcus Pluvialis (b), NS: Nannochloropsis spp.(c)) biorefinery in each season

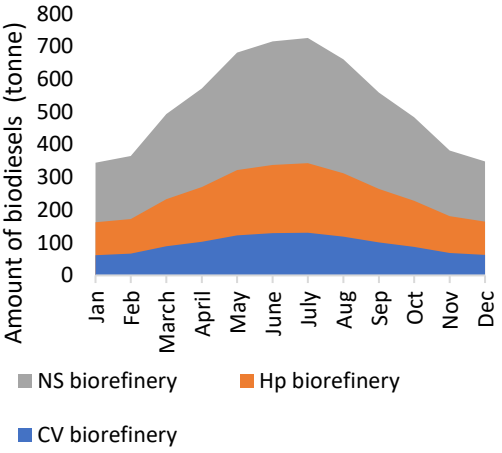
The amount of bioproducts extracted each month depends on the composition of microalgae and their productivity. Figure 4-9 shows the amount of different bioproducts extracted each month. Decision makers and planners can use this information to change the type of microalgae biorefinery based on market demand.

In July, Haematococcus Pluvialis biorefinery produced approximately twelve times more pigments than Nannochloropsis spp. biorefinery. However, the results of this comparison differ from those of other types of bioproducts. Biodiesel produced by Nannochloropsis spp. biorefinery in July is approximately two times higher than biodiesel produced by Haematococcus Pluvialis.

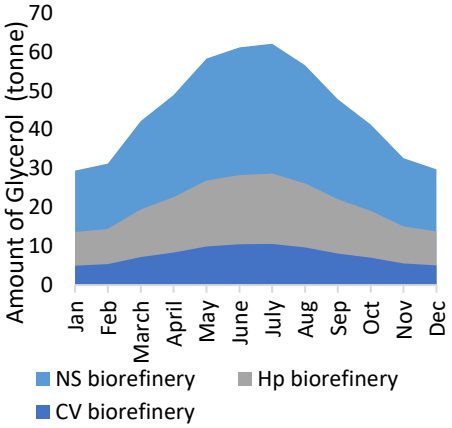
Haematococcus Pluvialis biorefinery can produce the maximum amount of omega-3 in July, which is 0.026 tonnes. As compared to two other biorefineries, this value is extremely low. The Chlorella Vulgaris biorefinery can produce 3.38 tonnes in the same period, while Nannochloropsis spp. can produce 2.81 tonnes. In other words, if the biorefinery is aimed at producing omega-3 fatty acids, then Haematococcus Pluvialis is not a good choice.


Amount of pigments (tonne)
60
50
40
30
20
10
0
Jan
Feb
March
April
May
June
July
Aug
Sep
Oct
Nov
Dec
Hp biorefinery
CV biorefinery
NS biorefinery

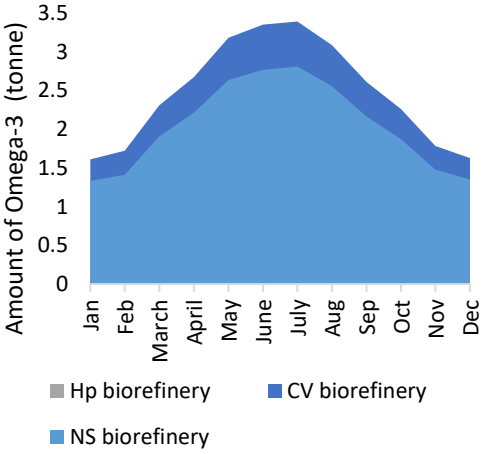
(a)


Amount of biodiesels (tonne)
800
700
600
500
400
300
200
100
0
Jan
Feb
March
April
May
June
July
Aug
Sep
Oct
Nov
Dec
NS biorefinery
Hp biorefinery
CV biorefinery

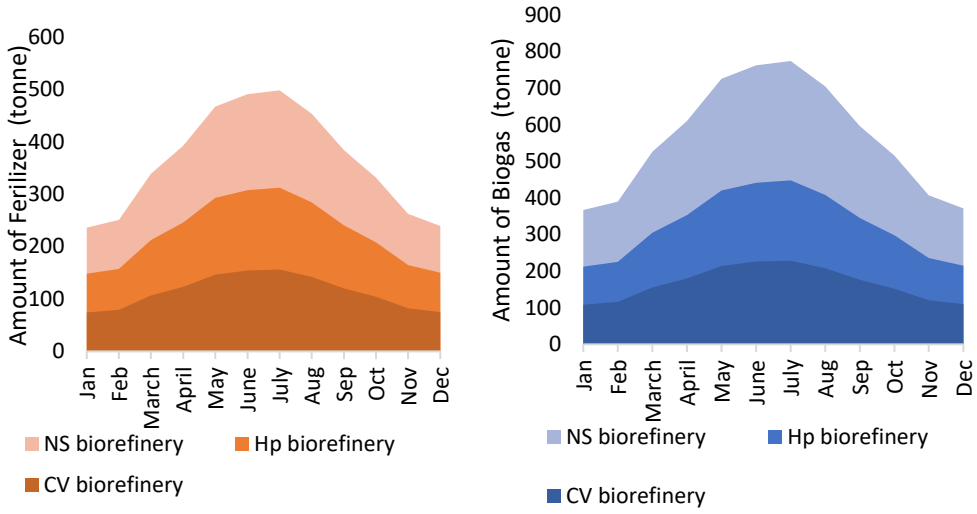
(b)


Amount of Glycerol (tonne)
70
60
50
40
30
20
10
0
Jan
Feb
March
April
May
June
July
Aug
Sep
Oct
Nov
Dec
NS biorefinery
Hp biorefinery
CV biorefinery

(c)


Amount of Omega-3 (tonne)
3.5
3
2.5
2
1.5
1
0.5
0
Jan
Feb
March
April
May
June
July
Aug
Sep
Oct
Nov
Dec
Hp biorefinery
CV biorefinery
NS biorefinery

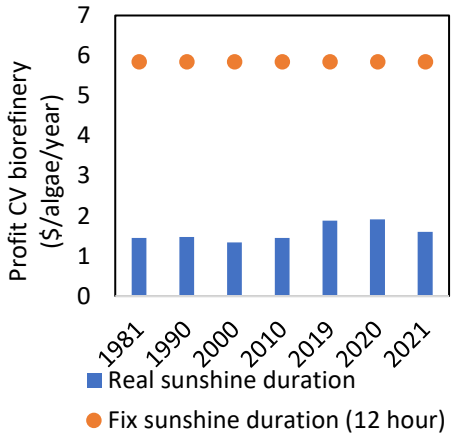
(d)



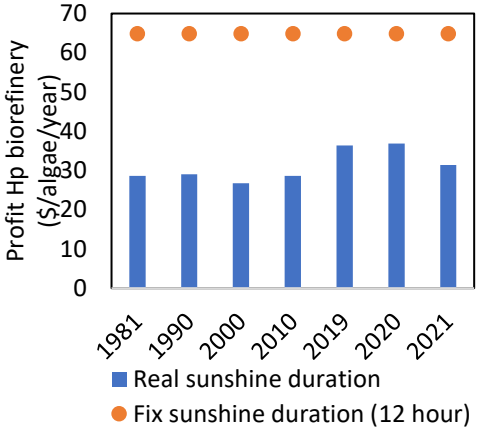
(e) (f)

Figure 4-9: Amounts of different bioproducts (pigments, biodiesel, biogas, glycerol, fertilizer, omega-3) of three microalgae (CV: Chlorella Vulgaris (a), HP: Haematococcus Pluvialis (b), NS: Nannochloropsis spp.(c)) biorefinery in each month

Photosynthesis is highly dependent on sunshine durations, which, in turn, affects microalgae productivity. With decreasing duration of sunshine, microalgae biorefinery profits decrease significantly. Figure 4-10 considers the uncertainty of sunshine duration on the annual profit margin of microalgae biorefinery. The annual profit margin per kg of microalgae of Chlorella Vulgaris, Haematococcus Pluvialis, and Nannochloropsis spp. biorefineries are estimated approximately to be 1, 32, and 0.4 dollars, respectively, according to actual data of sunshine duration. It has been estimated that approximately 50% of errors are caused by considering fixed values for sunshine periods. The total profit margin of microalgae biorefineries is therefore highly dependent on this parameter.



(a)



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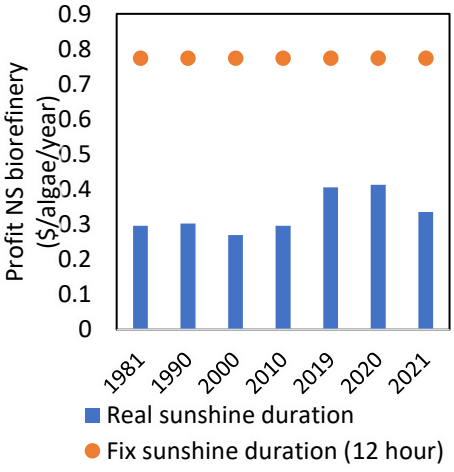


(c)

Figure 4-10: Profit margin of three microalgae (CV: Chlorella Vulgaris (a), HP: Haematococcus Pluvialis (b), NS: Nannochloropsis spp.(c)) biorefinery by considering real/fixed sunshine duration.

As a final step, the annual profit margin per kg of microalgae is calculated by considering both of these uncertainties (wastewater composition and sunshine duration) at the same time. Figure 4-11 shows the CDF of profit margin of Chlorella Vulgaris (a), Haematococcus Pluvialis (b), Nannochloropsis spp.(c) biorefineries under consideration of these two

uncertainties. The minimum profit margins per kg of microalgae Chlorella Vulgaris, Haematococcus Pluvialis, Nannochloropsis spp. are 1.3, 27, and 0.27 $, while the maximum profit margins per kg of microalgae are 1.9, 37, 0.42, respectively. The Haematococcus Pluvialis biorefinery has a relatively high standard deviation, which approximates 3.65 due to high number of profits (standard deviation of Nannochloropsis spp. biorefinery and Chlorella Vulgaris biorefinery are 0.05 and 0.2, respectively). The average annual profit margin under these two uncertainties of Chlorella Vulgaris, Haematococcus Pluvialis, Nannochloropsis spp. biorefineries are 1.6, 31, and 0.33 $ per kg of microalgae.

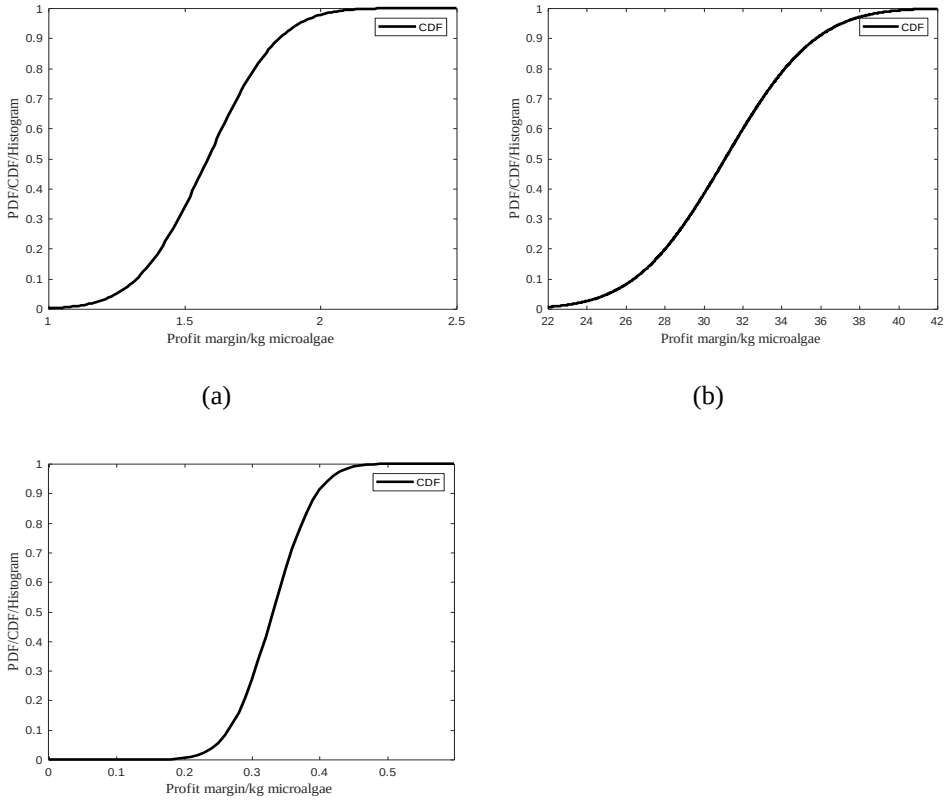


Figure 4-12: Profit margin of three microalgae (CV: Chlorella Vulgaris (a), HP: Haematococcus Pluvialis (b), NS: Nannochloropsis spp.(c)) biorefinery by considering uncertainty conditions of wastewater composition and sunshine duration

Conclusion

The effect of uncertainty of feedstock quality and availability on annual profit margin and productivity of three types of microalgae biorefineries is studied using Monte Carlo simulation. Two parameters that were investigated in this study are wastewater composition and sunshine duration. Three case study on these two uncertainties are defined :1- only investigating uncertainty of composition wastewater, 2: only considering uncertainty of sunshine duration, 3: uncertainty of composition of wastewater and sunshine duration in each year, simultaneously. The results of the study of uncertainty of composition of wastewater show that Haematococcus Pluvialis biorefinery has the highest profit margin per kg of microalgae (62.86 ($)) with an 80% probability of earning profit margin between 62.55 ($) and 62.98 ($), according to a cumulative distribution function curve. A second uncertain parameter is the sunshine duration that varies across seasons, which affects the estimation of profit margin and amount of bioproducts. Annual profit margin of microalgae biorefinery are reduced by approximately 50% when using actual sunlight duration. Finally, the profit margin of Chlorella Vulgaris, Haematococcus Pluvialis, Nannochloropsis spp. under uncertainty of composition of wastewater and sunshine duration are varied between (1.3-1.9, 27-37, and 0.27-0.42 ($/kg microalgae/year), respectively.

Chapter Five

Life cycle assessment of a microalgae biorefinery

Abstract

Microalgae biorefineries are recognized as one of the most valuable biomasses biorefinery due to source for various added-value products (such as pigment). A sustainability study of these biorefineries is an essential step toward improving microalgae production processes, coordinating strategic planning, and forming public policy. One of the tools for analyzing the sustainability of a microalgae biorefinery is a Life Cycle Assessment (LCA).

This chapter presents the LCA for three microalgae biorefineries (Cholera Vulgarise biorefinery, Haematococcus Pluvialis biorefinery, Nannochloropsis spp. biorefinery) when producing different added-value products: pigments, biodiesel, glycerol, omega-3, fertilizers, and biogas. For each of these biorefineries, different environmental impacts are considered, e.g., ozone layer depletion, global warming, eutrophication, ecotoxicity, acidification, and human toxicity. The Nannochloropsis spp. biorefinery has the highest environmental impact compared to other biorefineries. However, the functional unit of these biorefineries is the same (one cubic meter Dutch influent wastewater). The biorefinery that cultivates and processes Nannochloropsis spp. produces more microalgae biomass. These massive quantities of biomass require additional solvents for reactions and utilities, which significantly impacts the environmental categories. This biorefinery emits 903.146 kg CO_2 eq. of greenhouse gases using one cubic meter Dutch influent wastewater.

The environmental impacts of the individual processing intervals (cultivation, harvesting, dewatering, cell disruption, pigment extraction, lipid extraction, lipid production, remnant treatment) in the microalgae biorefinery are also analysed. It turns out that the remnant treatment step has the greatest impact on global warming, human toxicity, ecotoxicity, and acidification. Cultivation and harvesting have the most eutrophication impact.

A comparative environmental impact analysis between producing 1 kg β-carotene (as example of pigment) from carrot and these microalgae has been conducted. LCA results show that microalgae biorefinery have greater environmental impact in comparison with carrot refinery. Also, the cost of producing this pigment from carrots is lower than that of microalgae.

5.1 Introduction

In 1992, the United Nations Organization created the United Nations Framework Convention on Climate Change (UNFCC) because of the adverse impacts of global warming caused by carbon dioxide emissions on agriculture, forestry, ecosystems, and water resources. Achieving a level of greenhouse gas concentrations in the atmosphere that would prevent dangerous anthropogenic interference with the climate system is the goal of this association [176–178].

There is no doubt that fossil fuels are one of the most important contributors to carbon emissions. As an alternative energy source for transportation, various feedstocks have been examined for biofuel production, including cotton, soybean, sunflower, rapeseed, palm oil, and algae [19,179].

In recent studies, algae are considered a feedstock for a new generation of biofuels [180]. A significant advantage of algae as compared to other feedstocks is their fast growth, high lipid content, low water and land use, and ability to treat wastewater and capture carbon dioxide during the process [181]. Compared to other types of biomasses, algae provide a higher yield, year-round cultivation, the ability to use brackish water, and the ability to use lower-quality land. Still, there is a need to conduct a more critical assessment of this biomass scalability and its environmental impacts related to its application.

In addition, algae have been found to be useful for a wide range of additional applications, for example in pharmaceuticals, food, animal feed, and nutraceuticals. In Chapter 2, the different added-value components that can be extracted from the microalgae and their application have been discussed in more detail.

The cultivation of algae requires a large quantities of carbon dioxide and nutrients. For each kilogram of algae biomass produced, approximately 1.8 kilograms of CO_2 (depending on the species of algae) is captured without any extra cost for carbon storage. Fresh water, seawater, and wastewater contain the required nutrients and are all suitable for algae growth. [182–184].

As there can be various bioproducts obtained from microalgae, they are all need to be assessed for their environmental impact. In this chapter, an LCA methodology that evaluates three types of microalgae biorefineries for their impacts on ozone layer depletion, global warming, eutrophication, ecotoxicity, acidification, and human toxicity is employed. It is crucial to assess the life cycle of the overall process of bioproduct production in each microalga biorefinery from cradle to gate. In addition, the role of the individual microalgae processing steps in different environmental impact categories is assessed. A comparison of the environmental impact analysis between the current process and the microalgae process of producing β-carotene has been done.

5.2 Materials and methods

The LCA is a scientific tool that has been developed to allow researchers to assess environmental impacts of processes [181]. As a result of the LCA, the processes that adversely impact the environment can be identified and optimized to reduce their adverse impact on the environment [185,186].

LCA has four stages (as shown in figure 5-1): 1) goal and scope definition: Goal defines the purpose of the study and how its results will be used, while the scope specifies the parameters of the study (such as functional unit, system boundaries, and assumptions). 2) life cycle inventory (LCI): it is accounting every steam involved to the system. 3) life cycle impact assessment (LCIA): it is a systematic method of analysing how products and services will impact the environment during their entire life cycle. and 4) interpretation: outcomes are checked and evaluated. 'OpenLCA'is used as open-source software for this LCA study. The AgriBalyse, open-source database is utilized for the life cycle inventory.

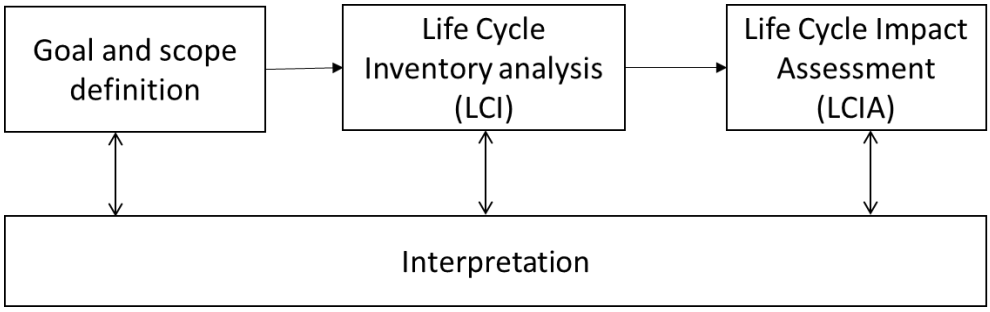


Figure 5-1: different stage of LCA study[187]

5.2.1 Goals and Scope

In Chapter 3, the superstructure optimization was used to identify the most promising production pathway from microalgae to final products in terms of profitability. This pathway produces added-value products besides biodiesel and biofuel from three different microalgae. This route will be now subjected to an LCA.

The goal is to compare the environmental impact of the three types of microalgae (Cholera Vulgarise biorefinery, Haematococcus Pluvialis biorefinery, Nannochloropsis spp. biorefinery) for each environmental impact category. The environmental impacts of the processes are further analysed by computing their contributions to each of the different processing steps required in the refinery. For an accurate comparison, production pathways and feedstock must be similar for each of these biorefineries. Finally, producing pigment from microalgae and current feedstock have been compared economically and environmentally.

5.2.2 Functional Unit (FU)

A functional unit describes the performance delivered by a product or system in its end-use application. Two functional units are chosen in this study:

1- Using the raw material as functional unit: Wastewater is required as nutrient source for the algae cultivation. To compare three types of microalgae biorefineries; one cubic meter of Dutch influent wastewater is selected as a functional unit. For this comparison, it is important to fix all parameters (such as composition and quantities of feedstock) except the species of algae.

2- Using the product as functional unit for comparing different steps of microalgae biorefineries and several types of biomasses (microalgae and carrot): 1 kg of pigment has been chosen as a functional unit. Since pigments are highly profitable, microalgae biorefineries should produce pigments as their primary goal.

5.2.3 Environmental impact categories

The following environmental impacts are considered in this study: ozone layer depletion, global warming, eutrophication, ecotoxicity, acidification, and human toxicity (cancer and non-cancer related). Each of these impact categories are explained below.

Ozon layer depletion: the emissions that are responsible for destroying the stratospheric ozone layer. The unit measurement of this impact category is the kilogram of Trichlorofluoromethane equivalent (kg CFC-11 eq).

Global warming: the potential for global warming that may occur due to greenhouse gas emissions into the atmosphere. The unit measurement of this impact category is the kilogram of carbon dioxide equivalent (kg CO_2 eq).

Eutrophication (freshwater/ marine/ terrestrial): nutrient enrichment of freshwater/ marine/ terrestrial ecosystems via nitrogen or phosphor emissions. The unit measurement of eutrophication-freshwater, eutrophication-marine, and eutrophication-terrestrial is the kilogram of phosphate equivalent (kg PO_4-eq) and mole of nitrate equivalent (mol N-eq), respectively.

Ecotoxicity (freshwater): toxic substances released into the environment on freshwater organisms. The unit measurement of this impact category is kg 1,4-dichlorobenzene equivalent (kg 1,4-DCB)

Acidification: nitrogen oxides and sulfur oxides release can cause acidification of soils and water. The unit measurement of this impact category is the kilogram of sulfur oxides equivalent (kg SO_2 eq).

Human toxicity (cancer and non-cancer related): the health condition of humans is affected by emissions of toxic substances into the atmosphere. There are two kinds of toxic substances: non-cancerous and cancerous. The unit measurement of this impact category is the comparative toxic unit for humans (kg 1,4-DCB) [188,189].

5.2.4 System boundary

A system boundary delineates which processes in the product life cycle are included in a life cycle assessment. In this research, the concept of life cycle assessment is applied to the use of microalgae in biorefineries from cradle to gate, I.e., (From raw materials to factory gates). As presented in figure 5-2, the system boundaries encompass (1) cultivation in the open pond, (2) dewatering in the sedimentation and flotation (3), harvesting in the flocculation, (4) not necessary to use the dryer, (5) cell disruption in the sonication, (6) pigment extraction with an organic solvent, (7) lipid extraction with n-butanol extraction, (8) lipid production, finally (9) remnant treatment. Materials that enter the system boundaries are shown in blue lines. The required utilities are shown in purple lines. The red lines show the materials existing the system boundary. More information about each of these streams and quantity of them can be found in table C-14-16.

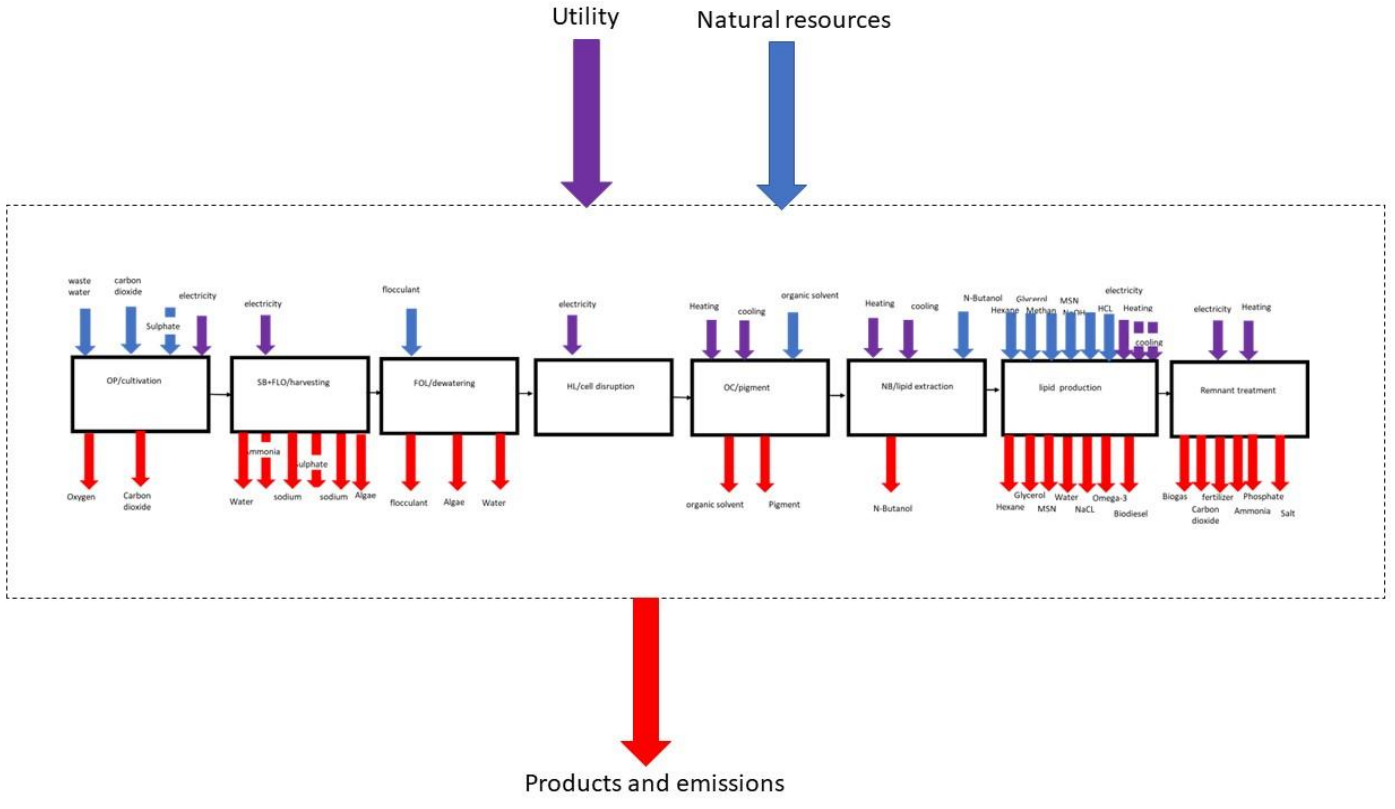


Figure 5-2: System boundary (dotted lines) with input/output flows

5.2.5 Life Cycle Inventory (LCI)

During the LCI phase, data and estimates for the outputs and inputs are collected following the system under study. The inputs and outputs of a process include energy, raw materials, and other physical inputs, products, co-products, and waste emissions to air, water, and soil, as well as other environmental effects.

In this study, the input and output flows are estimated based on mass and energy calculations of chapter 3. Thus, the amounts of materials and energy consumed or produced in each case study when using one cubic meter of Dutch influent wastewater (functional unit) or producing one kilogram of pigment (functional unit) are calculated. These data are available in table C-14-16. Then, these input/output flows need to be assessed for upstream (direct/indirect) environmental impacts. For this purpose, Agribalyse is used as a database. The AgriBalyse is a French LCI database focusing on agriculture and food. Upstream impacts of flows are stored in the LCI database and could be directly added to the inventory once the inputs/ outputs of a studied system are determined.

5.2.6 Assumptions

The transportation of flue gases through pipelines from the power plant to the open raceway ponds is not included. The same occurs for transporting primary algae products (e.g., algae oil) to other industries (e.g., transesterification facility) and transporting final products (e.g., biodiesel) from storage to the market. Furthermore, the construction of the chemical plant and the open raceway ponds are not included.

In the LCI study, to use open source database, some assumptions are made:

1- Although these three microalgae biorefineries produce/use different amounts of bioproducts/components, the microalgae that exist in various steps of biorefineries are assumed to be the same. Laminaria is the only microalga defined in the database;

2- Electricity with different production sources is used as a utility;

3- It is assumed that all the pigments produced are of the red types available in the database;

4- Omega-3 is not defined in this database. The fatty acid methyl ester is considered as omega-3;

5- Fatty acids C9-C13 are the main components of biodiesel;

6- All existing water is considered wastewater.

5.2.7 Life Cycle Impact Assessment (LCIA)

As part of the LCA study, there is an LCIA used to compile and document all emissions and resources consumed in a product throughout its lifetime. An impact assessment follows, which considers aspects such as human health, the environment, and the use of natural resources.

In Bussa et al., (2018) is mentioned that the ReCiPe impact assessment method is most recommended for microalgal biorefinery systems since it covers a multitude of environmental factors, as it covers more than just energy and greenhouse gas emissions [190]. Therefore, it was decided that the ReCiPe impact assessment method would be used in this study as a life cycle assessment method.

Many impact assessment methods (such as Eco-Indicator 99, LIME) are available to practitioners of life cycle assessment, but ReCiPe is the most recent and most updated. There are two levels to evaluate the environmental effects in this model: midpoint and endpoint. Indicators of midpoints describe how products contribute to specific environmental impacts. There are 18 midpoint category indicators. In endpoint terms, these are the final environmental effects caused by various environmental influences at the midpoint levels, such as the destruction of biodiversity, the loss of human health, and the exhaustion of raw materials. The model aggregates the midpoints into three different endpoint categories based on the number of concerns raised at the midpoint level (see Figure 5-3) [191–195]. In this study, The ReCipe midpoint method has been utilized via openLCA software to estimate the environmental impacts of microalgae biorefinery.

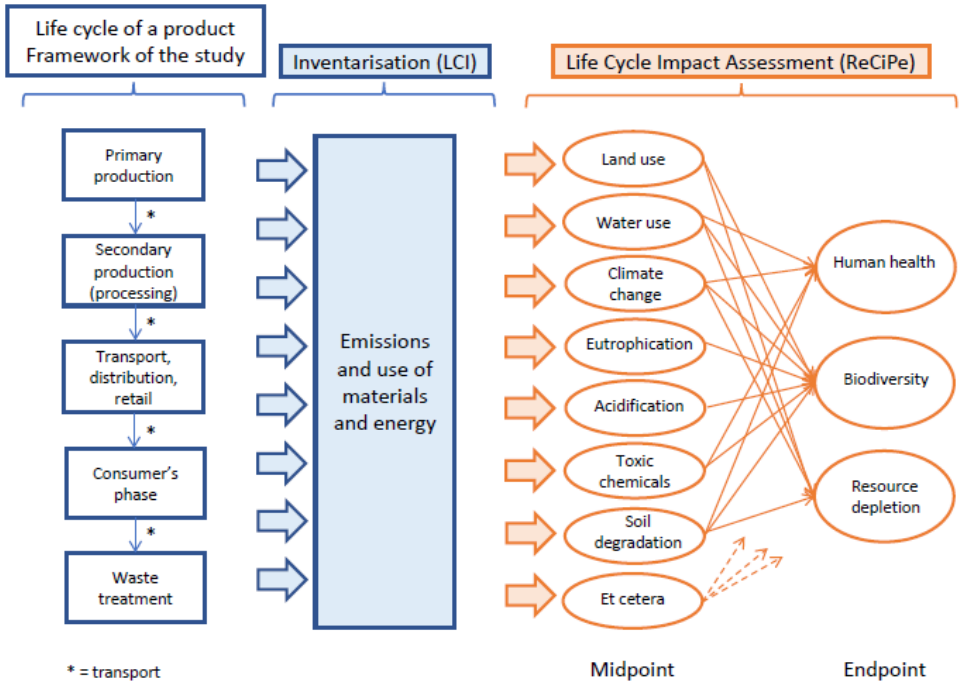


Figure 5-3: An overview of the impact categories included in ReCiPe 2016

5.3 Results:

This study compares the environmental impact of three microalgae biorefineries (Chlorella vulgaris, Haematococcus pluvialis, Nannochloropsis spp.) in the Netherlands, which produce added-value products. The environmental impact of different steps in the microalgae biorefinery and various biomass are also examined. The main indicators are ozone layer depletion, global warming, eutrophication, ecotoxicity, acidification, and human toxicity.

5.3.1 Comparison of three types of microalgae biorefineries

Global warming potential is a major environmental impact factor. The global warming impacts of these three microalgae biorefineries, when using one cubic meter of Dutch influent wastewater, are shown in figure 5-4. The highest greenhouse emissions stem from the Nannochloropsis spp. biorefinery which is equals 903.146 (kg CO_2 eq.). The productivity of this microalgae and percentage of protein and carbohydrate are more than the two others. Thus, more biogases can be produced, which directly affects global warming. The amount of biogas that can be produced in the Chlorella Vulgaris biorefinery is less than in the Haematococcus Pluvialis biorefinery (based on the result of chapter 3). Due to that, the fewest greenhouse emissions are related to the Chlorella Vulgaris biorefinery.

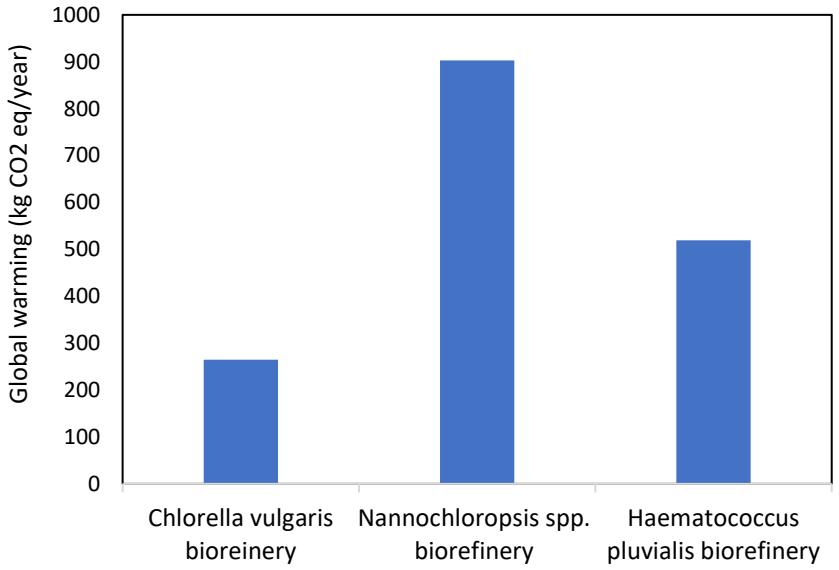


Figure 5-4: Global-warming potential of three types of microalgae biorefineries

Different relative environmental impact categories of these three microalgae are compared in figure 5-5. The absolute value of these environmental impact can be found in Appendix B. However, their functional unit (feedstock) is the same, Nannochloropsis spp. biorefinery produces more microalgae biomass. These enormous amounts of biomass need more required solvents for reaction and utilities, which affect different environmental categories.

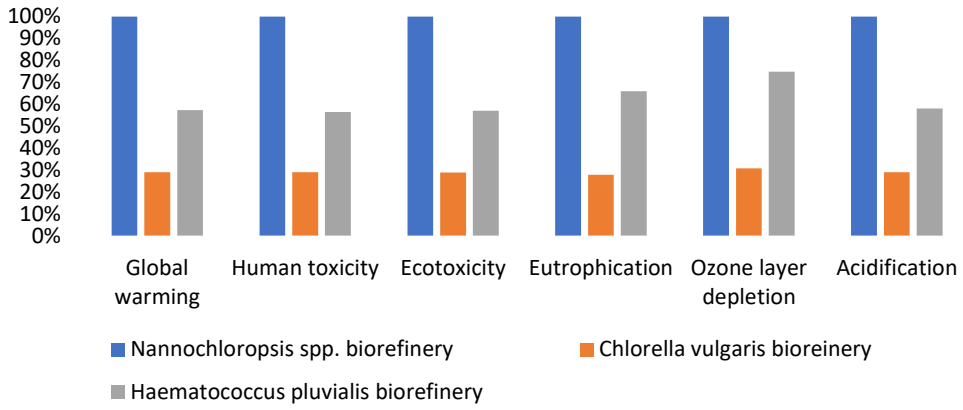


Figure 5-5: Relative environmental impacts of three types of microalgae biorefineries

5.3.2 Comparison of different steps of microalgae biorefinery

The breakdown of the environmental impact for each of the processing steps of Haematococcus Pluvialis biorefinery are presented in figure 5-6. Various environmental impacts of different steps of two other types of microalgae biorefineries can be found in appendix B (figure B-1, 2). The allocation of different products can be done on a mass basis because the inventories for these processes are built on a mass functional unit. The required parameter to calculate allocation coefficients of different products can be extracted from Table C-14-16. In this study different emissions are reported for whole process and products. The required parameter for Ozone molecules are destroyed when chlorine and bromine atoms contact them in the stratosphere [196]. Biomass releases these components during the change process (such as pyrolysis and gasification) [197]. According to the hypothesis in this study, microalgae cells are changed into different components (lipid, pigment, protein, etc.) in the cell disruption unit, and after extracting pigment and lipid, cell is burned in the remnant treatment. Microalgae biomass leaves the biorefinery during dewatering and harvesting process which are effect on Ozone layer depletion. Figure 5-6 (a) shows that cell disruption has the highest ozone depletion impact (0.17 kg CFC 11 eq.) followed by remnant treatment (0.15 kg CFC 11 eq.).

Nitrous oxides (NOx), sulfur dioxide (SO2), and reduced nitrogen (NHx) are the main gases responsible for acidification [198]. In the remnant treatment step, these gases are emissions. Furthermore, these components are present in the fertilizer. Due to these reasons, the remnant treatment has the most acidification impact, as shown in figure 5-6 (f). Furthermore, N-

butanol and acetone are solvents used in lipid extraction and pigment extraction, respectively. Due to the usage of these solvents, different acidification impacts result in microalgae biorefinery.

The term 'toxicity' refers to properties that can harm humans (human toxicity) or ecosystems (ecotoxicity). Phosphate is one of the common components that cause toxicity in humans, such as renal function and rhabdomyolysis [199]. This component left the microalgae biorefinery in the remnant treatment stage. Due to that, the remnant treatment has a high human toxicity impact (72322.3 kg 1,4-DCB) and ecotoxicity (62109.4kg 1,4-DCB), as shown in figure 5-6 (c & d). In addition to phosphate, ammonia is another main substance contributing to ecotoxicity [200]. Microalgal biorefineries also produce ammonia as a fertilizer. Figure 5-6 (d) shows that remnant treatment has the highest ecotoxicity impact (62109.4kg 1,4-DCB).

Several factors influence the eutrophication of water, such as nutrient enrichment, hydrodynamics, temperature, salinity, carbon dioxide, element balances, microbial diversity, and environmental factors, such as temperature and salinity [201]. Regarding Dutch influent wastewater does not present in the Agribalyse database, ammonia, phosphate, and sulfate are added to water to simulate wastewater. Due to this assumption, cultivation and harvesting have the highest eutrophication impact, as seen in figure 5-6 (e). Some water leaves the biorefinery as the waste stream that have eutrophication effect as well.

A remnant treatment has the highest global warming effect due to energy consumption and methane production (biogas) [202]. Figure 5-6 (b) shows that remnant treatment has the highest global warming impact (718814 kg CO_2 eq.) followed by cell disruption (253642 kg CO_2 eq.). It is assumed that biomass is changed to different cell components in cell disruption, which impacts global warming. Thus, cell disruption has a 253642(kg CO_2 eq.) global warming impact. Furthermore, energy consumption is a reason for global warming's impact on pigment extraction, cultivation, harvesting, lipid extraction.

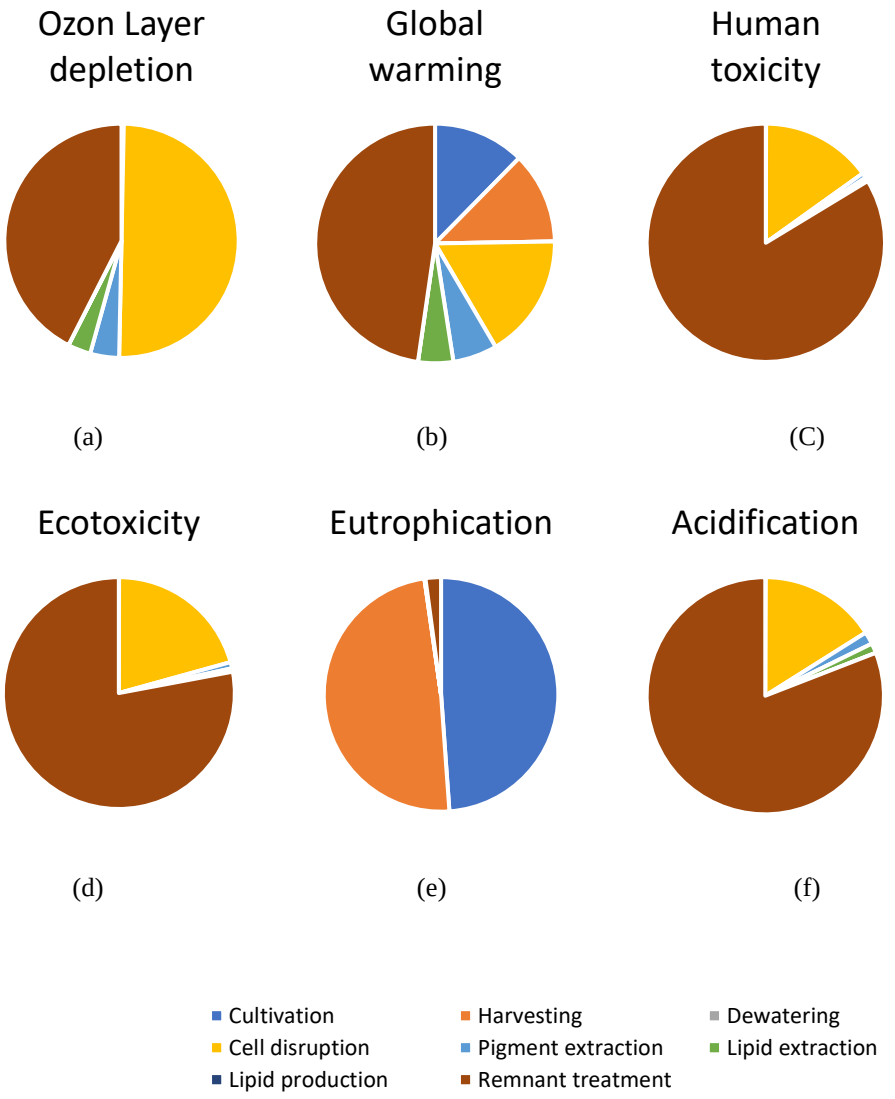


Figure 5-6: (a) Ozon layer depletion, (b) global warming, (c) human toxicity, (d) ecotoxicity, (e) eutrophication, (f) Ozone formation, and (g) acidification impact of different steps of Haematococcus Pluvialis biorefinery

The remnant treatment has the highest different environmental impacts. Although huge investments (capital and operating cost) are needed (due to remaining part of microalgae cell is coming to this step of biorefinery), small revenue are brought to the biorefinery (due to the low prices of biogas and fertilizers) with high negative effects on the environment. Increasing profitability and reducing the environmental impact of this step should be further considered.

5.3.3. Environmental impact versus economics of conventional and microalga processes

To compare the environmental impact of producing 1 kg pigment in a microalgae biorefinery and a conventional production pigment process, the extraction of β-carotene from Carrot Daucus carota L is considered. The functional unit for this study is 1 kg β-carotene. The results of a previous LCA study when producing β-carotene from carrot with conventional solvent, are utilized[203]. While 0.1%-3% of microalgae cell is pigment, 10% weight of carrot is β-carotene [203]. In comparison with carrots, more microalgae are needed to cultivate and more solvent and utilities are used to produce 1 kg β-carotene. Figure 5-7 shows that the microalgae biorefinery has a greater impact on global warming than the carrot refinery which is like the comparative study [203]. For instance, the global warming effect of chlorella vulgaris, Nannochloropsis spp. Haematococcus Pluvialis biorefineries are 18,231, 1.6 times more than carrot refinery. The production cost of different microalgae/ carrot (bio)refineries versus different environmental indicators are shown in appendix B. Figure 5-7 is also shown the production cost of microalgae biorefinery is more than carrot refinery when producing 1 kg β-carotene. As a result, microalgae biorefineries will need to be improved to make them more productive, both from an economic and environmental perspective.

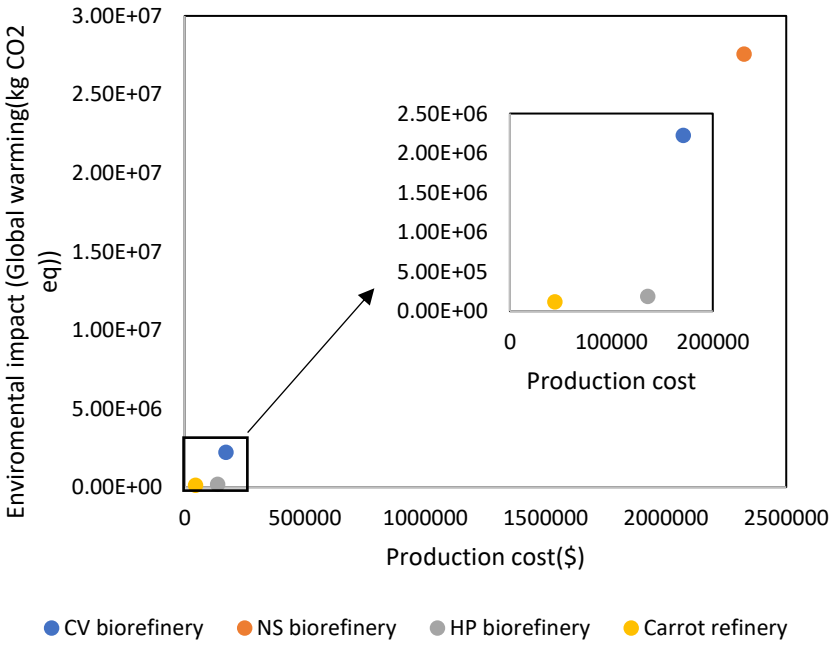


Figure 5-7: Environmental impact (global warming) of producing β-carotene from carrot and microalgae

Conclusion

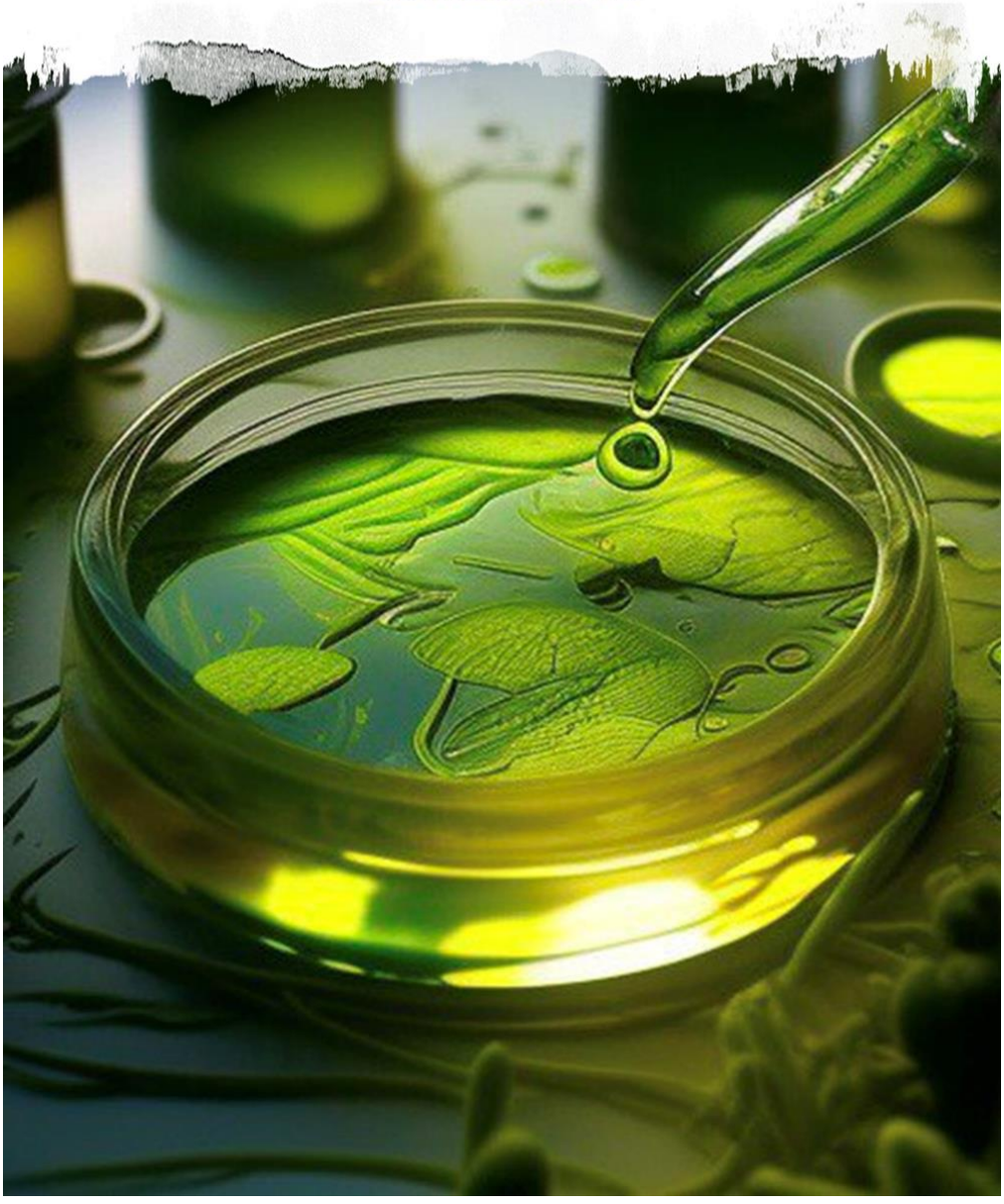
A cradle-to-gate LCA study was performed to compare the performance of three types of microalgae biorefineries in terms of environmental impact. The focus was twofold; in one case the feedstock (wastewater) was used as functional unit in the other case the main product (pigment) was used as a functional unit. The ReCiPe2016 method and Agribalyse database are used to assess the environmental impact in terms of the following midpoint categories: ozone layer depletion, global warming potential, human toxicity, ecotoxicity, eutrophication and acidification. The LCA results show that although the feedstock (one cubic meter of Dutch influent wastewater) and environmental condition are same for these microalgae biorefineries, Nannochloropsis spp. has high productivity and biomass production. Due to that, the environmental impacts of this biorefinery are more than two others biorefineries.

The environmental impacts of different steps of microalgae biorefinery are compared when producing 1 kg of pigment. By disrupting the cells, microalgae are converted into lipid, pigment, and other components, and most of these components are burned in the remnant treatment to produce biogas and fertilizers. The remnant treatment has the highest global warming potential, human toxicity, ecotoxicity, and acidification impacts forward by cell disruption.

A comparison was conducted between the production of 1 kg of β-carotene (as an example of pigment) from carrots and these microalgae. As compared to a carrot refinery, microalgae biorefineries have a more significant environmental impact (For instance, the global warming effect of chlorella vulgaris Nannochloropsis spp. Haematococcus Pluvialis biorefineries are 18,231, 1.6 times more than carrot refinery). In addition, carrots produce this pigment at a lower cost than microalgae (approximately 50000$).

Chapter Six

Conclusions



In this thesis, a framework is developed to find a pathway (economically and environmentally) for producing added-value products from several types of microalgae, considering the uncertain conditions of feedstocks. Three types of microalgae biorefineries (Cholera Vulgarise, Haematococcus Pluvialis, Nannochloropsis spp.) are considered that produce six added-value products (pigment, biodiesel, glycerol, omega-3, fertilizer, and biogas). To reach this goal three tasks must be executed: 1) development of superstructure optimization framework for identifying promising production routes, 2) The systematic assessment of uncertainty throughout the decision process and 3) the balancing of economic gains with environmental impact via a life cycle assessment.

First, a superstructure with all available 23 technologies for nine steps (cultivation, harvesting, dewatering, drying, cell disruption, pigment extraction, lipid extraction, lipid production, and remnant treatment) of microalgae biorefineries is developed. This superstructure model is formulated as a mixed integer nonlinear program (MINLP) containing 6710 variables and 6161 constraints. The model is optimized with two solvers (global solver: BARON, local solver: AOA) in AIMMS version 4.82.3.29 64-bit.

To decrease the problem size and to increase the computational efficiency a new modelling approach is proposed. In this approach, individual blocks can be lumped with a so-called block integration. In addition to optimizing the superstructure of microalgae biorefineries, this approach can also be used to optimize the superstructures of other types of refineries. Results of optimizing the MINLP model show that the type of microalgae does not influence the decision-making regarding the different processing steps and the optimized production pathway is: open pond, sedimentation and flotation, flocculation without any dryer, sonication, organic solvent pigment extraction, n-butanol solvent lipid extraction, lipid production, and anaerobic digestion. However, the annual profit of the Haematococcus Pluvialis biorefinery is highest with (62$/ kg of microalgae due to the high pigment price (3608.5 $/kg). This biorefinery produces approximately 500 t of pigment bioproducts from 24 Kt biomass by using 200 Kt of wastewater and 164 Kt of carbon dioxide

The uncertain character of these feedstocks affects the annual profit and quantities of bioproducts of microalgae biorefineries. For this reason, three types of uncertainty in the feedstocks (composition of wastewater and sunshine duration, and simultaneous effect of composition of wastewater and sunshine duration) are considered in the decision-making process. For this Dutch influent wastewater data of the period 1989-2019 and sunshine duration data of the Netherlands for this period are extracted from the literature. From a Monte Carlo simulation, the average profit margin of Haematococcus Pluvialis biorefinery (as highest profitable microalgae biorefinery) is 62.98 $/kg. This microalgae biorefinery has

80% probability of earning profit margin between 62.55 ($/kg) and 62.98 ($/kg), according to a cumulative distribution function curve.

Furthermore, annual profit margin of microalgae biorefinery are reduced by approximately 50% when using actual sunshine duration in the Netherlands. Under uncertainty of wastewater composition and sunshine duration, the profits margins of Chlorella vulgaris, Haematococcus pluvialis, and Nannochloropsis spp. have 95% probability to reach 1.6±0.4 ,31±7, 0.33±0.1 ($/kg), respectively.

The selected pathways from the superstructure optimization are used to compute the environmental impacts using two functional units: 1) the feedstock, where one cubic meter of Dutch influent wastewater is used as a base and 2) the product, where 1 kg of pigment, is used. These biorefineries are compared via different midpoint categories: ozone layer depletion, global warming, human toxicity, eutrophication, ecotoxicity and acidification. LCA studies of these three microalgae biorefineries show that Nannochloropsis spp. biorefinery has the most different environmental impacts due to high productivity. The remnant treatment has the greatest impact on global warming, human toxicity, ecotoxicity, and acidification. Cultivation and harvesting have the most eutrophication impact.

Chapter Seven

Outlook

This study suggests that pigment extraction from microalgae is economically competitive with biodiesel or biogas. As pigments have a variety of polarities, choosing the best extraction method is crucial [204]. It is also important that the right solvents be chosen according to the application of the pigment. Microalga types and cultivation conditions also affect their quantity. Another factor that influences the efficiency of extraction is optimizing the extraction process's operating conditions. Pressure, for example, has a negligible effect in contrast to temperature, which has a significant impact [205]. On the other hand, the experimental research study at pilot/industrial scale of pigment extraction is underpowered. The use of supercritical CO_2 for large-scale astaxanthin extraction has received an increasingly interest due to its efficiency and environmental friendliness as compared to other solvents [206].

Standardizations and simplifications are required to effectively use superstructure optimization. This approach can be used to get first estimates on energy consumption, unit operations' sizes, and costs associated with setting up and maintaining production, but individual operations are not described in detail. It is therefore essential to analyse the most promising pathways in a more rigorous manner. A sophisticated process modelling and simulation software should be used to determine whether the production costs of both added-value products and fuels are competitive with market prices under various conditions. For this purpose, the optimal unit operation and process conditions must be chosen to fulfil a given step in the production line.

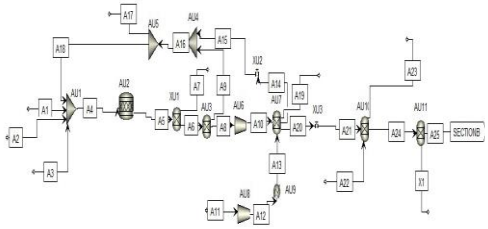
In addition, the value chain must be represented accurately and effectively to ensure that the economic potential of the operation is not overestimated. Very few simulation studies and analyses in terms of economic outlook have been dedicated to combining the process of extracting bioproducts from microalgae. These researchers concentrated almost entirely on specific sections of an algae biorefinery, simplifying the remaining sections significantly and ignoring, for example, the impact of tertiary unit operations [207–209].

In this study, some efforts have been made to simulate the production pathway chosen by optimizing the superstructure in Aspen Plus software. As seen in figure 7-1, the pathway is divided into six sections named:

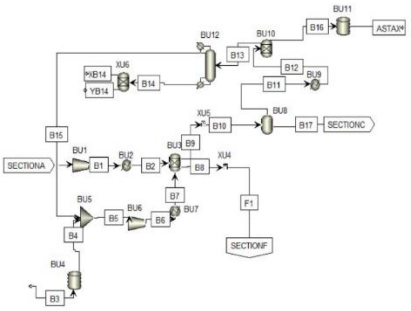
- Section A (harvesting, dewatering, and cell disruption)
- Section B (extraction)
- Section C (glycerol production)
- Section D (biodiesel production)
- Section E (omega-3 production)

- Section F (remnant treatment)

Such division helps to increase the speed of simulating, (de)activate section(s) of study and optimize the operation of conditions of one section separately.



(Section A)



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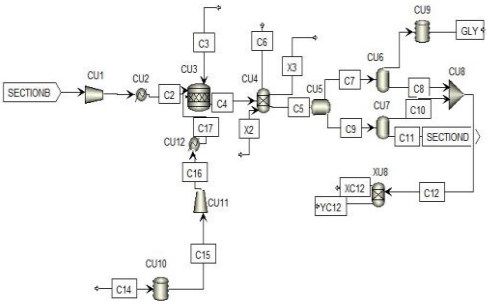

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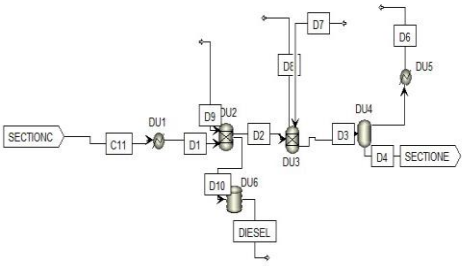

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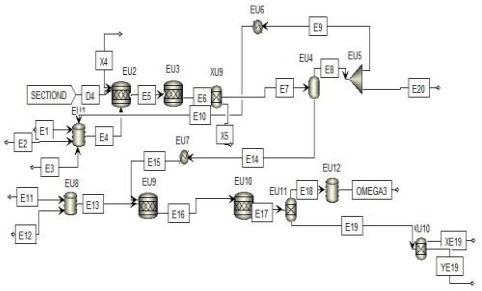

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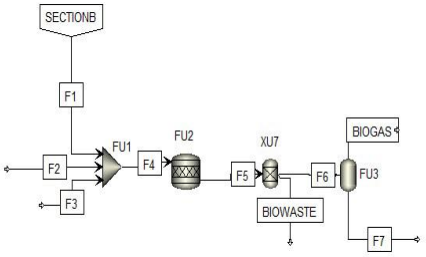

(Section F)

Figure 7-1: Flowsheet of microalgae biorefinery

Some assumptions are made to simulate the pathway completely. Some important ones are:

- Microalgae biomass is simulated in a liquid phase instead of a solid.
- Splitters are utilized instead of distillation columns.
- Instead of simulating the cell as a component, different components of the cell (lipid, pigment, etc.) are simulated separately.
- Pigments only contain astaxanthin. Phenylalanine, triolein, and sucrose are considered to be amino acids, lipids, and carbohydrates, respectively.
- The growth pond is simulated as a reactor. To simulate a pond in different environmental temperatures, external energy is defined.

Some efforts to improve simulation model should be done to improve accuracy of results. For instance, parameters of components (such as astaxanthin) should be evaluated and complete. Appropriate equation of state should be chosen to cover all operation conditions (high/low pressure or temperature). In addition, simulating solid and liquid phase at the same time and components which are like polymers are challenge of simulation model.

Furthermore, the superstructure model can be solved more accurately by providing a detailed cost calculation, especially for land costs. The accuracy of the current system is 30% to 35% due to Lang's factor method [210] used to calculate capital investment cost, uncertainty price of land and material and quantity of products. Over the past two decades, genome-scale models have steadily improved to study cellular growth. With this model, we can accurately predict microalgae biomass, one of the sources of uncertainty regarding the quantity of products[211].

Although in this thesis the outcomes of the superstructure in terms of economic performance have been compared to life cycle analysis, a systematic multi-criterion optimization has not been executed and, such extension would surely be welcomed [212].

Abbreviations and nomenclature

Abbreviations

ASP	Aquatic Species Initiative
BPBR	Bubble column photobioreactors
CDF	Cumulative distribution function
CV	Chlorella Vulgaris biorefinery
FPBR	Flat plate photobioreactors
GGE	Gasoline gallon equivalent
GHGs	Greenhouse gases
GWP	Global warming potential
HP	Haematococcus Pluvialis biorefinery,
IPCC	International Panel on Climate Change
LCA	Life cycle assessment
LCI	Life cycle inventory
LCIA	Life cycle impact assessment
MINLP	Mixed-integer nonlinear programming
NPV	Net present value
NREL	National Renewable Energy Laboratory
NS	Nannochloropsis spp.
OP	Open pond
PDF	Probability density function
PUFA	Polyunsaturated fatty acids
SC	Supply chain
SWOT	Strengths, weaknesses, opportunities, and threats
TPBR	Turbo photobioreactors,
UNFCC	United Nations Framework Convention on Climate Change

WWT	Wastewater treatment

Nomenclature

1. Variables

M	Mass flow (t/h)
F	Feedstock stream (t/h)
U	Utility
PM	Profits margin ($/t)
AIC	Annualized investment cost ($)
AOC	Annualized operating cost ($)
PS	Product sales ($/t)
TIPC	Total installation plant cost ($)
P	Price ($)
K	Coefficient (Factor)
Y	Binary decision variable

2. Parameters

X	Concentration of reactant
S	Mass stochiometric coefficient
CF	Conversion factor
D	Distribution coefficient
SF	Split factor
SUC	Specific utility consumption factor
IR	Interest rate
LT	Lifetime
EC	Equipment cost
LC	Land cost

IDX	Cost index
F	Sizing factor
UC	Utility cost
OMC	Operating and maintenance cost ($)
WTC	Waste treatment cost ($)
RMC	Raw material cost ($)

3.Subscripts

H	stage
J	alternative
K	component

4.Superscripts

IN	Input stream
U	Upstream
R	Reactant stream
Out	Output stream
W	Waste stream
P	Product stream
E	Electricity
H	heating
C	cooling
ref	reference
OM	Operating and maintenance
ENG	Engineering

Appendix

Appendix A: Block integration approach

The lipid production stage involves the production of biodiesel, omega-3, and glycerol, which need to be separated from one another. It involves three reactions and four separations[101]. Figure A-1 shows the required unit operations for this stage. Several series of technical data should be used to describe this stage, e.g., using a split factor for each separation unit and a conversion factor for each reaction unit.

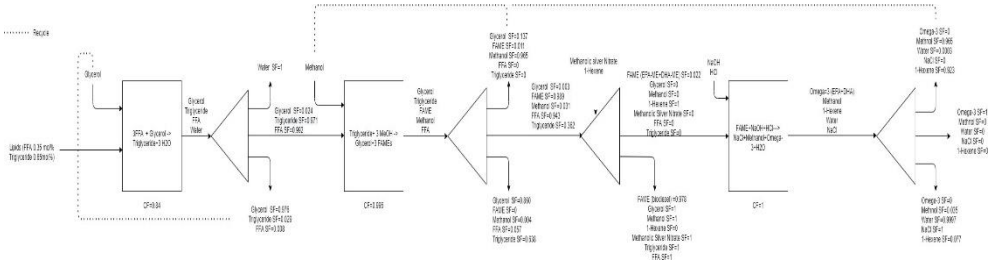

Figure A-1: Process sketch for lipid production

It is possible to integrate all the unit operations of the lipid production stage into one, which means there is only one overall reaction and one overall separation in this alternative. In this overall reactor, all the reactants are added at once, and all the products can be produced simultaneously. Also, the overall separation can separate all the products at once. All processes can be integrated as one by only considering inputs and outputs. The modelling process should now be based on only one set of technical data. The approaches to calculate overall reaction, conversion, and split factor are explained below.

A-1: Overall reaction

Lipid production involves three main reactions. Based on the compositions of the lipid, the overall reaction should be defined. 1 mol lipid is assumed to contain 0.35 mol free fatty acids (FFA) and 0.65 mol triglycerides (Eq. A-1) [101]. Free fatty acids are converted to triglycerides with glycerol (Eq. A-2). A second reaction involves converting triglyceride into glycerol and fatty acid methyl esters (FAMEs) with methanol (Eq. A-3). The combination of these reactions is shown in Eq. A-4.

Lipid = 0.35 Free fatty acid + 0.65 Triglyceride (Eq. A-1)

3 Free fatty acid + Glycerol → Triglyceride + 3 H2O (Eq. A-2)

Triglyceride + 3 Methanol → Glycerol + 3 FAMEs (Eq. A-3)

Lipid + 2.3 Methanol → 2.3 FAMEs + 0.65 Glycerol + 0.35 H2O (Eq. A-4)

FAMEs consist of two types of fatty acid components (long-chain fatty acids and short-chain fatty acids). Instead of considering FAMEs as final products (biodiesel), long-chain fatty acids can be separated from short ones with sodium hydroxide and hydrochloric acid to produce omega-3. About 10% of long chain fatty acids can be separated to produce omega-3 fatty acids, and the remaining is used in biodiesel flow. The percentage of lipids' long-chain and short-chain fatty acids varies based on the type of microalgae species. For instance, approximately 22 mole % of lipid of Chlorella Vulgaris is a long chain fatty acid. Eq. A-5-7 shows the overall mass reaction of lipid production of Chlorella Vulgaris, Haematococcus Pluvialis, and Nannochloropsis spp. respectively.

Lipid + 0.003 Sodium hydroxide + 0.003 Hydrochloric acid + 0.104 Methanol → 0.984 Biodiesel+ 0.024 Omega-3 + 0.004Sodium chloride + 0.087Glycerol + 0.010Water

(Eq. A-5)

Lipid + 9.88×10^{-6} Sodium hydroxide + 9.01×10^{-6}Hydrochloric acid + 0.113 Methanol → 1.012 Biodiesel+ 7.47×10^{-5}Omega-3 + 1.44×10^{-5}Sodium chloride + 0.092Glycerol + 0.010Water

(Eq. A-6)

Lipid + 5.0×10^{-4} Sodium hydroxide + 4.6×10^{-4}Hydrochloric acid + 0.115 Methanol → 1.008 Biodiesel+ 0.004Omega-3 + 7.0×10^{-4}Sodium chloride + 0.094Glycerol + 0.010Water

(Eq. A-7)

A-2: Overall conversion factor and split factor

Calculating a conversion factor of an overall reaction based on biodiesel is possible. This is a ratio between the amount of biodiesel produced in a real and the ideal situation. Stoichiometric coefficients of the overall reaction represent the amount of biodiesel in an ideal situation. The simulation study can calculate the amount of biodiesel in the real situation [101].

A previous simulation study shows that Nannochloropsis can produce four streams (waste, glycerol, biodiesel, omega-3) with different compositions. Table A-1 shows different streams' compositions when 1-mole lipid enters the lipid production stage. The main components of biodiesel are triglycerides, free fatty acids, and FAMEs. In this case, the total amount of biodiesel is the summation of these components, which is 2.205 moles. This means that in real situations, 1 mole of lipid can produce 2.205 mol of biodiesel product. In the other hand, the Eq-A-5 can be used to find the amount of biodiesel in an ideal situation which is 2.292. Thus, the overall conversion factor is 2.205/2.292=0.962. The conversion factor for Chlorella vulgaris and Haematococcus pluvialis is assumed to be the same as Nannochloropsis spp.

Table A-1: Component amounts for different streams.

Component	**Waste**	**Glycerol**	**Biodiesel**	**Omega-3**	**Total**
Triglyceride(mol)	0.021	0.002	0.001	0	0.024
Free fatty acids (mol)	0.0005	0.003	0.052	0	0.056
FAMEs(mol)	0.024	0	2.101	0	2.125
Glycerol(mol)	0.244	0.626	0.002	0	0.872
Methanol(mol)	0.288	0.001	0.009t	0	0.298

An overall split factor needs to be calculated for each component. The split factor is calculated as the amount towards one stream over the total amount towards all streams. An example is biodiesel, a mixture of triglycerides, free fatty acids, and FAMEs. According to table A-1, the split factors for biodiesel in waste, glycerol, biodiesel, and omega-3 are 0.021,0.002,0.977, and 0, respectively.

This approach is implemented for other components of different microalgae biorefinery.

Appendix B: LCA results

Table B-1: Environmental impacts of microalgae biorefineries when using one cubic meter of Dutch influent wastewater

Indicators	**Nannochloropsis spp. biorefinery**	**Chlorella vulgaris biorefinery**	**Haematococcus pluvialis biorefinery**	**Unit**
Global warming	2.65E+02	9.03E+02	5.19E+02	kg CO2 eq
Human toxicity	1.04E+01	3.55E+01	2.01E+01	kg 1,4-DCB
Ecotoxicity	1.06E+01	3.64E+01	2.08E+01	kg 1,4-DCB
Eutrophication	-1.78E+00	-6.34E+00	-4.19E+00	kg N eq
Ozone layer depletion	6.66E-04	2.28E-03	1.33E-03	kg CFC11 eq
Acidification	4.96E+00	1.70E+01	9.87E+00	kg SO2 eq

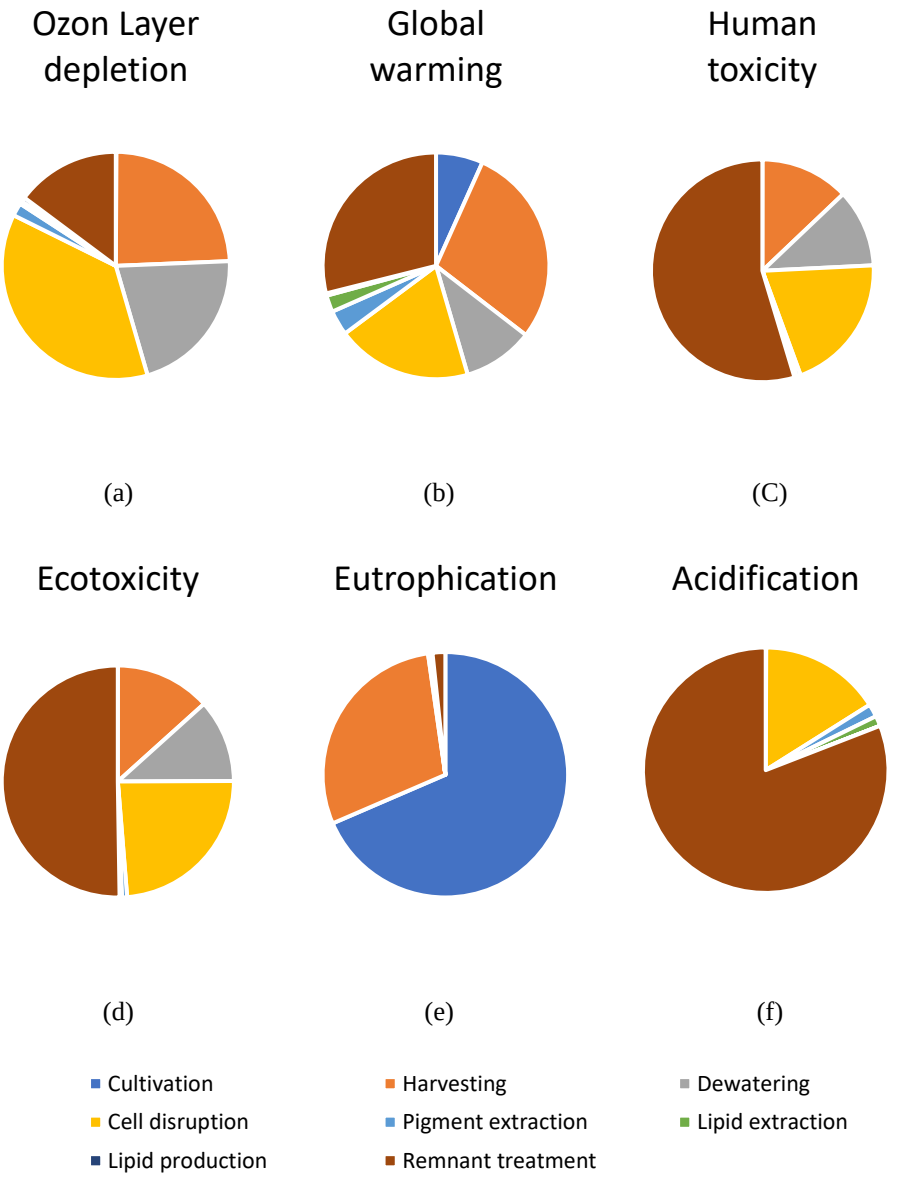

Figure B-1: (a) Ozon layer depletion, (b) global warming, (c) human toxicity, (d) ecotoxicity, (e) eutrophication, (f) Ozone formation, and (g) acidification impact of different steps of Chlorella vulgaris biorefinery

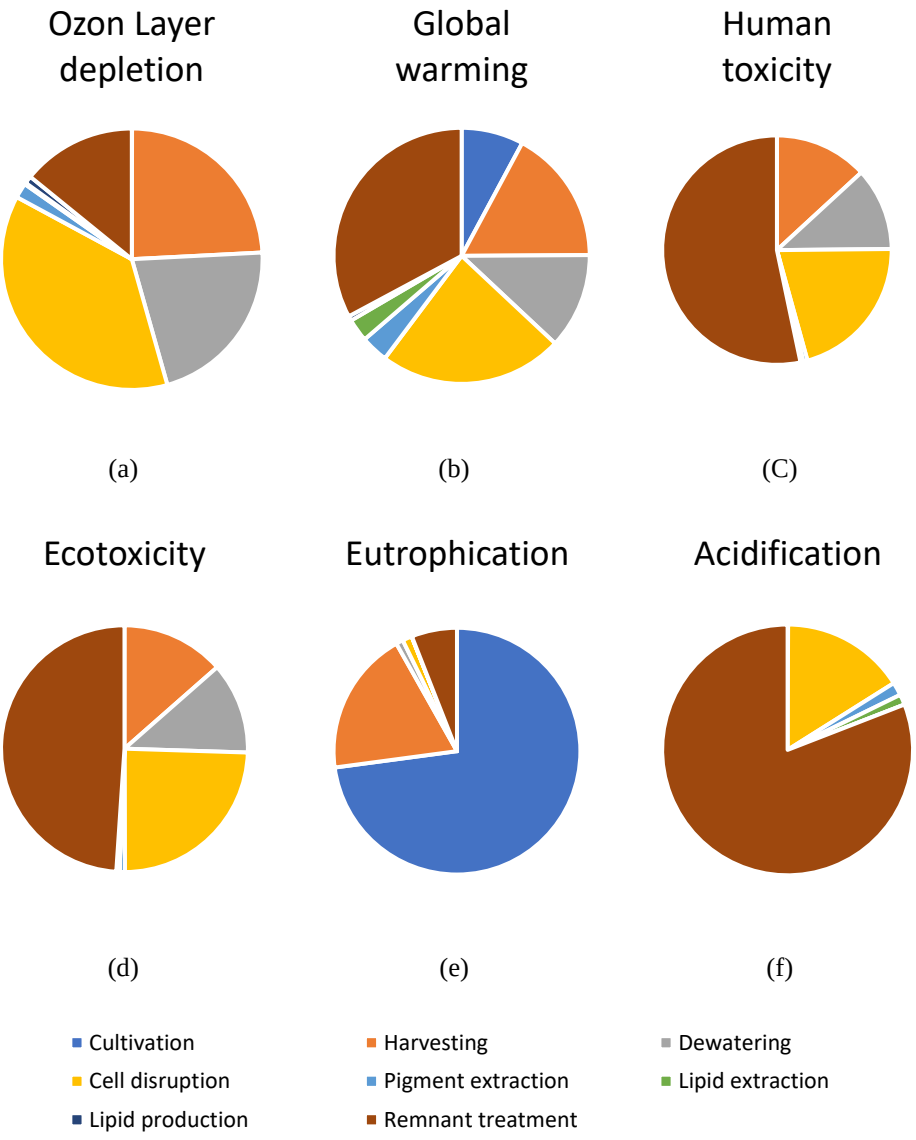

Figure B-2: (a) Ozon layer depletion, (b) global warming, (c) human toxicity, (d) ecotoxicity, (e) eutrophication, (f) Ozone formation, and (g) acidification impact of different steps of Nannochloropsis Spp. biorefinery

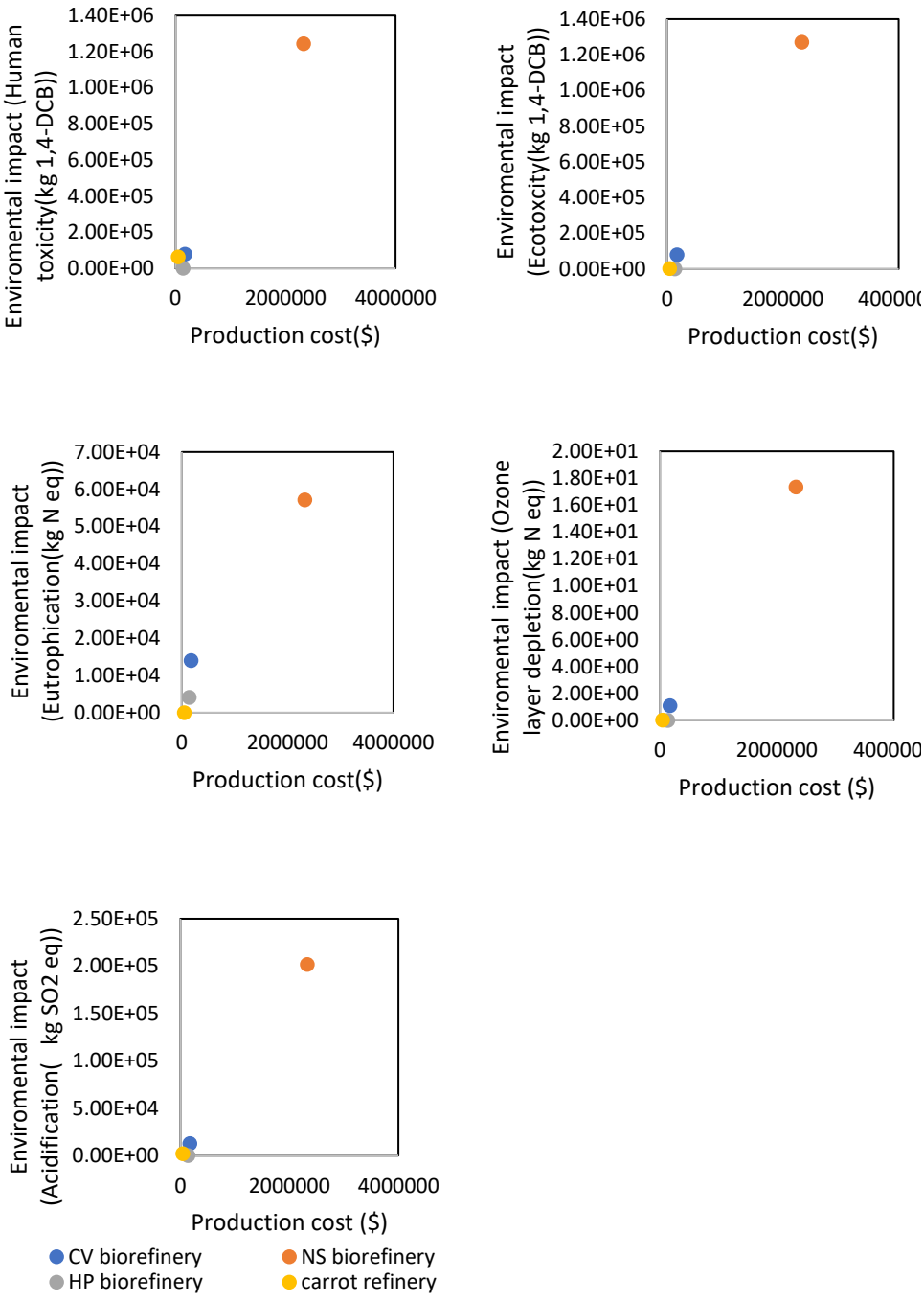

Figure B-3: The production cost of different microalgae/ carrot (bio)refineries versus different environmental indicators

Appendix C: Supplement data

Table C-1: Composition of influent wastewater [132]

Component	**Influent wastewater (t/h)**
water	224.59
Chemical oxygen demand (COD)	128.35
Biochemical oxygen demand (Bod)	53.83
Nitrogen compounds as N (total)	76.37
Phosphorus compounds as P (total)	11.88
Copper	0.02
Chromium	0.01
Zinc	0.06
Lead	0.01
Cadmium	5.11E-05
Nickel	0.01
Mercury	2.65E-05
Arsenic	8.41 E-04

Table C-2: Concentration factor (k, j) of component (k) in alternative (j)[100,145,213]

Alternative (*j*)	**Component (*k*)**	**Concentration factor (g/g)**
Flocculation	Flocculant	3×10^{-6}
Organic solvent-based pigment extraction	Organic solvent	15.7
Supercritical carbon dioxide-based pigment extraction	Carbon dioxide	5
Supercritical carbon dioxide-based pigment extraction	Acetone	0.56
Hexane solvent-based lipid extraction	Hexane	2
n-butanol solvent-based lipid extraction	n-butanol	1
Supercritical carbon dioxide-based lipid extraction	carbon dioxide	6.85

Table C-3: Concentration factor ($k, 22$) of component (k) in lipid production intervals (22) (for different type of microalgae[101,142,214,215]

Microalgae species	**Component (*k*)**	**Concentration factor (g/g)**
Chlorella vulgaris	Hexane	3.01×10^{-5}
	Methanol	0.11
	Methanolic Silver Nitrate	5.51×10^{-7}
	Sodium hydroxide	0.003
	Hydrochloric acid	0.003
Haematococcus pluvialis	Hexane	3.18×10^{-5}
	Methanol	0.12
	Methanolic Silver Nitrate	5.83×10^{-7}
	Sodium hydroxide	1.03×10^{-5}
	Hydrochloric acid	9.37×10^{-6}
Nannochloropsis spp.	Hexane	3.24×10^{-5}
	Methanol	0.12
	Methanolic Silver Nitrate	5.95×10^{-7}
	Sodium hydroxide	5.23×10^{-4}
	Hydrochloric acid	4.78×10^{-4}

Table C-4: Mass stoichiometric coefficient of reactions in lipid production intervals [101,142,215,216]

Microalgae species	**Component (*k*)**	**Stoichiometric coefficient (-)**
Chlorella vulgaris	Water	0.01
	Lipid	-1
	Glycerol	0.08
	Methanol	-0.104
	Sodium hydroxide	-0.003
	Hydrochloric acid	-0.003
	Sodium chloride	0.004
	Omega-3	0.024
	Biodiesel	0.984
Haematococcus pluvialis	Water	0.01
	Lipid	-1
	Glycerol	0.09
	Methanol	-0.11
	Sodium hydroxide	-9.89×10^{-6}
	Hydrochloric acid	-9.01×10^{-6}
	Sodium chloride	1.44×10^{-5}
	Omega-3	7.47×10^{-5}
	Biodiesel	1.01
Nannochloropsis spp.	Water	0.01
	Lipid	-1
	Glycerol	0.094

Methanol	-0.12
Sodium hydroxide	-5.0×10^{-4}
Hydrochloric acid	-4.6×10^{-4}
Sodium chloride	7.0×10^{-4}
Omega-3	0.004
Biodiesel	1.01

Table C-5: Reaction conversion factor ($CF_{k,j}$) of component (k) in alternatives (j) [101,145,216,217]

Alternative (*j*)	**Component (*k*)**	**Conversion factor (-)**
Open pond/Flat plate photobioreactor/Bubble column photobioreactor/ turbo column photobioreactor	Carbon dioxide	0.75
Bead beating/high-pressure homogenization/ Microwaving/ Sonication	Microalgae	1
Lipid production	Lipid	0.96

Table C-6: Distribution factor ($D_{k,j}$) of component (k) in anaerobic digestion alternative (j) [145]

Component (*k*)	**Distribution factor (g/g)**
Water	0.29
Ammonia	0.01
Phosphate	0.001
Biogas	0.39
Salt	0.107
Solid	0.20

Table C-7 (a): Split factor ($SF_{k,j}$) of component (k) in downstream flows of alternative (j) [98,100,222,101,126,145,152,218–221]

Alternative (*j*)	**Split factor of different components (-)**
Open pond/Flat plate photobioreactor/Bubble column photobioreactor/ turbo column photobioreactor	Water 1, Ammonia 1, Phosphate 1, Sulphate 1, Microalgae 1
Sedimentation and flotation	Water 0.001, Microalgae 0.893
Sedimentation and filtration	Water 0.00032, Microalgae 0.89, flocculant 0
Flocculation	Water 0.246, Microalgae 0.95
Centrifugation	Water 0.086, Microalgae 0.95
Filter press	Water 0.086, Microalgae 0.95
Dryer	Water 0.176, Microalgae 1
Bead beating/high-pressure homogenization/ Microwaving/ Sonication	Pigment 1, Lipid 1, Other cell composition 1
Organic solvent pigment extraction	Water 1, Pigment 0.33, Lipid 1, Other cell composition 1
Supercritical carbon dioxide pigment extraction	Water 1, Pigment 0.41, Lipid 1, Other cell composition 1
Hexane based lipid extraction	Water 1, Pigment 1, Lipid 0, Other cell composition 1, Hexane 0.002
n- butanol based lipid extraction	Water 1, Pigment 1, Lipid 0, Other cell composition 1, n-butanol 0.017
Supercritical carbon dioxide lipid extraction	Water 1, Pigment 1, Lipid 0, Other cell composition 1

Table C-7 (b): Split factor ($SF_{k,j}$) of component (k) in waste flows of alternative (j) [98,100,222,101,126,145,152,218–221]

Alternatives (*j*)	**Split factor of different components (-)**
Open pond/Flat plate photobioreactor/Bubble column photobioreactor/ turbo column photobioreactor	Carbon dioxide 1, oxygen 1
Sedimentation and flotation	Water 0.999, Microalgae 0.117
Sedimentation and filtration	Water 0.99968, Microalgae 0.117, flocculant 1
Flocculation	Water 0.754, Microalgae 0.05
Centrifugation	Water 0.914, Microalgae 0.05
Filter press	Water 0.914, Microalgae 0.05
Dryer	Water 0.824, Microalgae 0
Organic solvent pigment extraction	Organic solvent 1
Supercritical carbon dioxide pigment extraction	Ethanol 1, Carbon dioxide 1
Hexane based lipid extraction	Hexane 0.998
N- butanol based lipid extraction	n-butanol 0.983
Supercritical carbon dioxide lipid extraction	Carbon dioxide 1
Lipid production	Water 1, Hexane 1, Glycerol 0.28, Methanol 0.966 methanolic silver nitrate 1, sodium hydroxide 1, Hydrochloric acid 1, sodium chloride 1, biodiesel 0.021
Anaerobic digestion	Water 0.9, carbon dioxide 0.9, Ammonia 1, phosphate 1, sulphate 1, biogas 0.01, salt 0.9

Table C-7 (c) : Split factor ($SF_{k,j}$) of component (k) in different products flows of alternative (j) [98,100,222,101,126,145,152,218–221]

Alternatives (*j*)	**Types of product flow**	**Split factor of different components (-)**
Organic solvent pigment extraction	Pigment	Water 0, Pigment 0.67, Lipid 0, Other cell composition 0
Supercritical carbon dioxide pigment extraction	Product	Water 0, Pigment 0.59, Lipid 0, Other cell composition 0
Hexane based lipid extraction	Lipid	Water 0, Pigment 0, Lipid 1, Other cell composition 0
n- butanol based lipid extraction	Lipid	Water 0, Pigment 0, Lipid 1, Other cell composition 0
Supercritical carbon dioxide lipid extraction	Lipid	Water 0, Pigment 0, Lipid 1, Other cell composition 0
Lipid production	Biodiesel	Glycerol 0.003, Methanol 0.03, biodiesel 0.977
	Glycerol	Glycerol 0.717, biodiesel 0.002
	Omega-3	Methanol 0.004, omega-3 1
Anaerobic digestion	Biogas	Carbon dioxide 0.1, biogas 0.99,
	Fertilizer	Water 0.1, salt 0.1, solid 1

Table C-8: Specific utility (electricity, heating, cooling) consumption (SUC_j) of alternative (j) [101,125,229,145,222–228]

Alternatives (j)	**Specific electricity consumption (MWh/t)**	**Specific heating consumption (MWh/t)**	**Specific cooling consumption (MWh/t)**
Open pond	1.5×10^{-3}	-	-
Flat plate photobioreactor	1.5×10^{-3}	-	-
Bubble column photobioreactor	1.5×10^{-3}	-	-
turbo column photobioreactor	1.5×10^{-3}	-	-
Sedimentation and flotation	6.25×10^{-2}	-	-
Sedimentation and filtration	8.8×10^{-4}	-	-
Centrifugation	6.0×10^{-2}	-	-
Filter press	8.8×10^{-4}	-	-
Dryer	19.445	-	-
Bead beating	140	-	-
high-pressure homogenization	146.94	-	-
Microwaving	116.67	-	-
Sonication	36.67	-	-
Organic solvent pigment extraction	-	4.972	0.195
Supercritical carbon dioxide pigment extraction	2.25	81.926	0

Hexane based lipid extraction	-	4.972	0.195
n- butanol based lipid extraction	-	4.304	9.342
Supercritical carbon dioxide lipid extraction	2.25	81.926	-
Lipid production	0.01	0.154	0.157
Anaerobic digestion	13.93	0.1	-

Table C-9: List and number of required equipment for each alternative

Alternatives (j)	**(number) type of equipment**
Open pond	(1) Open pond
Flat plate photobioreactor	(1) Flat plate photobioreactor
Bubble column photobioreactor	(1) Bubble column photobioreactor
Turbo column photobioreactor	(1) Turbo column photobioreactor
Sedimentation and flotation	(1) Sedimentation, (1) Flotation
Sedimentation and filtration	(1) Sedimentation, (1) Filtration
Centrifugation	(1) Centrifugation
Filter press	(1) Filtration
Dryer	(1) Dryer
Bead beating	(1) Bead beating
high-pressure homogenization	(1) high-pressure homogenization
Microwaving	(1) Microwaving
Sonication	(1) Sonication
Organic solvent pigment extraction	(1) Extractor, (1) Distillation column, (1) Heat exchanger
Supercritical carbon dioxide pigment extraction	(1) Supercritical extraction
Hexane based lipid extraction	Extractor, (1) Distillation column, (1) Heat exchanger
n- butanol based lipid extraction	Extractor, (2) Distillation column, (2) Heat exchanger
Supercritical carbon dioxide lipid extraction	Extractor, (1) Distillation column, (1) Heat exchanger

Lipid production	(2) Distillation column, (8) Heat exchanger, (3) Decanter, (14) reactor, (3) vessel, (4) Storage tank
Anaerobic digestion	(1) Anaerobic digestion, (1) adsorption, (1) Centrifugation

Table C-10: Required parameters of Eq. (12) [125,126,131,145,151,152,226,230,231]

Type of equipment	$\left(\frac{EC_j^{ref}}{m_j^{ref}}\right)^{f_j}$	IDX_j^{ref}
Open pond	0.0087	521.9
Flat plate photobioreactor	0.0968	521.9
Bubble column photobioreactor	0.0878	521.9
Tubular column photobioreactor	0.0878	521.9
Sedimentation	0.0785	567.5
Flotation	0.0717	567.5
Filtration	1.5081	381.1
Centrifugation	0.1641	394.1
Flocculation	0.0785	567.5
Dryer	1.7717	539.1
Sonication	0.0011	567.7
Bead beating	0.0318	567.7
High pressure homogenization	0.0002	567.7
Microwave	1.9270	433.2
Extractor	0.0045	500
Distillation Column	0.0249	521.9
Heat Exchanger	0.0088	521.9
Supercritical extraction	1.4125	395.6
Decanters	0.0001	541.7
Reactors	0.0624	394.1
Vessels	0.3765	585.7

Storage Tank	0.2533	521.9
Anaerobic digestion	4.3010	585.7
Adsorption	0.711	567.7

Table C-11: Productivities(g/L/day) of Chlorella Vulgaris, Haematococcus Pluvialis, and Nannochloropsis Spp. in an open pond, flat plate photobioreactor, bubble column photobioreactor, and turbo column photobioreactor [232–240]

	open pond	**flat plate photobioreactor**	**bubble column photobioreactor**	**turbo column photobioreactor**
Chlorella vulgaris	0.022	0.085	0.068	0.085
Haematococcus Pluvialis	0.153	0.67	0.12	0.55
Nannochloropsis Spp.	0.04	0.9	0.04	0.65

Table C-12: Price of different pigment's components[147,241]

pigment's components	**Price ($/kg)**
Astaxanthin	2500-7000
beta-carotene	1500
Lutein/Zeaxanthin	2000-3000
Canthaxanthin	600-5000
Chlorophyll	48.2
Vaucheriaxanthin	2000-3000 (assumed same as Zeaxanthin)

Table C-13: Prices of materials [145,242]

Materials/ utility	**Price(($/t)/ ($/mWh)**
Carbon dioxide	34.99
Sulphate	116.38
Flocculant	2500
Ethanol	780
Hexane	1300
n-butanol	1500
Methanol	400
Methanolic silver nitrate	472300
Sodium hydroxide	540
Hydrochloric acid	200
Organic solvent	789
Electricity	33.1
Heating	29
Cooling	14

Table C-14: Input/output flows of system boundaries of Chlorella vulgaris biorefinery

Stage of biorefinery	Components	Energy (MWh)	Input (t/h)	Input from outside (t/h)	Down stream (t/h)	Waste (t/h)	Product (t/h)
Cultivation	water		22.46	22.46	21.90		
	carbon dioxide		10.38	10.38		6.49	
	Ammonia		1.189	1.189	1.02		
	phosphate		0.17	0.17	0.08		
	Sulphate		0.03	0.03	0.02		
	Algae				2.07		
	oxygen					3.19	
	electricity	0.051					
Harvesting	water		21.90		0.02	21.88	
	Ammonia		1.02			1.02	
	phosphate		0.08			0.08	
	Sulphate		0.02			0.02	
	Algae		2.07		1.85	0.24	
	electricity	1.57					
Dewatering	water		0.02		0.002	0.02	
	algae		1.85		1.76	0.09	
	flocculant		5.55 E-06	5.55E-06		5.55E-06	
	electricity						

Cell disruption	water		0.002		0.002		
	algae		1.76				
	Pigment				0.04		
	lipid				0.21		
	others				1.51		
	electricity	64.472					
Pigment extraction	water		0.002			0.002	
	Pigment		0.04		0.015		0.03
	lipid		0.21		0.21		
	others		1.50		1.51		
	organic solvent			0.70		0.70	
	heating	12.21					
	cooling	0.48					
Lipid extraction	Pigment		0.014		0.015		
	lipid		0.21				
	others		1.51		1.51		
	n- butanol			0.21		0.21	
	heating	8.35					
	cooling	18.12					
Lipid production	lipid		0.21				
	Hexane			6.33E-06		6.33E-06	

					0.006	1.58E-02
	Glycerol		0.004			
	Methanol		0.021		0.002	
	methanolic silver nitrate		1.16E-07		1.16E-07	
	sodium hydroxide		6.49E-04		4.17E-05	
	chloric acid		5.92E-04		4.42E-05	
	water				0.002	
	Solid-NaCl				0.001	
	omega-3					0.006
	biodiesel					0.195
	electricity	0.002				
	heating	0.037				
	cooling	0.038				
	pigment	0.015				
	others	1.50				
Remnant treatment	carbon dioxide			0.39		0.04
	Ammonia			0.013		
	phosphate			0.002		
	biogas			0.003		0.30
	water			0.002		0.0002

salt		0.53	
fertilizer			0.23
electricity	21.148		
heating	0.158		

Table C-15: Input/output flows of system boundaries of Haematococcus Pluvialis biorefinery

Stage of biorefinery	Components	Energy (MWh/t)	Input (t/h)	Input from outside (t/h)	Down stream (t/h)	Waste (t/h)	Product (t/h)
Cultivation	water		22.46	22.46	21.54		
	carbon dioxide		20.75	20.75		12.97	
	Ammonia		1.19	1.19	0.8		
	phosphate		0.17	0.17	0.08		
	Sulphate		0.31	0.31	0.19		
	Algae				3.86		
	oxygen					6.36	
	electricity	0.07					
Harvesting	water		21.54		0.02	21.51	
	Ammonia		0.80			0.8	
	phosphate		0.089			0.08	
	Sulphate		0.19			0.19	
	Algae		3.85		3.44	0.45	
	electricity	1.65					
Dewatering	water		0.02		0.002	0.02	
	algae		3.44		3.28	0.17	
	flocculant			1.03E-05		1.03E-05	
	electricity						
	water		0.002		0.002		

Cell disruption	algae		3.28			
	Pigment			0.10		
	lipid			0.50		
	others			2.68		
	electricity	120.16				
Pigment extraction	water		0.002		0.002	
	Pigment		0.104	0.03		0.07
	lipid		0.49	0.50		
	others		2.68	2.68		
	organic solvent		1.63		1.63	
	heating	24.41				
	cooling	0.96				
Lipid extraction	Pigment		0.034	0.03		
	lipid		0.49			
	others		2.67	2.68		
	n- butanol		0.49		0.49	
	heating	15.91				
	cooling	34.54				
lipid production	lipid		0.49			
	Hexane		1.47E-05		1.47E-05	
	Glycerol		0.009		0.015	3.90E-02
	Methanol		0.06		0.002	

	methanolic silver nitrate		2.86E-07	2.86E-07	
	sodium hydroxide		4.84E-06	1.84E-07	
	chloric acid		4.42E-06	1.68E-07	
	water			0.004579	
	solid - NaCl-			6.81E-06	
	omega 3				3.52E-05
	biodiesel				0.47
	electricity	0.006			
	heating	0.08			
	cooling	0.09			
	pigment		0.015		
	others		1.51		
Remnant treatment	carbon dioxide			0.71	
	Ammonia			0.02	
	phosphate			0.003	
	biogas			0.005	0.61
	water			0.005	
	salt			0.94	
	fertilizer				0.42

	electricity	37.84
	heating	0.27

Table C-16: Input/output flows of system boundaries of Nannochloropsis Spp. biorefinery

Stage of biorefinery	Components	Energy (MWh/t)	Input (t/h)	Input from outside (t/h)	Down stream (t/h)	Waste (t/h)	Product (t/h)
Cultivation	water		22.46	22.46	18.82		
	carbon dioxide		30.53	30.53		22.21	
	Ammonia		1.19	1.19	0.59		
	phosphate		0.17	0.17	0.084		
	Sulphate		0.62	0.62	0.44		
	Algae				7.28		
	oxygen					10.44	
	electricity	0.09					
Harvesting	water		18.82		0.021	18.80	
	Ammonia		0.60			0.60	
	phosphate		0.08			0.08	
	Sulphate		0.44			0.45	
	Algae		7.29		6.51	0.85	
	electricity	1.71					
Dewatering	water		0.02		0.002	0.02	
	algae		6.51		6.18	0.32	
	flocculant			1.95E-05		1.95E-05	
	electricity						

Cell disruption	water		0.002		0.002		
	algae		6.18				
	Pigment				0.01		
	lipid				1.14		
	others				5.04		
	electricity	226.77					
Pigment extraction	water		0.002			0.002	
	Pigment		0.01		0.003		0.006
	lipid		1.14		1.135		
	others		5.04		5.04		
	organic solvent			0.15		0.15	
	heating	31.51					
	cooling	1.24					
Lipid extraction	Pigment		0.003		0.003		
	lipid		1.14				
	others		5.04		5.04		
	n- butanol			1.14		1.14	
	heating	31.47					
	cooling	68.32					
lipid production	lipid		1.14				
	Hexane			3.68E-05		3.68E-05	
	Glycerol			0.02		0.035	9.19E-02

	Methanol		0.14	0.005	
	methanolic silver nitrate		6.75E-07	6.75E-07	
	sodium hydroxide		5.71E-06	2.17-05	
	chloric acid		5.21E-06	1.98E-05	
	water			0.01	
	solid (NaCl)			8.03-04	
	omega 3				4.16E-03
	biodiesel				1.07
	electricity	0.01			
	heating	0.2			
	cooling	0.2			
	pigment		0.003		
	others		5.04		
Remnant treatment	carbon dioxide			1.32	
	Ammonia			0.05	
	phosphate			0.006	
	biogas			0.01	1.15
	water			0.001	
	salt			1.77	

	fertilizer		0.73
	electricity	70.24	
	heating	0.5	

List of publications

Journal papers:

- M. Raeisi, E. Zondervan “The role of bioprocess systems engineering in extracting chemicals and energy from microalgae", Phys. Sci. Rev. (https://doi.org/10.1515/psr-2020-0059)

- M. Raeisi, T.A. Huynh, M. B. Franke, & E. Zondervan, E. “Sustainable process technology to extract biochemicals from microalgae: A mini-review". Chem. Scary. Trans. (https://doi.org/10.3303/CET2188148)

- M. Raeisi, M. B. Franke, E. Zondervan “valuable bioproducts from microalgae - A superstructure optimization approach ", Alga research (submitted)

- M. Raeisi, M. B. Franke, E. Zondervan “Life cycle assessment study of microalgae biorefinery ", J. Food Engineering (in preparation)

Conference papers:

- M. Raeisi, j. Huang, T.A. Huynh, M Franke, E. Zondervan “Optimal design of an algae biorefinery for the production of added value products", PSE 2021+, Kyoto, Japan, 19-23 June, 2022

- M. Raeisi, T.A. Huynh, M Franke, E. Zondervan “Superstructure optimization for sustainable design of an algae biorefinery ", ESCAPE32, Toulouse, France, 12-15 June, 2022

- M. Raeisi, E. Zondervan “Optimization of microalgae biorefinery design and operation", CAPE Forum 2020, Kgs. Lyngby, Denmark, 7-9 October 2020

- M. Raeisi, M. B. Franke, E. Zondervan “Uncertainty analysis in the sustainable design of microalgal biorefineries ", CAPE Forum 2022, Enschede, Netherlands 14-16 September, 2022

Collaborations:

- T.A. Huynh, M. Raeisi, M. B. Franke, & E. Zondervan “Novel dynamic cleaning model for cyclic operation of biodiesel membrane reactors". Chem. Scary. Trans. (https://doi.org/10.3303/CET2188147)

- T.A. Huynh, V. Reurslag, M. Raeisi, M Franke, E. Zondervan “Superstructure optimization of biodiesel production from continuous stirred tank and membrane reactors", PSE 2021+, Kyoto, Japan, 19-23 June, 2022

- T.A. Huynh, M. Rossi, M. Raeisi, M Franke, E. Zondervan “promising future for biodiesel: superstructure optimization from feed to fuel ", ESCAPE32, Toulouse, France, 12-15 June, 2022

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