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ENGINEERING & THE ENVIRONMENT

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Energy Balance and Techno-economic Assessment of Algal Biofuel Production Systems

by

John James Milledge BSc MPhil

Thesis for the degree of Doctor of Philosophy

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Abstract

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ENERGY BALANCE AND TECHNO-ECONOMIC ASSESSMENT OF ALGAL BIOFUEL PRODUCTION SYSTEMS

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There has been considerable discussion in recent years about the potential of micro-algae for the production of sustainable and renewable biofuels. Unfortunately the scientific studies are accompanied by a multitude of semi-technical and commercial literature in which the claims made are difficult to substantiate or validate on the basis of theoretical considerations.

To determine whether biofuel from micro-algae is a viable source of renewable energy three questions must be answered:

a. How much energy can be produced by the micro-algae?
b. How much energy is used in the production of micro-algae?
c. Is more energy produced than used?

A simple approach has been developed that allows calculation of maximum theoretical dry algal biomass and oil yields which can be used to counter some of the extreme yield values suggested in the 'grey' literature.

No ready-made platform was found that was capable of producing an energy balance model for micro-algal biofuel. A mechanistic energy balance model was successfully developed for the production of biogas from the anaerobic digestion of micro-algal biomass from raceways. Preliminary calculations had suggested this was the most promising approach. The energy balance model was used to consider the energetic viability of a number of production scenarios, and to identify the most critical parameters affecting net energy production. These were:

a. Favourable climatic conditions. The production of micro-algal biofuel in UK would be energetically challenging at best.
b. Achievement of 'reasonable yields' equivalent to ~3 % photosynthetic efficiency (25 g m^{-2} day^{-1})
c. Low or no cost and embodied energy sources of CO_2 and nutrients from flue gas and wastewater
d. Mesophilic rather than thermophilic digestion
e. Adequate conversion of the organic carbon to biogas (≥ 60 %)
f. A low dose and low embodied energy organic flocculant that is readily digested, or micro-algal communities that settle readily
g. Additional concentration after flocculation or sedimentation
h. Exploitation of the heat produced from parasitic combustion of micro-algal biogas in CHP units
i. Minimisation of pumping of dilute micro-algal suspension

It was concluded that the production of only biodiesel from micro-algae is not economically or energetically viable using current commercial technology, however, the production of micro-algal biogas is energetically viable, but is dependent on the exploitation of the heat generated by the combustion of biogas in combined heat and power units to show a positive balance.

Two novel concepts are briefly examined and proposed for further research:
a. The co-production of *Dunaliella* in open pan salt pans.
b. A 'Horizontal biorefinery' where micro-algae species and useful products vary with salt concentration driven by solar evaporation.

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Declaration of authorship

I, John James Milledge, declare that the thesis entitled;

ENERGY BALANCE AND TECHNO-ECONOMIC ASSESSMENT OF ALGAL BIOFUEL PRODUCTION SYSTEMS

and the work presented in the thesis is both my own, and has been generated by me as the result of my own original research. I confirm that:

- this work was done wholly or mainly while in candidature for a research degree at this University;
- where any part of this thesis has previously been submitted for a degree or any other qualification at this University or any other institution, this has been clearly stated;
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- I have acknowledged all main sources of help;
- where the thesis is based on work done by myself jointly with others, I have made clear exactly what was done by others and what I have contributed myself;
- parts of this work have been published (see Appendix 2)

Signed: ....

Date:..

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Abbreviations and nomenclature

Abbreviation	Description
AD	Anaerobic digestion
ATAD	Auto-thermal thermophilic aerated digestion
BOD	Biological oxygen demand
CCS	Carbon capture and storage
CHP	Combined heat and power
COD	Chemical oxygen demand
DAF	Dissolved air flotation
DO	Dissolved oxygen
DW	Dry weight
EROEI	Energy return on energy investment
EROI	Energy return on energy investment
EROOI	Energy return on operational energy investment
GHG	Greenhouse gas
HHV	Higher heating value
HPDE	High density polyethylene
HRAP	High rate algal pond (raceway)
LCA	Life cycle assessment
LHV	Lower heating value
MFC	Microbial fuel cell
NREL	US National Renewable Energy Laboratory
PAR	Photosynthetically active radiation
PBR	Photo-bioreactor
PE	Photosynthetic efficiency
PVC	Polyvinyl chloride
ROC	UK Renewable fuel obligation certificate
SCWG	Supercritical water gasification
TS	Total solids
VS	Volatile solids

English letters used in equations

Symbol	Description	Unit
A	Area	M^2
C_c	Concentration dry weight of algae per unit volume	$Mg\ l^{-1}$
C_f	Concentration factor	
C_o	Concentration of the gas in the fluid	$kg\ m^{-3}$
C_p	Specific heat at constant pressure	$kJ\ kg^{-1}\ K^{-1}$
C_s	Saturation concentration of the gas	$kg\ m^{-3}$
d	Depth	m
d_p	Light penetration depth	cm
D	Diameter	m
D_c	Characteristic dimension	m
D_w	Dewatering energy	$kW\ m^{-3}$
e	Absolute pipe roughness	m
E	Energy per unit of volume	$kW\ m^{-3}$
f	Friction factor (Strickler)	
f_D	Friction factor (Darcy-Weisbach)	
g	Acceleration due to gravity (9.81)	ms^{-2}
G	Root mean square velocity gradient	s^{-1}
h_b	Head loss in a bend	m
h_g	Head loss in a sump	m
h_p	Head loss in a pipe	m
h_s	Head loss in a straight	m
H	Heat requirement	$KJ\ s^{-1}$
H_l	Heat loss	$KJ\ s^{-1}$
H_t	Total heat requirement	$KJ\ s^{-1}$
k	Head loss factor	
K_{gt}	Gas transfer constant	s^{-1}
L	Length	m
L_s	Length raceway straight	m
M	Mass flow	$kg\ s^{-1}$

n	Manning Friction Factor	
N	Power law exponent	
p_{in}	Pressure in	Pa
p_{out}	Pressure out	Pa
P_h	Hydraulic Power	w
q_{o2}	rate of oxygen production from algal grow	kg m^{-3} s^{-1}
Q	Volumetric flow rate	m^3 s^{-1}
r_c	Cell radius	m
R_e	Reynolds Number	
R_h	Hydraulic radius	m
T	Temperature	° K
T_{in}	Temperature in	° K
T_{out}	Temperature out	° K
U	Heat transfer coefficient	W m^{-2} K^{-1}
v	Average fluid velocity	ms^{-1}
v_l	Local fluid velocity	ms^{-1}
v_{max}	Maximum fluid velocity	ms^{-1}
w	Width	m
y	Distance from the bottom normal to the flow	m
z	Ratio of specific heat at constant pressure to specific heat at constant volume	

Greek symbols used in equations

Symbol	Description	Unit
ε	energy dissipation per unit mass	W kg^{-1}
λ	micro-eddy length	m
μ	Viscosity	Pa s
π	Pi	
ρ	Density	kg m^{-3}
ρ_l	Density of solid	kg m^{-3}
ρ_s	Density of liquid	kg m^{-3}

1. Introduction

1.1 Fossil fuels and global warming

Fossil fuel has been produced over ‘geological time’, primarily by photosynthesis using solar energy, and has powered an industrial revolution: but reserves are finite. Liquid fossil fuel has allowed a transport and organic chemical industry revolution, but estimated reserves are being rapidly depleted. BP has estimated proven world oil reserves in 2010 as sufficient to meet 46 years of current global production. Demand is increasing due to industrial growth and development particularly in the Asia Pacific region; and increasing demand and limited supply are putting upward pressure on oil prices: Brent crude oil prices increased 29 % from 2009 to 2010 (BP, 2011).

Limited supply together with increasing demand and prices will normally drive a search for alternatives in any market, but the search for alternatives to energy provided by fossil fuel is also being driven by concerns over climate change and continuity of supply due to ‘political instability’. The overwhelming majority of scientists agree that the world climate is changing, with average global temperature increasing; and that this is due to rising concentrations of heat-trapping gases in the atmosphere caused by human activities. Carbon dioxide present in the atmosphere is transparent to incoming short wave radiation, but absorbs outgoing infra-red radiation. As the concentrations of carbon dioxide and other gases (methane, nitrous oxide, ozone and certain chlorofluorocarbons) increase it is believed that heat radiating into space will be reduced, resulting in the average world temperature rising - a phenomenon known as Global Warming. The effect of carbon dioxide and other gases, produced by human activity on global temperature is often termed the Greenhouse Effect with the gases involved being known as Greenhouse Gases (Cannell and Hooper, 1990). Concern about global warming has resulted in a variety of legislation throughout the world to reduce greenhouse gas (GHG) emissions resulting in many cases in a cost for GHG production and a market to ‘trade’ emissions of GHG. In 2011one tonne of carbon dioxide was trading, at €13 to €16 on the European Energy Exchange.

In addition to warming due to greenhouse gases, concerns have recently been expressed on 'waste heat warming' of the Earth. It has been concluded that the main type of "energy that is not going to additionally heat the Earth is solar and its derivatives" (Ananthaswamy, 2012).

Most of the world production of liquid fossil fuel is not in the developed democratic countries and there are considerable concerns, particular in the USA and the UK, about the supply of imported oil and the risk of political instability and price. The UK government has recently published a report "to highlight specific areas of concern in the security of the UK's energy supply" (House of Commons Energy and Climate Change Committee, 2011). In 2007 the Energy Independence and Security Act (EISA) was introduced in the United States to increase energy independence and security by increasing the efficiency of products, buildings, and vehicles; promoting research on and deploying greenhouse gas capture and storage options; and increasing the production of clean renewable fuels (US Congress, 2007). In the EU the two main objectives of energy policy are, to reduce greenhouse gas emissions and increase the security of energy supply (European Commission, 2011) .

Reserves of solid fossil fuel in the form of coal are considerably greater than of liquid fossil fuel and many developed democratic nations, including the USA and UK, have considerable reserves (BP, 2011, DECC, 2011, World Energy Council, 2010). Coal could have an important continuing place in the fuel mix and particularly in the production of electricity and the political security of energy supply (DECC, 2010). Jerry Costello, a US Democratic House Representative and member of the sub-committee on energy and the environment believes that: "coal is absolutely critical to national (USA) economic health and global competiveness".

Mary Harris Jones (1830 to 1930) an American labour rights activist, sometimes called "the most dangerous woman in America", said; "not all the coal that is dug warms the world". Although this statement was used in another context it could be apt for the debate about the continued and future use of coal and its effect on global warming. Coal, when burnt, will increase the concentration of GHG in the atmosphere, and some form of carbon capture or storage will be essential for the future exploitation of coal or "Carbon capture and storage – sine qua non" (DECC, 2010). A variety of carbon capture and storage (CCS) technologies have been suggested including, amine stripping, chilled ammonia and oxy-combustion (Chung et al., 2011), but it has been concluded that although technically possible generation of electricity from coal using CCS could be up to 130 % more expensive and significant cost reductions need to be made (Kadam, 2002). Amine stripping is the closest method to commercial application, but the potential for lowering cost is limited, and although both chilled ammonia and oxy-combustion may have greater scope for cost reduction there is considerable uncertainty concerning scale-up (Chung et al., 2011).

1.2 Renewable and bioenergy

Concern over global warming, dwindling fossil fuel reserves, increasing fuel prices and fuel security has prompted research into the more efficient use of fuel together with investigations of alternative and renewable energy sources. Renewable energy normally includes sources such as solar, hydroelectric, tidal, wave, wind and biofuels from biomass (DECC, 2011), but is more narrowly defined by the Energy Independence and Security Act of 2007 (EISA) as "fuel that is produced from renewable biomass and that is used to replace or reduce the quantity of fossil fuel" (US Congress, 2007). Biomass may be defined as; renewable and recently produced organic materials, such as wood, agricultural crops or wastes, and municipal wastes from recent sources that can be burned directly or processed into biofuels such as ethanol, biodiesel and methane. The term bioenergy is used to cover both energy generated from biomass and from biofuels derived from biomass (Twidell and Weir, 2006). In the UK over 80 % of renewable energy usage, excluding solar, is from biomass (DECC, 2011) and in the EU two thirds of total renewable energy is from biomass (European Commission, 2011).

Humanity has been using bioenergy for millennia since the discovery of how to start fire (Figure 1) and biomass was the most important energy supply until the industrial revolution, but as fossil energy use grew biomass usage became "almost non-existent in the industrialised nations" (Quaschning, 2010). Bioenergy is significant worldwide, accounting for 13 % of the world's energy consumption (Twidell and Weir, 2006, World Energy Council, 2010) and is of vital importance in many non-industrialised nations with Mozambique, Ethiopia and Nepal using biomass for 90 % of their primary energy needs (Quaschning, 2010, Sims, 2002).

Figure 1 Firewood; seasoned wood for domestic use in Kent, England

Although the contribution of bioenergy is low in most industrial countries, e.g. 2 to 3 % of the total energy demand in the UK, USA and Germany, its use is growing and can be significant in sugar producing nations, accounting for 40 % of their energy supply (Quaschning, 2010, Twidell and Weir, 2006). Bioethanol and biodiesel production have increased every year from 2000 to 2010 with a 16 % increase in 2010 to an output equivalent to 59 million tonnes of crude fossil oil (BP, 2011). In 2009, biofuels made up 2.9 % of total petrol and diesel sales in the UK, with the majority of this being accounted for by biodiesel. Biodiesel for use in diesel vehicles accounted for 77 % of total biofuels (1 billion litres, 4 % of diesel sales), but only 9 % of biodiesel in the UK market was produced using domestic feedstocks (Committee on Climate Change, 2011). Europe is currently the main market for biodiesel; Figure 2 shows the importance of the European market in world biodiesel trade. The EU Renewable Energy Directive (RED) set targets for biofuel energy content in transport fuels as follows: 2005 – 2 %, 2010 - 5.75 %, 2020 – 10 %, but only Sweden and Germany met the 2005 target (Lemon, 2008). The UK met neither the 2005 or 2010 targets. Although fuels suppliers are responsible for delivering the EU Fuel Quality Directive of a 10 % reduction in average life-cycle emissions of fuels between 2010 and 2020, the EU and Member States spent approximately €3.1 billion on biofuel support in 2010 (House of Commons Energy and Climate Change Committee, 2011). It would appear that demand for biodiesel in EU will continue to grow, but there are reservations in UK. The UK budget of 2008 removed the reduced fuel duty for biodiesel, although secondary legislation was introduced to maintain the 20 pence per litre duty differential for biodiesel produced only from waste cooking oil for a period of two years. The UK Coalition Government has concerns about the sustainability of biofuels: it does not plan to increase biofuel targets before 2014 and it is reviewing biofuel policy, but targets are likely to increase from 2014 to 2020 to meet European targets (Bennett, 2011).

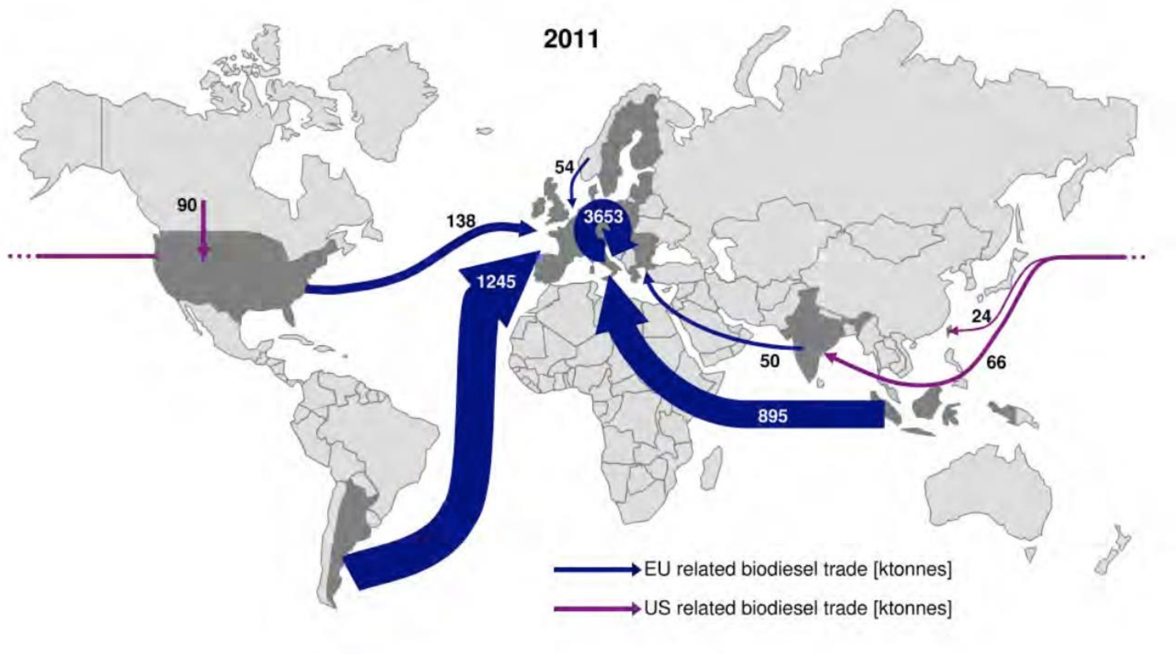


Figure 2 Global biodiesel trade streams in 2010 [ktonne](Ecofys, 2011).

Sustainability has been defined as; "Meeting the needs of the present generation without compromising the ability of future generations to meet their needs" (World Commission on Environment and Development, 1987). If biofuel production is to continue to increase and be considered renewable the biomass must be sustainable with the growth of material as great or greater than its use. There have been numerous examples in agriculture and forestry of overuse of a resource with serious environmental consequences and bioenergy should not accelerate the loss of scarce resources in attempting to reduce greenhouse gas emissions and resolve fuel scarcity, price and security issues.

Bioenergy was initially seen as an ‘easy’ solution to energy and environmental issues as the organic carbon within the biofuel is produced via photosynthesis from carbon dioxide in the atmosphere. When biofuel is burnt the carbon dioxide is returned or recycled to the atmosphere. If the amount of carbon dioxide used in the production of bioenergy is equal to the amount released into the atmosphere there would be no increase or decrease in the concentration of carbon dioxide in the atmosphere, a situation often called carbon neutral. Unfortunately, it is now understood that the effect of the production and use of biofuels is more complex than this simple analysis. Biomass growth requires not only solar energy, but energy for planting, harvesting, processing, transport and production of fertilisers. These energy inputs and the resultant greenhouse gas emissions can be considerable, and in some cases the energy input may be greater than the energy in the fuel. Energy return on energy investment (EROEI or EROI) is the ratio of the energy produced compared to the amount of energy invested in its production. This ‘simple’ ratio can be useful in assessing the viability of fuels. A ratio of less than one indicates that more energy is used than produced, and an EROI of 3 has been suggested as the minimum that is sustainable (Clarens et al., 2011a). EROI for crude oil is currently about 20, but has declined over time. This decline is likely to continue as crude oil is extracted from more difficult locations, with oil extracted from Canadian oil shale having an EROI of 3 (Beal, 2011, Hall and Klitgaard, 2012). The best practice in crops is an EROI between 11 and 16, but biofuels produced from biomass are often considerably lower with sugar ethanol having a reported EROI between 1.25 to 8 and corn ethanol between 1 and 1.34 (Beal, 2011, Clarens et al., 2011a, Hall and Klitgaard, 2012, Mulder and Hagens, 2008, Twidell and Weir, 2006). The EROI can also be a useful indicator of economic viability and the emissions of a fuel. In general the lower the EROI the higher the operational cost of producing and processing the fuel. If a renewable fuel has an EROI of one and all the energy is supplied by fossil fuel it would have greenhouse gas emissions roughly equivalent to the fossil fuel for its production. Care should be taken in interpreting EROI as the boundaries used for energy inputs and outputs may vary and incorporating all the energy values for non- energy co-products and inputs, although the fullest measure, can also be the most imprecise. EROI also assumes that all energy is fungible (freely exchangeable and replaceable) and ignores the 'quality' (cost and usefulness) of the energy inputs and outputs (Mulder and Hagens, 2008). However, "energy balance EROI or net energy gain is central to any evaluation of biofuels" (Walker, 2010).

Biofuels are often classified by the origin of the biomass from which they are derived. First generation biofuels, also termed conventional biofuels, are made from sugar, starch or vegetable oil contained in part of a plant with the remainder of the plant becoming a non-fuel fuel product or waste. Examples of first generation biofuels are; bioethanol produced from sugarcane and corn; and biodiesel from rapeseed and palm oil. This type of biofuel is in direct competition with food production, as it uses crops and parts of crops that are also used for both human and animal food. Second generation biofuels are made from the whole of plant or 'waste' biomass and do not necessarily compete with food production as they can use a crop or part of a crop not used for food. Matter that is not digested by humans such as, cellulose, hemi-cellulose or lignin can be processed into second generation biofuels such as cellulosic bioethanol. Third generation biofuels are not dependent on conventional agricultural or forestry and although production systems, such as algal cultivation systems, are currently under development no large-scale commercial facilities yet exist. The term 'fourth generation biofuels' has appeared recently and can have a variety of definitions. It may refer to biofuels that are not easily classified as first, second or third generation biofuels, but the vast majority of definitions refer to genetically modified organisms. A definition of fourth generation biofuels might be the direct production of readily useable fuels from carbon negative processes involving genetic modified organism: examples could be the direct 'excretion' of hydrogen and alkanes from the growth of genetically modified microorganisms. Fourth generation biofuels are not only the least well defined, but would also appear be at the most fundamental stage of research.

1.3 Algae and algal biofuels

The demand for renewable biofuels is predicted to continue to grow, but the use of first generation biofuels, derived from food crops such as soya and sugarcane, to meet this demand is controversial due to their influence on world food markets and competition for agricultural land. The Gallagher Review, a major study commissioned by the UK Labour Government on the impacts of biofuels on carbon emissions from land use change and on food security, concluded; the EU targets for biofuels for transport fuel is unlikely to be met sustainably from first generation biofuels (Renewable Fuels Agency, 2008). The UK Department of Energy puts current sources of biomass at the bottom of the list of desirable renewable energy sources (Walker, 2010). Third-generation biofuels are proposed as a possible solution and are currently receiving a great deal of attention as they can be cultivated on marginal or non-agricultural land, can use brackish or salt water and may have productivities greater than first or second generation biofuels.

Algae are a diverse range of aquatic 'plants', ranging from unicellular to multi-cellular forms, and generally possess chlorophyll, but are without true stems and roots. The diverse nature of algae is clearly illustrated by an examination of the 'Tree of Life'(Figure 3) where both plants and animals are shown circled on single branches and the various algal taxa are highlighted in rectangles on numerous branches (Schlarb-Ridley, 2011). It is suggest, however, that all algae may share a common cyanobacteria ancestor (Stephenson et al., 2011). The algae can be divided by size into two groups: macro-algae commonly known as 'seaweed' and micro-algae, microscopic single cell organisms ranging in size from a few micrometres to a few hundred micrometres (μm) (Sheehan et al., 1998). The term micro-algae is often used to include the prokaryotic cyanobacteria (blue green algae), although these are no longer classified as algae, together with the eukaryotic micro-algae such as diatoms and green algae (Mata et al., 2010).

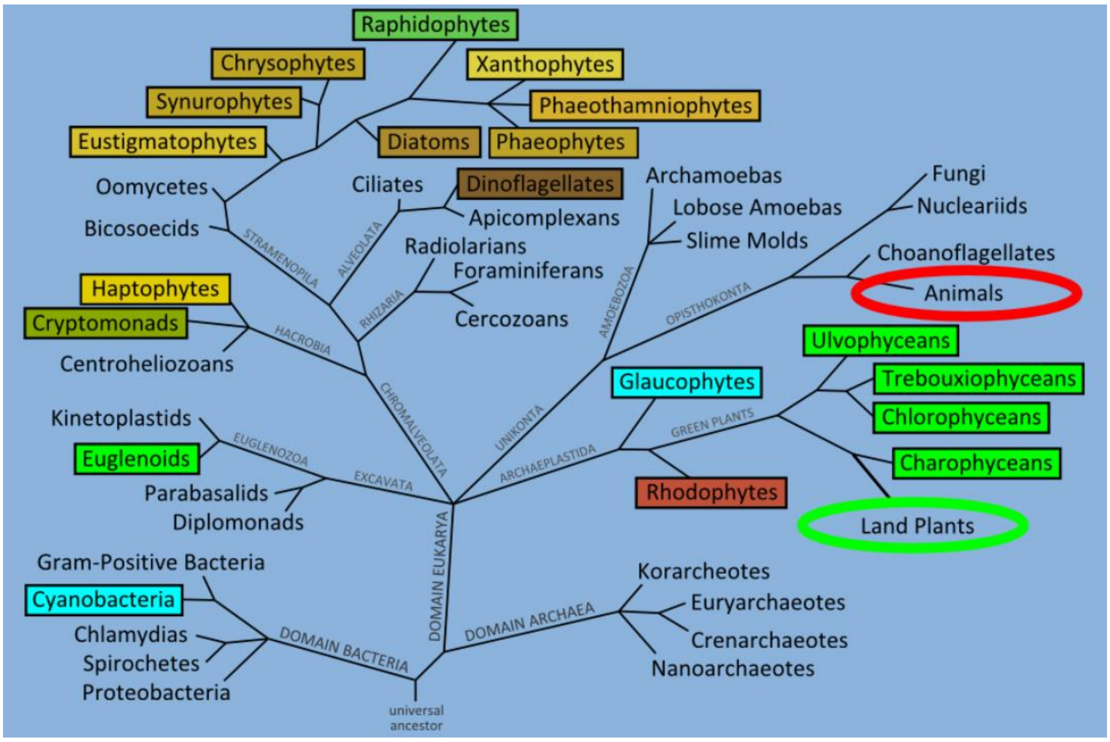


Figure 3 Algae on the 'Tree of Life' (Schlarb-Ridley, 2011)

Macro algae or seaweeds have been used by mankind for generations as a food and for soil conditioning or fertiliser. Seaweeds are used as food by a number of cultures around the world for example: Laver in Wales, Dillisk in Ireland, Dulse in Scotland, Nori in Japan and Limu in Hawaii. The main markets are in Asia. China accounted for over 70 % of the world's macro-algal production in 2006 with aquaculture accounting for 15.1 million wet tonnes of the annual world production and harvest from wild stocks about 1.1 million wet tonnes (Kellly and Dworjanyn, 2008, Roesijadi et al., 2010). The macro-algal industry is currently 100 times bigger in wet tonnage terms than the micro-algal industry (Lundquist et al., 2010).

The high water content (80 to 90 %) of macro-algae has led to a focus on energy production from wet algae due to the relatively high energy demand of evaporating water from the biomass compared to the energy content within it (Ecofys, 2008). Seaweed cultivation for bioethanol and biogas is being explored in Asia, Europe and South America, while bio-butanol from macro-algae is attracting research interest and investment in the USA (Parliamentary Office of Science & Technology, 2011). The use of macro-algae as a commercial feedstock for fuels production is currently 'non-existent' (Roesijadi et al., 2010) and the cost of production is high, with recent estimates suggesting that biogas from seaweed could be 7-15 times more expensive than natural gas (Parliamentary Office of Science & Technology, 2011).

Humans have used micro-algae for thousands of years. The Chinese used *Nostoc* to survive famine and the blue green algae species, *Aphanizomenon*, has also been used (Singh et al., 2005, Spolaore et al., 2006). *Spirulina* has been exploited by ancient peoples in both Chad and Mexico as a source of food (Chisti, 2006, Henrikson, 2008). The use of algae for therapeutic purposes has a long history, but the search for biologically active substances from algae, especially examination for antibiotic activity, began only in the 1950s. Much of the laboratory work up until the 1980s focused on macro-algae (Borowitzka, 1995). Approximately 15,000 natural marine products have now been screened for biological activity and 45 marine-derived natural products have been tested for medical use in preclinical and clinical trials. Only two have been developed into registered drugs, one from a marine snail and the other from a sea squirt, with none as yet from micro-algae (Wijffels, 2007).

Micro-algae are responsible for over 50 % of primary photosynthetic productivity on earth and are sunlight-fuelled factories for a wide range of potentially useful products, but are barely used commercially compared to terrestrial plants and other microorganisms (Chisti, 2000, Chisti, 2006, Gavrilescu and Chisti, 2005, Milledge, 2011a, Wijffels, 2007). In 2004 the global market for micro-algal biomass for non-fuel use was estimated as 5000 tonnes of dry matter per year and generated an annual turnover of US$1250 million (Spolaore et al., 2006). Reviews of the non-fuel uses of micro-algae have been published (Milledge, 2011a, Milledge, 2012b) and some typical examples are:

a. β-carotene, a substance converted by the body to Vitamin A, and used as food supplement and colourant, is produced from *Dunaliella* (Chisti, 2006, Singh et al., 2005).
b. Lina Blue, a blue Phycobiliprotein food colourant, which is used in chewing gum, ice slush, sweets, soft drinks, dairy products and wasabi, is produced from *Spirulina* (DIC, 2008, Raja et al., 2008, Spolaore et al., 2006).

c. Docosahexaenoic acid (DHA), a polyunsaturated omega-3 fatty acid, used as a dietary supplement and supplement in infant formulas, is produced by heterotrophic culture of the dinoflagellate *Crypthecodinium cohnii* (Borowitzka, 2006, Spolaore et al., 2006).
d. Food and feed additives for the commercial rearing of many aquatic animals are produced from a variety of micro-algal species (Borowitzka, 2006).
e. Sulphated polysaccharides, are being produced in Israel for cosmetic products, from *Porphyridium* (Arad and Levy-Ontman, 2010).

The culture of micro-algae is one of the modern biotechnologies, with uni-algal culture (the culture of a single algal species) being first achieved in 1890 with *Chlorella vulgaris* (Borowitzka, 2006). Algae can be photoautotrophic, heterotrophic or mixotrophic. In photoautotrophic growth organic materials are synthesised from inorganic carbon using energy from light. In heterotrophic growth energy is derived from an organic substance.

Heterotrophic growth of micro-algae has been used to convert distillery waste to algal biogas and other high value algal products (Aylott, 2010). Solazyme, in 2010, delivered 80,000 litres of heterotrophic algal-derived marine diesel and jet fuel to the US Navy (Solazyme, 2012), but the cost of over $26 a US gallon was approximately fifteen times the price of crude oil (Kelly, 2011). The major drawback with heterotrophic growth is that it requires an organic carbon source derived from autotrophic growth and the conversion of one form of organic carbon chemical energy to another is not as ‘efficient’ as autotrophic growth (Chisti, 2007). The cost of the carbon source has been found to be over 60 % of the biomass cost of heterotrophically grown Chlorella and energy losses due to respiration and conversion of organic material are significant (Liu et al., 2010).

Mixotrophic algal growth is a combination of phototrophic and heterotrophic growth and may have advantages over purely autotrophic growth in low light and oligotrophic environments (low nutrients and rates of photosynthesis) (Arenovski, 1994).

The use of photoautotrophic algae allows the direct production of biomass for biofuel by exploiting solar radiation and avoids the losses in biomass energy in the conversion between different organisms. There is a vast amount of solar radiation that could be exploited by photoautotrophic organisms. Total solar radiation delivers a total energy input over the entire earth's surface of $2.7x10^{27}$ J (Smil, 1999). This exceeds the total energy demand of humans by three orders of magnitude (Stephenson et al., 2011). Only a small fraction of the total solar radiation (0.1 to 0.5 %)(Packer, 2009) enters into the global biological system, but this is still many times the total anthropogenic energy consumption (Sims, 2002). Energy production from the autotrophic growth of micro-algae is the focus of this research study.

The lipid content of algae can be high at over 70 %, with oil levels of 20 to 50 % being common, but more typically 10 to 30 % when grown under nutrient replete conditions (Campbell et al., 2009, Gavrilescu and Chisti, 2005). This high lipid content led to serious consideration of the large-scale cultivation of micro-algae and use of the biomass for fuel production in Germany during World War II (Becker, 1994, Tamiya, 1957). The oil crisis of the 1970s led to a programme carried out by the US National Renewable Energy Laboratory (NREL), to develop renewable transportation fuels from algae which ran from 1978 to 1996 at a cost of $25.05 million. The overall conclusion of these studies was, that "in principle and practice large-scale micro-algae production is not limited by design, engineering, or net energy considerations and could be economically competitive with other renewable energy sources" (Sheehan et al., 1998).

Current concerns about fuel cost and global warming have resulted in micro-algal biomass cultivation as a potential source of biofuel receiving a great deal of attention. There are now over 50 algal biofuel companies, but none, as yet, are producing commercial-scale quantities at competitive prices and the process of producing fuel from algae would appear to be currently uneconomic (Pienkos and Darzins, 2009, Sills et al., 2012, St John, 2009). Estimates for the reduction in production cost needed for algal biofuel to become economic vary from a reduction of a factor of five up to two orders of magnitude (Bruton et al., 2009, Kovalyova, 2009, Wijffels, 2007).

Nearly 70 years of sometimes intensive research on micro-algae fuels, and over two billion US dollars of private investment since 2000 (Service, 2011) have not produced economically viable commercial-scale quantities of algal fuel and this suggests that there are major technical and engineering difficulties to be resolved before economic algal biofuel production can be achieved.

1.4 Aims and objectives

The overall aim of the research was to develop tools for the assessment of the energy balance of micro-algal biofuel production.

To achieve this, the following objectives were identified:

1) To gather, critically review and assess information from the literature on process options for micro-algal biofuel production and identify key areas in which further information is required in relation to their contribution to the overall energy balance.
2) To determine whether existing software packages are suitable for determination of the energy balance of micro-algal biofuel production.
3) To establish maximum theoretical and 'achievable' micro-algal biomass growth and oil yields as a basis for estimation of the energy output of micro-algal biofuel production processes.
4) To establish 'energy return on operational energy input' as a criterion for assessment of the energy balance of micro-algal biofuel production, using the example of paddlewheel mixing of a raceway cultivation system.
5) To develop a mechanistic model of energy inputs and outputs as a rational basis for the assessment of algal biomass and biofuel production systems.
6) To determine the energetic viability of a number of micro-algal biofuel production scenarios, and to identify the most critical parameters affecting net energy production.
7) To carry out a case study of micro-algal biofuel production including assessment of its energetic and economic viability.

1.5 Structure of thesis

The thesis consists of 8 chapters:

The current chapter introduces key concepts and issues in renewable energy production from micro-algae, and summarises the aims and objectives of the work.

Chapter 2 provides a critical review and analysis of process operations in micro-algal biofuel production, and identifies areas in which further information is required in relation to their contribution to the overall energy balance.

Chapter 3 evaluates the suitability of available software for modelling and assessment of energy balances for micro-algal biofuel production.

Chapter 4 describes initial investigations carried out to provide a baseline for assessment of micro-algal biofuel production by establishing maximum and realistic algal yields and operational energy returns for micro-algal growth in raceways.

Chapter 5 describes the construction of a mechanistic energy balance model for micro-algal biofuel production in open raceway systems.

Chapter 6 presents the results of energy balance modelling for the production of micro-algal biofuel under different scenarios.

Chapter 7 presents a case study on the energetic and economic viability of micro-algal biodiesel production and presents some novel non-fuel micro-algal processes in which energy production takes a secondary role.

Chapter 8 consists of brief conclusions and suggestions for further work.

2. Algal biofuel process operations

This chapter provides a contextual analysis of the processes for micro-algal biofuel production; reviewing, summarising and identifying gaps in the knowledge for each area.

Process operations for algal biofuel production can be grouped into three areas: growth, harvesting and energy extraction. The methods available are summarised in Figure 4, and the success of micro-algal biofuel will be dependent on the achievement of an optimised process for these three areas.

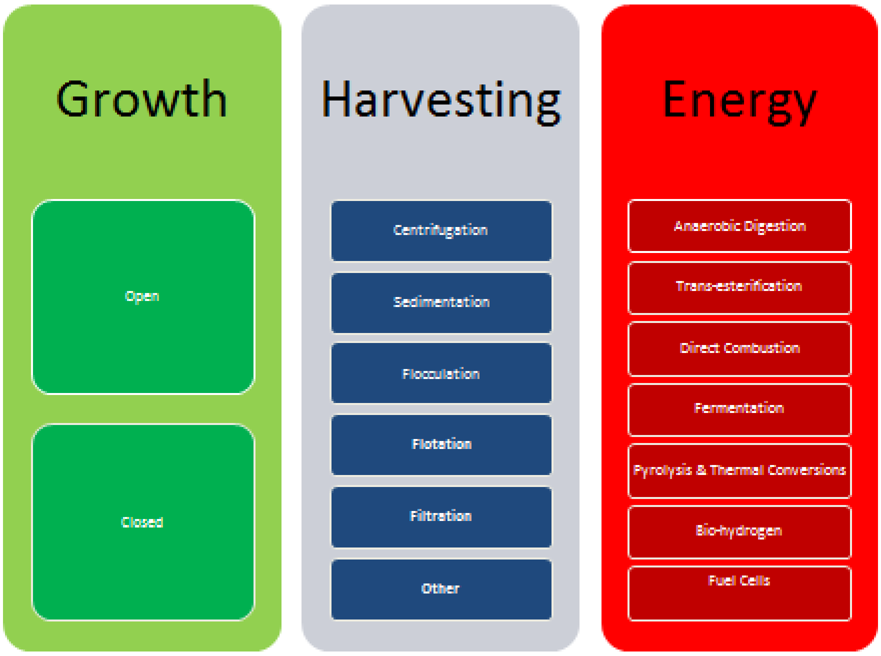


Figure 4 Summary of algal biofuel production options

2.1 Growth systems

Algae can be grown in simple open systems or closed systems known as photo-bioreactors (PBRs) (Becker, 1994, Chisti, 2007). Currently the majority of micro-algal production occurs in outdoor ponds (Spolaore et al., 2006), although at present only three taxa (*Spirulina, Dunaliella* and *Chlorella*) are grown commercially, where the use of highly selective environments make it possible to suppress the growth of competitive species. *Chlorella* grows well in nutrient-rich media, *Spirulina* requires a high pH and bicarbonate concentration and *Dunaliella salina* grows at very high salinity (Huntley and Redalje, 2007).

2.1.1 Open systems

The growth of micro-algae in natural open water bodies has been exploited around the world for centuries, especially in time of famine, and simple unmixed unlined ponds are still used for micro-algal cultivation and wastewater treatment.

Shallow natural or man-made ponds (normally under 0.5 m deep) are the lowest capital cost and least technically complex of all micro-algal mass culture methods (Shen et al., 2009). *Dunaliella salina* is cultivated, commercially in Australia, in very large, shallow unlined and highly saline ponds for the production of β-carotene as shown in Figure 5. This type of culture is suited to areas where land costs are low and climatic conditions favourable, with low rainfall and high solar insolation (Borowitzka, 2005), but productivity is low at only 1.5 tonnes ha^{-1} $year^{-1}$ (0.4 g m^{-2} day^{-1}) (Campbell et al., 2009).

The yields of *Dunaliella* can be increased by mixing the culture in the ponds; work in Spain using paddlewheel-mixed ponds found average daily productivity increased to 1.65 g m^{-2} day^{-1}, four times that of unmixed systems (Garcia-Gonzalez et al., 2003). Agitation or mixing of the culture medium has been found to be one of the most important factors in achieving "consistently high yields of biomass"; ensuring frequent exposure of all algal cells to light, prevention of settlement of algal cells, elimination of thermal stratification, even distribution and improved gaseous transfer (Becker, 1994). Since the 1950s a number of different stirred open systems have been investigated for the production of micro-algae by a number of research groups, but the two most commercially successfully are (Becker, 1994, Shen et al., 2009):

a. Circular ponds with mixing provided by a rotating arm
b. Race-ways shallow ponds, where algal growth medium is circulated around a central rib.

Figure 5 *Dunaliella* growth ponds in Australia. Courtesy Cognis

2.1.1.1 **Algal wastewater stabilisation ponds**

Simple unmixed waste stabilisation ponds were introduced in the USA in the early 1900s as a low cost solution to wastewater treatment for a growing population. They were initially simply used for containment without discharge, rather than being designed and optimised for wastewater remediation (Banks, 2011). In these relatively shallow man-made ponds the organic content of the effluent is converted to bacterial and micro-algal biomass with the symbiotic growth of micro-algae and bacteria reducing odours and pathogenic microorganisms (Buhr and Miller, 1983). Algae provide the oxygen for the growth of the bacteria to breakdown the organic waste matter, and the bacteria in turn provide carbon dioxide for the growth of the algae (Buhr and Miller, 1983, Goldman, 1979a, Green et al., 1995). The ponds are also known as facultative ponds, having an aerobic zone at the top and an anaerobic zone at the bottom as shown in Figure 6.

In wastewater treatment aeration by mechanical means can be the most energy intensive process, with 0.4 to 1.1 kWh required to transfer 1 kg of oxygen (Green et al., 1995). The use of micro-algae for wastewater oxygenation is considered to be the most economical and energy efficient method available (Oswald, 1988).

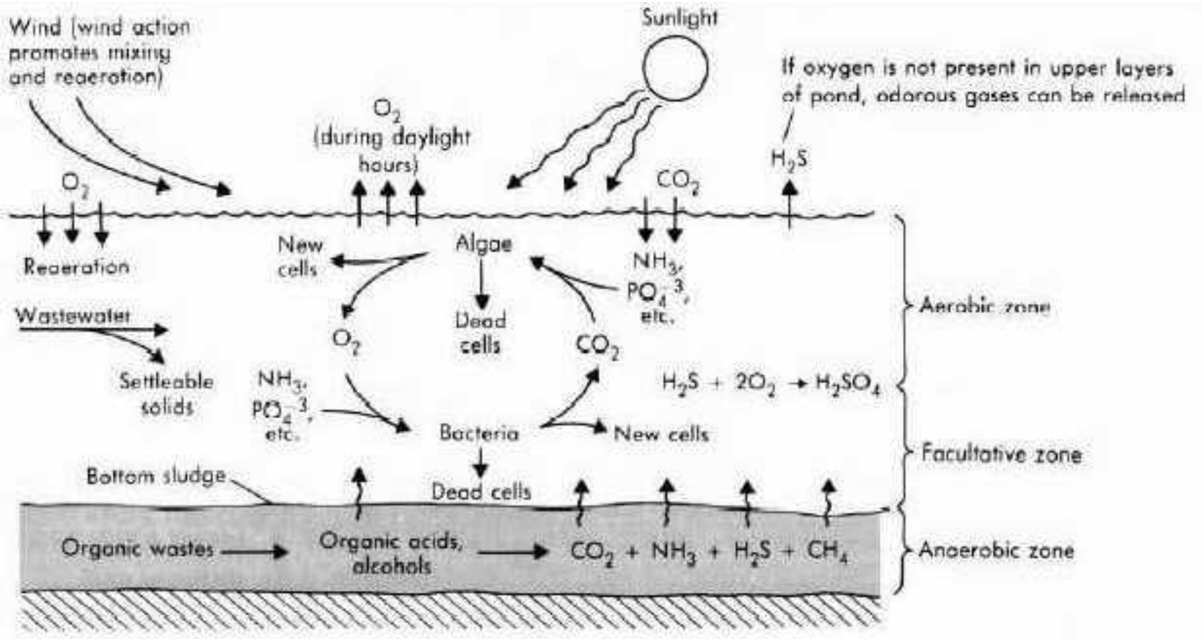


Figure 6 Operation of the facultative pond (Tchobanoglous and Schroeder, 1985)

Algal waste stabilisation ponds gained wide acceptance and by 1980 over 7000 such ponds were in operation in the USA (Water Treatment, 2010). One of the major drawbacks of waste stabilisation ponds is that they require a large area of land (Goldman, 1979a). Favourable climate conditions, however, led to very large pond systems of 60 to 340 hectares being established in California (Benemann et al., 1980).

The growth of bacteria and micro-algae in waste stabilisation ponds can not only reduce dissolved nutrients and Biological Oxygen Demand (BOD), but can also produce significant amounts of micro-algal and bacterial biomass. This biomass can present an environmental threat if it is allowed to flow out of the pond into the surrounding environment, but it can also be an opportunity if harvested as a source of organic material for feed and fuel (Goldman, 1979a).

Algal waste stabilisation ponds (1 to 3 m deep) were designed for optimal waste treatment and not micro-algal production (Benemann et al., 1977). The potential to increase and exploit micro-algal biomass and reduce the land demands of waste stabilisation ponds led to attempts to produce improved wastewater treatment systems involving micro-algae and the development of the High Rate Algal Pond or Raceway (Benemann et al., 1980, Buhr and Miller, 1983).

2.1.1.2 **Circular algal cultivation ponds**

Circular ponds with a centrally pivoted mixing arm have been used in both Taiwan and Japan for the production of *Chlorella* (Becker, 1994, Borowitzka, 2005, Shen et al., 2009). Ponds are normally 0.3 m to 0.7 m deep and up to 45 m in diameter (Shen et al., 2009). The maximum diameter of the ponds is limited to approximately 50 m as mixing efficiency is poor when the rotating arm is too long (Borowitzka, 2005, Shen et al., 2009) and 'mechanical problems' occur with large diameter mixing arms (Becker, 1994). Other disadvantages of circular ponds are; low turbulence and mixing in the central part of the pond; supplying CO_2 to the culture; and high capital costs relative to raceway ponds (Becker, 1994). Although circular ponds are the oldest large-scale mixed algal growth system (Borowitzka, 2005) they are now currently only used to a limited extent and the most common open stirred growth system is the raceway (Becker, 1994, Borowitzka, 2005).

2.1.1.3 **Raceway systems**

Raceways (Figure 7) have become the most common method of growing algae in open systems (Becker, 1994, Ferrell and Sarisky-Reed, 2010, Oswald, 1988, Sheehan et al., 1998). They are suggested as "the most efficient design for large scale culture of micro-algae" (Borowitzka, 2005) and are probably the least expensive option for cultivation of micro-algae in both capital and operating costs (Sills et al., 2012).

A raceway is a shallow closed-loop recirculation channel (James and Boriah, 2010) where algal growth medium is circulated around a central rib. The raceway can be either a single loop (as shown in Figure 7) or may be serpentine, but all raceways may be subdivided into four design areas; straights, bends, gaseous transfer and fluid propulsion areas.

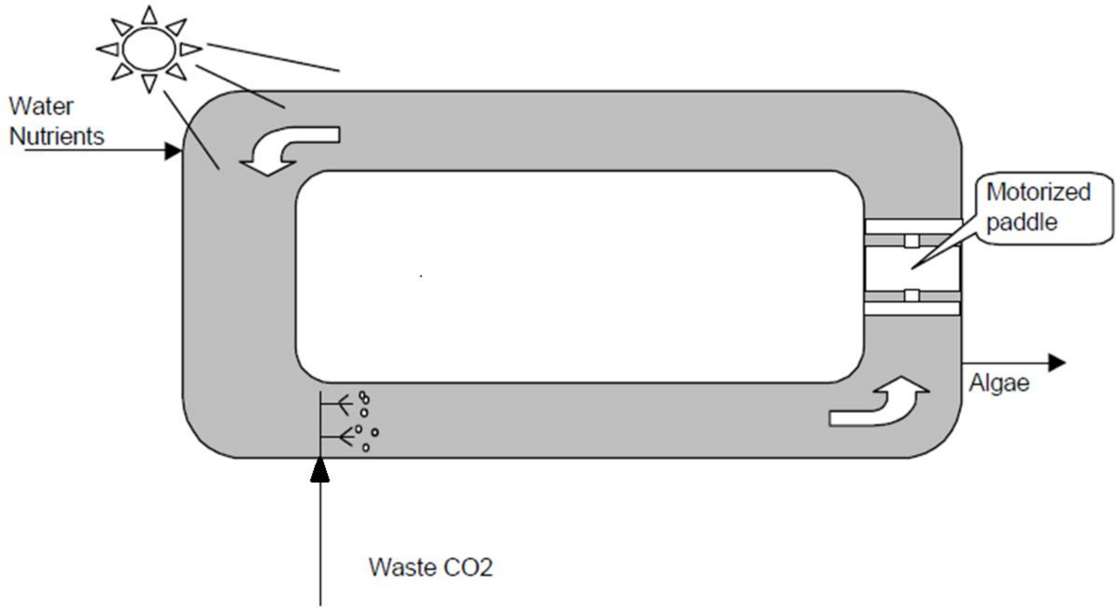


Figure 7 Raceway system (Sheehan et al., 1998)

2.1.2 Closed systems – Photo-bioreactors (PBRs)

Although the term photo-bioreactor (PBR) has been applied to open and closed micro-algal growth systems, it is primarily reserved for closed devices (Molina Grima et al., 1999) and a photo-bioreactor can be defined as a closed system for micro-algae cultivation (Tredici, 2010). In photo-bioreactors the growth medium is not directly exposed to the atmosphere, but is contained within a transparent material (Shen et al., 2009) that allows autotrophic micro-algal growth while isolating the culture from potential contamination (Molina Grima et al., 1999).

Numerous photo-bioreactor configurations have been reported (Acién Fernández et al., 2013, Brennan and Owende, 2010, Tredici, 2012). Most may be classified into one of two types: either tubular devices (Figure 8) or flat panels (Figure 9), with additional categorisation according to orientation of tubes or panels, method of circulation of the culture, mechanism of light provision, type of gas exchange system and materials of construction (Molina Grima et al., 1999).

Figure 8 Tubular Photo-bioreactor at University of Almeria, Cajamar Experimental Station in Southern Spain

Figure 9 Flat Panel Photo-bioreactor at University of Almeria, Cajamar Experimental Station in Southern Spain

2.1.3 **Comparison of open and closed systems**

A comparison of photo-bioreactors and open raceway ponds is given in Table 1. Although, photo-bioreactors can have benefits, for example controlled environment and reduced contamination, their higher capital and operational costs may prohibit their use for the production of biofuel (Benemann, 2008).

The circulation energy in photo-bioreactors has been estimated to be 13 to 28 times that of open raceway ponds for the production of the same mass of micro-algae, although the algal concentration in the PBR was assumed to be five times that in an open system (Stephenson, 2009, Stephenson et al., 2010). It has been argued that lower extraction costs due to the higher algae concentrations, can make bioreactors more competitive (Chisti, 2007), but the claims that photo-bioreactors (PBRs) are many times more productive than open systems are ‘unsupported’ (Lundquist et al., 2010).

Control over the micro-algal species grown is considered easier in a closed system PBR than in open system (Chisti, 2007, Mata et al., 2010), but maintaining a stable single species algal culture at full commercial biofuel production scale would be "exceptionally challenging" due to the large volumes of liquid and gas requiring sterilisation (Smith et al., 2010). The NREL study in the USA concluded that open systems were the only economic solution for large-scale production and a study by Auburn University reached the same conclusion (Putt, 2007, Sheehan et al., 1998). Although some micro-algal products are now being produced commercially in photo-bioreactors, the development of micro-algae biotechnology has been slowed by the limited performance of photo-bioreactors (Spolaore et al., 2006). The use of photo-bioreactors will probably be restricted to very high value products, for example when purity is essential, or the production of inoculum for open ponds (Becker, 1994).

Table 1 Comparison of closed and open micro-algal growth systems (adapted from Mata et al. (2010))

Culture systems for micro-algae	Closed systems (PBRs)	Open systems (Raceway Ponds)
Contamination control	Easy	Difficult
Contamination risk	Reduced	High
Process control	Easy	Difficult
Species control	Easy	Difficult
Mixing	Uniform	Very poor
Area/volume ratio	High (20–200 m−1)	Low (3–10 m−1)
Algal cell density	High	Low
Investment	High	Low
Operation costs	High	Low
Capital/operating costs ponds	Ponds 3–10 times lower cost	PBRs > Ponds
Light utilisation efficiency	High	Poor
Temperature control	More uniform	Difficult
Productivity	3–5x more productive	Low
Hydrodynamic stress on algae	Low–high	Very low
Evaporation of growth medium	Low	High
Gas transfer control	High	Low
O_2 inhibition	Greater problem in PBRs	PBRs > Ponds
Biomass concentration	3–5 times in PBRs	PBRs > Ponds

The cost of producing dry algal biomass in a tubular photo-bioreactor, similar to the type shown in Figure 8, was stated as US$ 32.16 kg^{-1} (Molina-Grima et al., 2003). Some estimates have indicated that closed reactor systems will only be able to compete with crude oil at US$ 800 per barrel (US$ 5 per litre) (Broere, 2008). Solix Biofuels has developed technologies to produce oil derived from algae, but at a cost of about $ 32.81 a gallon (over US$ 8 per litre) (Kanellos, 2009). The production cost of oil from algae, grown in open saline ponds in a project involving Murdoch University in Perth, Western Australia, was reported below US$ 4 kg^{-1} (Lewis, 2009). A review of the potential of marine algae as a source of biofuel in Ireland estimated that the cost of algal biodiesel feedstock produced in open ponds in Israel is over $ 2.80 kg^{-1} (Bruton et al., 2009), both costs considerably below the previous estimates for PBRs. The cost of biomass productions in PBRs would appear to be an order of magnitude greater than in open raceways (Mata et al., 2010).

The mass production of food and protein to solve world food/protein shortages by the use of bioreactors has not materialised and traditional farming is still the means by which the vast majority of food is produced. Using food production as an analogy for the cultivation of micro-algae for biofuel, open systems could be considered as farming and may be the route by which algal biofuels is produced. It will be the intention of this research to concentrate on the energy analysis and proposals for the energy optimisation of open algal growth systems.

2.2 Engineering aspects of raceway systems

2.2.1 Materials of construction

Algal raceway ponds can be excavated or they can be constructed above ground with walls of concrete blocks or similar. Excavated raceways are normally cheaper, but it is more difficult to maintain a constant depth for even flow and there may be higher risks of contamination (Borowitzka, 2005). Lined ponds are approximately twice the cost of unlined ponds (Lundquist et al., 2010), and a number of authors have suggested that this increase in costs will make lined ponds uneconomic for algal fuel production (Lundquist et al., 2010, Putt, 2007, Sheehan et al., 1998). An examination of photographs of open pond production facilities would indicate that liners are almost always used in both commercial and experimental open raceways. Operational problems have been reported with the use of unlined ponds (Ben-Amotz, 2008b). Liners should; result in lower head losses due to the smoother surface; lower seepage into the surrounding soil; be easier to clean; and reduce water clouding problems due to the suspension of soil and clay particles (Borowitzka, 2005). No difference in algal yields was found between lined and unlined raceway ponds (Sheehan et al., 1998).

Liners need to be cheap and UV resistant (Ben-Amotz, 2008b). Possible liner materials are:

a. HPDE – High Density Polyethylene

b. PVC – Polyvinyl Chloride
c. Polypropylene
d. Butyl rubber
e. Geo-synthetic Clay Liners i.e. Bentomat a layer of clay fixed to a membrane

Both PVC and HPDE can inhibit micro-algal growth due to the presence plasticisers, other additives and contaminates, but the use of food grade materials can prevent growth inhibition (Borowitzka, 2005) . HDPE has been used successfully in water and wastewater treatment. It is probably the most popular geo-membrane liner due to its high UV and chemical resistance, flexibility and relatively low cost compared to other liners. HPDE has a relatively high coefficient of thermal expansion which could present problems in areas where there are wide temperature variations and HPDE also requires a high degree of soil compaction as any movement under the liner may cause damage (Butyl Rubber Ltd private communication, 2009). Problems have been encountered with its use for algal raceways in Israel and it was replaced by PVC (Ben-Amotz, 2008b). PVC can last for over 5 years in temperate desert climates (Borowitzka, 2005). It has lasted for over 20 years in algal ponds in Israel (Ben-Amotz, 2008b) and was used as a liner in the extensive studies by NREL (Sheehan et al., 1998). Food grade PVC has also been used in experimental growth ponds with an area of up to 450 m^2 in Spain (Jimenez et al., 2003).

Reinforced polypropylene and butyl rubber are both durable and flexible, but relatively expensive at over twice the price of HDPE (Butyl Rubber Ltd private communication, 2009). Geo-synthetic Clay liners have the advantage that they can self-seal, but are normally more expensive than PVC or HDPE and need to be covered with earth after installation (Butyl Rubber Ltd private communication, 2009). The use of Geo-synthetic Clay liners could have additional costs for excavation and maintaining flow and increased problems of suspended solids in the algal growth media. The advantages and disadvantages of the various liners are summarised in Table 2.

Table 2 Raceway liners advantages & disadvantages

Liner Type	Advantages	Disadvantages
HPDE – High Density Polyethylene	✓ UV Resistant ✓ Durable ✓ Widely used in the water and wastewater treatment	• High coefficient of thermal expansion • Requires high degree of soil compaction • Problems encountered in algal raceway
PVC – Polyvinyl Chloride	✓ Durable ✓ UV Resistant ✓ Successfully used in raceways	• Possible growth inhibition from plasticisers and other additives
Polypropylene	✓ Durable and flexible	• Cost
Butyl rubber	✓ Durable and flexible	• Cost
Geo-synthetic Clay Liners	✓ Ability of clay to seal punctures, and self-seam	• Cost • Suspended matter

2.2.2 Raceway channels

It has been suggested that the main objective of the design of an algal cultivation system is the selection of the flow and depth combination that will result in the optimum biomass concentration and yield (Goldman, 1979b). Optimising the design of the straights involves not only selecting width and length, but also depth of fluid, fluid velocity, material selection, freeboard, side slope and devices for mixing.

2.2.2.1 Mixing and fluid velocity in raceways

Mixing is required to expose all cells to light, distribute nutrients, enhance gaseous transfer and prevent settlement (Oswald, 1988, Terry and Raymond, 1985, Williams and Laurens, 2010). The extent of thermal stratification in water bodies can be an indication of the degree of mixing. Unmixed 0.3 m deep ponds have been shown to have temperature variations of up to 8° C (Oswald, 1988). Velocities above 0.05 m s^{-1} have been shown to prevent thermal stratification and maintain algae in suspension, although it is suggested that an average velocity of 0.15 m s^{-1} is required within a raceway to maintain this velocity in all areas of a raceway (Oswald, 1988). The Arthur D

Little Company showed considerable micro-algal settlement at velocities below 0.075 m s^{-1} in shallow micro-algal growth systems (<0.1m), but above 0.25 m s^{-1} settling was eliminated (Terry and Raymond, 1985). In other open systems, however, significant algal settlement has been shown to occur at velocities of 0.32 m s^{-1} (Weissman et al., 1989). The recommended velocity to maintain silt in suspension is considerably higher at between 0.6 to 0.9 m s^{-1} (2- 3fts^{-1}) (Chanson, 1999), but silt, although of a similar size to micro-algae (2 to 50 μm), is considerably denser (US Department of Agriculture Natural Resources Conservation Service, 2012) and settles out of suspension more readily than micro-algae. It has also been suggested that increased velocity and faster mixing can induce flocculation in micro-algae (Benemann et al., 1980).

Optimum mixing in algal ponds is suggested to occur between 0.2 to 0.3 m s^{-1} (Lundquist et al., 2010), while use of a CFD model showed no increase in micro-algal growth above 0.3 m s^{-1} (James and Boriah, 2010). Typical flow rates in micro-algal growth ponds are normally 0.15 to 0.3 m s^{-1} (Borowitzka, 2005, Chiaramonti et al., 2013). Literature values for fluid velocities in raceway ponds are summarised in Table 3.

Fluid velocity has tended to be used for the description of flow in open raceway systems despite the differences in system dimensions. However, the type of flow that occurs in a channel depends, not only flow velocity, but also on the characteristics of the fluid and dimension of the flow channel (Cheng, 2007, Chow, 1959).

Table 3 Fluid velocities in raceway

Prevention of thermal stratification	0.05 to 0.15 m s^{-1}	(Oswald, 1988)
Prevention of sedimentation of algae	0.25 m s^{-1}	(Terry and Raymond, 1985)
Suggested optimal mixing	0.2 to 0.3 m s^{-1}	(Lundquist et al., 2010)
Fluid velocity above which growth does not increase	0.3 m s^{-1}	(James and Boriah, 2010)
Typical flow rates in algal raceways	0.15 to 0.3 m s^{-1}	(Borowitzka, 2005, Chiaramonti et al., 2013)

2.2.2.2 **Reynolds number and turbulence**

Reynolds number (R_e) has been described as one of the most basic concepts in fluids and the most important dimensionless number (Lowe, 2003): it is the ratio of inertia forces to viscous forces within the fluid and can be expressed as (Coulson and Richardson, 1999);

Equation 1

$$R_e = \frac{\rho v D_c}{\mu}$$

where v is the average fluid velocity, D_c is characteristic dimension, ρ density and μ viscosity. The characteristic dimension used in the calculation of Reynolds Number is different for full flow systems and open systems (Lowe, 2003). Open systems use hydraulic radius (area of stream cross-section divided by wetted perimeter) and closed systems use hydraulic diameter or pipe diameter which is four times hydraulic radius (Lowe, 2003). The hydraulic radius (R_h) is expressed as;

Equation 2

$$R_h = \frac{dw}{w + d}$$

Where d is fluid depth and w is channel width.

Use of the Reynolds number allows the flow to be described as turbulent or laminar, comparison of flow in similar shaped structures of different dimensions, and the scale-up of flowing fluid systems (Coulson and Richardson, 1999, Vennard and Street, 1976). At low Reynolds numbers viscous forces dominate and flow is laminar, characterised by smooth motion of the fluid in layers, without significant mixing between layers. Turbulent flow occurs when inertial forces dominate, producing circulating currents or eddies, and there is exchange of momentum across the primary direction of fluid flow (Coulson and Richardson, 1999). Flow is said to be turbulent in open channels; above a Reynolds Number 1000, and 4000 for full flow in closed systems; flow is laminar below 500 for open systems, and 2000 for full flow closed systems (Lowe, 2003, Vennard and Street, 1976).

2.2.2.2.1 **Effect of Reynolds number and turbulence on algal productivity**

Flows in all of the micro-algal growth systems described above are turbulent, but variations in the reported velocity required to prevent micro-algal settlement might be due to differences in system dimensions together with different algal species. Increasing the Reynolds number can increase micro-algal growth yield, but growth can be supressed if turbulence is too high. Flow rates with Reynolds Numbers as low as 6500 in closed tubular micro-algal growth systems have been found to have a negative effect on micro-algal productivity due to cell damage (Grobbelaar, 1994, Grobbelaar, 2009b). Disruption of the cells may occur as a result of localised velocity gradients within an eddy or between eddies (Rodriguez et al., 1993). The exact nature of the micro-eddy hydrodynamic forces acting on the cells; shear, compression, torsion or impact, causing cell disruption are not fully understood (Clarke et al., 2010), but micro-algal cells appear to be damaged when the size of the micro-eddies is the same or smaller than the algal cell (Fernandez et al., 2001, Molina et al., 2000, Molina Grima et al., 2010). Micro-eddy dimensions reduce with increasing velocity and decreasing hydraulic diameter or depth and the very low Reynolds number found to damage micro-algal cells in a closed photo-bioreactor (Grobbelaar, 1994) may be due to the narrow tube diameter,

<0.05 m, used in the study. Increasing mixing and the length of time algae are mixed have been suggested to improve the 'harvestability' of micro-algae by encouraging flocculation, but no details of the mechanism that encourages flocculation or the fluid velocity or Reynolds required has been given (Benemann et al., 1980).

Mixing in raceways is a complex concept and may not be adequately described by either fluid velocity or Reynolds number (Chiaramonti et al., 2013). Recent work by the University of Southampton and University of Almeria has shown that there is little vertical or horizontal movement of small particles across the flow direction in open raceways in turbulent flow (Mendoza et al., 2013a). Computational Fluid Dynamic analysis of the flow field of a raceway and practical studies by the University of Florence have produced similar observations and it was concluded that in raceway straights there is little vertical mixing (Chiaramonti et al., 2013, Tredici, 2012). This observation may be explained by the velocity within eddies being substantially lower than average fluid velocity (Vennard and Street, 1976). Studies of small-scale and full-scale raceway ponds have found that in turbulent flow there is good cross-sectional mixing, but the flow is essentially plug flow with little or no back mixing (Buhr and Miller, 1983, Chiaramonti et al., 2013).

2.2.2.3 **Head loss**

The resistance to flow in a channel can be reported in terms of head loss. Head loss is unavoidable due to friction between the fluid and the walls of the channel and between adjacent fluid particles as they flow. Head loss in a straight channel can be expressed as:

Equation 3

$$h_s = \frac{L_s n^2 v^2}{R_h^{\frac{4}{3}}}$$

Where v is the average fluid velocity, L_s is the length of the straight, n is the Manning Friction Factor (a dimensionless coefficient used to describe the relative roughness of a channel) and R_h is the hydraulic radius. The head loss in the straight is a function of the square of the fluid velocity. The hydraulic power (P_h) required to move a fluid in channel is a function of the head loss:

Equation 4

$$P_h = Q\rho g h_s$$

where Q is volumetric flow rate, the product of flow area and average fluid velocity. Hydraulic power is, therefore, a function of the cube of fluid velocity, and small changes in fluid velocity can cause large changes in the energy and power required to move fluid within a straight channel. The minimisation of fluid velocity needed in raceway ponds for optimum micro-algal growth could have a significant effect on the energy balance of algal cultivation.

Inducing mixing by introduction of 'flow obstructions' acting as static mixers could offer a low energy method of maintaining algal suspension and improving mass transfer within the raceway rather than increasing fluid velocity. Both vertical and horizontal foils or vanes, made of 6 mm profile stainless steel, have been found to improve algal production in raceways (9 % and 14 % respectively) (Laws and Berning, 1991). A more recent study has also suggested that a large energy-saving opportunity may exist if the average flow velocity in raceways is reduced and mixing promoted by the installation of static mixing devices in the flow (Chiaramonti et al., 2013).

The power and energy requirements of maintaining fluid flow in raceways are discussed further in subsequent Sections (2.2.3).

2.2.2.4 **Depth of raceways**

The depth of an algal raceway is a compromise between being sufficiently shallow to give adequate light for the algal cell growth; and deep enough for fluid to be moved around the raceway while avoiding the costs of raceway bed grading (Borowitzka, 1999). Raceways can be up to 0.5 m deep (Brennan and Owende, 2010), but are typically between 0.2 and 0.3 m deep (Aquafuels, 2011, Jimenez et al., 2003, Johnson et al., 1988, Lundquist et al., 2010). Below 0.15 m problems have been found with algal biomass sedimentation and with achieving sufficiently even grading of the pond bottom to ensure consistent flow around the entire raceway (Weissman et al., 1989). Mixing problems, temperature variation and high rates of carbon dioxide outgassing may also occur at depths below 0.25 m (Lundquist et al., 2010).

2.2.2.5 **Width of raceways**

In wide channels, where the width of the channel is 10 times or more than the depth of the channel, the velocity distribution in the centre of the channel is essentially the same as an infinitely wide channel (Chow, 1959): this may indicate that the minimum width of the channel should be 3m if the depth of the channel is 0.3 m. Two dimensional channel flow, where all flow occurs in a set of parallel planes with no flow normal to them, occurs at aspect ratios, width-to-depth, of 7 to 1, but the development of such flow can take a distance of up to 500 times the depth (Monty, 2005). The US Energy Department study suggested a length to width ratio of 11 to 1 or greater (Weissman et al., 1989). Length is discussed in Section 2.2.6.

No maximum channel width appears to have been given in the literature, but problems of consistent flow are likely to occur with excessively wide raceways. The fiction factor of an open raceway increases with increasing hydraulic radius and therefore with width as described in the Strickler friction factor equation:

Equation 5

$$n = 0.115\, f^{1/2}\, R_h^{\ 1/6}$$

where n is the Manning friction factor and f is a friction factor dependent on roughness, especially in highly turbulent flow (high Reynolds Number), and R_h is hydraulic radius (Chow, 1959, Vennard and Street, 1976).

The velocity profile of the turbulent flow in open channels will also change with increasing width as describe by the Prandtl Power Law (Chanson, 1999):

Equation 6

$$\frac{v_l}{v_{max}} = \left(\frac{y}{d}\right)^{\frac{1}{N}}$$

Where v_l is local fluid velocity, v_{max} is maximum fluid velocity at the surface, y is the distance from the bottom normal to the flow direction and N is the power law exponent. The value of N can vary from 4 for shallow flow in a wide rough channel to 12 in a smooth narrow channel, with a typical figure of 6 being representative of channel flow in smooth concrete channels (Chanson, 1999, Cheng, 2007). Using the Prandtl Power Law to calculate the extreme values of N, 4 and 12 the graphs of flow in Figure 10 can be derived.

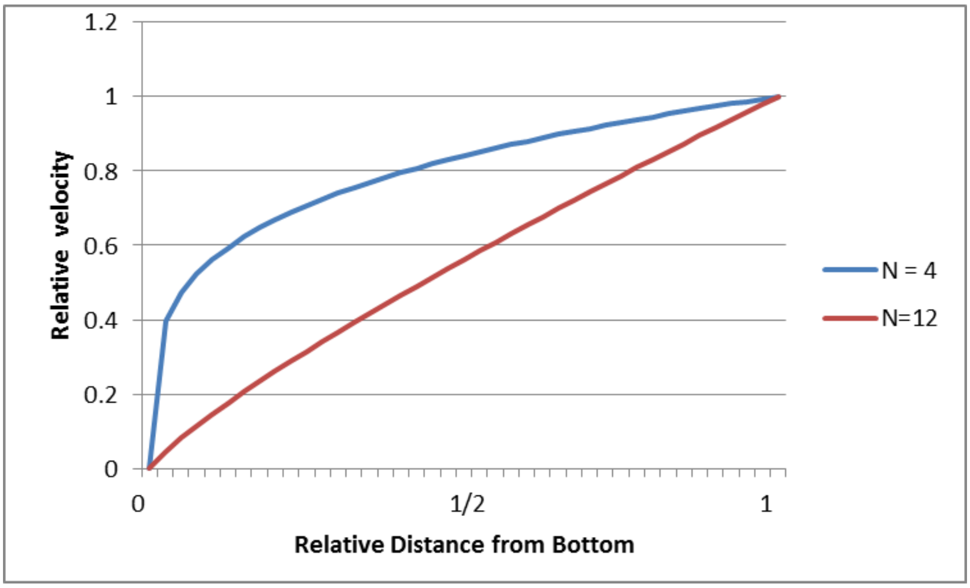


Figure 10 Velocity profiles of flow in open channel for extreme values of N.

The value in the wide channel, with N of 4, gives the classic 'snub nosed' profile of turbulent flow, while the narrow smooth channel, with N of 12, results in a profile of the type found in laminar flow. These profiles may also help to explain the observations at the University of Southampton and University of Almeria that showed little vertical movement of micro-algal across the flow direction (Mendoza et al., 2013a). The experimental raceway used was narrow, 1m wide, and constructed of relatively smooth glass-fibre reinforced plastic which could have resulted in a laminar-type flow velocity profile despite the flow being turbulent.

The Froude number (a dimensionless ratio of inertial and gravitational forces that describes different flow regimes of open channel flow) is always below 0.3 for the range of fluid velocities (0.1 to 0.3 m s^{-1}) and depth (0.1 to 0.3m) typically found in typical open raceways and the flow is subcritical or tranquil in all cases and will tend to show little surface disturbance. "Waves are not usually a problem in raceway ponds" (Oswald, 1988), although wind fetch could be a problem in ponds larger than one hectare (Benemann and Oswald, 1996).

2.2.2.6 **Raceway bends**

The head loss in a bend (h_b) can be expressed as:

Equation 7

$$h_b = \frac{kv^2}{2g}$$

where k is the head loss factor. High head losses factors of 2.4 to 2.5 have been reported for bends in raceways (Chiaramonti et al., 2013, Green et al., 1995), much higher than for hydraulic optimised bends (Chow, 1959, Idel'chik, 1986, Vennard and Street, 1976).

The high head losses on the bends relative to straights indicate that a simple raceway rather than a serpentine route would be more energy efficient. In a 'realistic' assessment of algal biofuel production it was also concluded that a single loop rather than a serpentine layout was the preferred design to minimise head losses (Lundquist et al., 2010). Most open commercial raceways are of a single loop rather than serpentine design. Figure 11 shows the extensive single loop raceways for the production of non-fuel micro-algae in Hawaii.

Head loss reductions could be made by increasing internal radii and by the addition of flow guides. Flow guides can reduce the head-loss factor of a 90° elbow from 1.15 to 0.55 (Idel'chik, 1986) and flow guides have been shown to even out flow and reduce areas of stagnation in raceways (Weissman et al., 1989). Curved baffle boards directing flow around raceway bends have been shown to reduce the head loss factor in bends to 2 resulting in a raceway mixing energy reduction of 7.5 % compared to a raceway with unbaffled bends (Chiaramonti et al., 2013). Three curved baffle boards in raceway bends were found to reduce the energy required to pump fluid around a 0.5 hectare raceway by 17 % compared to a raceway with unbaffled bends and also to reduce 'dead zones' (areas of low mixing with a flow velocity < 0.1 m s^{-1}) from 14.2 % of the raceway area to 0.9 % (Sompech et al., 2012).

Figure 11 Single loop raceways in Hawaii. Courtesy Cyanotech.

Bends offer the largest resistance where the curvature of the inner wall is sharp causing the flow to separate from the inner wall. In Figure 12 vortices (highlighted red) can be seen shedding from the sharp inner edge of an algal raceway. Rounding of the inner wall lowers resistance by making flow separation smoother; with the optimum internal radius being between 1.2 and 1.5 times the channel width (Idel'chik, 1986). Such large internal radii may be impractical and uneconomic due to the large land requirement needed for the centre of the raceway; but small internal bend radii can give significant reductions in head loss, and commercial producers of micro-algae in open ponds often add a bulbous end to the internal wall to reduce head losses as can be seen in the raceways in Figure 11. A bulbous inner radius to a raceway bend was found to 'fill in' the potential dead zone region and reduce the energy required to circulate fluid around a raceway, but curved flow guides were more effective (Sompech et al., 2012). A combination of flow guides and a bulbous inner radius to raceway bends was found to effectively remove all ‘dead zones’, but was considered too costly for micro-algal cultivation raceways (Sompech et al., 2012)

Computational fluid dynamic analysis has shown that most vertical mixing occurs in raceway bends rather than the straights (Chiaramonti et al., 2013, James and Boriah, 2010, Tredici, 2012). Interaction between the fluid and the bend walls and bottom sets up a ‘helical flow pattern’ that can be beneficial to algal growth by bringing micro-algal from the bottom to the top (James and Boriah, 2010).

Figure 12 Vortices being shed from sharp inner corner of algal raceway

2.2.3 **Fluid propulsion in raceways**

Sorguven and Ozilgen (2010) stated that "the challenge of algal cultivation is to maintain mixing with the minimum of energy". The minimum circulation velocity to ensure adequate mixing has been discussed in Section 2.2.2.1, but how is this fluid motion to be generated? The paddlewheel has become the universal method of producing fluid motion in raceways as they are mechanically simple, high volume, low head devices with a gentle mixing action (Borowitzka, 2005).

Pumping is less energy efficient than stirring for mixing in a tank (Perry and Chilton, 1973) and paddlewheels may be more analogous to a 'stirrer' than a 'pump'. Algal growth has been shown to be inhibited by high shear (Hondzo et al., 1997) thereby probably excluding the use of many pump types. Axial pumps have been shown to be more efficient than centrifugal designs in pumping algae especially as volumetric flows increase and hydraulic head decreases. They are suitable for low head and high volume and could be an alternative to paddlewheels particularly in very shallow ponds e.g. ≤ 0.005 m (Chiaramonti et al., 2013).

Fluid is returned by a pump into the raceway from the harvesting system to recycle water and nutrients. The use of the post algal harvest fluid stream to drive fluid around the raceway could potentially be used to provide fluid momentum to circulate liquid around the raceway. However, calculating the fluid velocity and pipe diameter using conservation of momentum and a simple mass balance have shown that this is impractical due to the very high fluid velocity (~150 m s^{-1}) and narrow diameter pipes (1-2mm).

In marine propulsion paddlewheels have been replaced by screw propellers, but this was for practical ship design and operational factors rather than advantages in efficiency (Rennie, 1853). Screw propellers have not replaced paddlewheels in raceways as they can have operational problems in the shallow depth of raceway ponds due to cavitation, and paddlewheels have been shown to be superior to screw propellers in open growth systems (Laws and Berning, 1991). In New Zealand, paddlewheels have now also replaced propellers in advanced micro-algae waste treatment raceways as they were found to be more energy efficient (Craggs, 2003).

Both Archimedes screws and gas lift systems have been found to be superior to paddlewheels for algal growth (Laws and Berning, 1991) (Ketheesan and Nirmalakhandan, 2011), but both require considerably greater head differences than paddlewheels for effective operation. The pumping energy of an air-lift pump to circulate liquid in a raceway has been estimated at being twice that of a paddlewheel (Becker, 1994).

The advantages and disadvantages of the various methods of circulating fluid around a raceway are summarised in Table 4.

Table 4 Summary of advantages and disadvantage of raceway circulation methods

Circulation Mechanism	Advantages	Disadvantages
Paddlewheel	✓ Mechanically simple, with a gentle mixing action ✓ Widely used and proven in raceways	• Low efficiency in many designs used in raceways • Design needs to optimised
Axial Pump	✓ Found to be effective in low depths ~ 5 mm	• Cost • Shear
Air-lift	✓ Can combine gaseous transfer and fluid flow.	• Operational energy • Sump depth
Screw Propeller	✓ Light weight ✓ Low drag	• Cavitation • Paddlewheel found to be more efficient in raceways
Archimedes' Screw	✓ More efficient than most pumps and paddlewheels	• Requires greater head than typically found in raceways

Paddlewheels can be extremely efficient and are showing considerable promise as hydropower converters at very low head differences (Senior et al., 2010), but figures quoted for paddlewheel efficiency in algal growth systems are low at 10 to 20 % (Borowitzka, 2005, Chiaramonti et al., 2013, Green et al., 1995, Putt, 2007). The paddlewheel efficiency generally increases with increasing number of blades, increasing diameter and reducing clearance between the paddlewheel and the bottom (Müller et al., 2007, Rennie, 1853, Senior et al., 2010). Paddlewheels of more than eight blades have been suggested as impractical for raceway ponds, and a paddlewheel of 1.5 m in diameter with 8 blades, with a blade height of 0.35 m, running in a 0.3 m deep sump (0.1 m below the pond bed depth) with a clearance of 0.02 m has been suggested as a suitable design for a 0.2 m deep raceway (Borowitzka, 2005). Efficiencies of 40 % have been suggested for optimised paddlewheel designs in raceways (Benemann and Oswald, 1996), and efficiencies of up to 75 % have been found for paddlewheels extracting energy from flows with low-head differences

(Senior et al., 2010). Reducing energy input for mixing micro-algal raceways has not been extensively studied (Chiaramonti et al., 2013, Ketheesan and Nirmalakhandan, 2011, Sorguven and Ozilgen, 2010) and it would appear that considerable improvements could be made to micro-algal raceway paddlewheel design. Work is currently being undertaken by the University of Southampton on improving paddlewheel efficiency.

2.2.4 **Light penetration and light climate in raceways**

The 'light climate' experienced by micro-algae will depend not only on culture depth and fluid mixing, but also on the light adsorption and dispersion from the micro-algal biomass, other particulate matter, dissolved materials and the water itself (Martinez, 1996). The intensity of bright light, 3.2×10^5 Lx (30,000 foot-candles), over three times the light intensity of a bright summers day (MAF NZ, 2007), is reduced by over 99 % in a depth of 0.1 m in an algal suspension of 0.025 % dry weight of *Chlorella* and if the algal concentration is increased to 0.25 % the 99 % reduction in light intensity is achieved within 0.015m (Rabe and Benoit, 1962). The exponential nature of the Beer-Lambert law ensures that even substantial variations in light intensity will make little difference to light penetration in micro-algal cultures (Buhr and Miller, 1983). Very dense cultures of micro-algae (undefined) can absorb over 90 % of light within the first 0.01m (Love, 2011). Field observation using a Secchi disc established a simple correlation between light penetration depth d_p in centimetres and the concentration of algal C_c expressed as mg l^{-1} dry weight of algae per unit volume of algal suspension given in (Oswald, 1988).

Equation 8

$$d_p = \frac{6000}{C_c}$$

Using micro-algal concentrations in the above equation of 0.025 % (250 mg l^{-1}) and 0.25 % (2.5 g l^{-1}) light penetration depths can be calculated of 0.24 m and 0.024 m respectively.

Light irradiance has been found to be a limiting factor in the growth of *Spirulina* in southern Spain during the summer. The maximum algal yield in

0.3 m deep ponds was at micro-algal dry weight concentrations of between 0.04 to 0.05 % with higher concentrations of algae reducing yield due to poor light penetration into the medium (Jimenez et al., 2003). It has been suggested that in typical raceway conditions only the micro-algae in the upper 0.005 m to 0.02 m of a pond are light saturated and that 90 to 95 % of the culture depth is light limited (Williams and Laurens, 2010). Improving light availability to all micro-algal cells within a culture continues to be one of the greatest challenges of micro-algal mass culture (Williams and Laurens, 2010).

The rapid cycling of micro-algal cells from dark to light has been reported to increase growth rates (Grobbelaar, 2009a, Janssen et al., 2001), but the optimal light dark frequency appears to be culture specific and also depends on the light intensity (Molina et al., 2000). The effect may be enhanced by longer dark than light periods (Bosma et al., 2007), but phytoplankton have been shown to become acclimatised to the rapid light dark cycle and any increase in micro-algal production rates can be lost within three days (Grobbelaar, 2010). The increase in productivity is attributed to the slower dark reactions of photosynthesis catching up on light reactions of photosynthesis (Benemann and Oswald, 1996, Williams and Laurens, 2010). The minimum frequency to achieve an increase in micro-algal production rate from a rapid dark light cycle or flashing light effect is 1Hz (Molina Grima, 2011, Molina Grima et al., 2010, Williams and Laurens, 2010), but the maximum effect is probably achieved at much higher frequencies with a light period of under 10 ms (Bosma et al., 2007, Grobbelaar, 2010, Janssen et al., 2001). The achievable frequency of the light-dark cycling is considerably slower in open systems (Grobbelaar, 2010) with typical values around 0.1 Hz at flow rates between 0.15 and 0.2 m s^{-1} in open ponds 0.15 to 0.2 m deep (Williams and Laurens, 2010). No effect on micro-algal productivity could be attributable to a light-dark cycle at fluid velocities between 0.02 to 0.5 m s^{-1} when other conditions (CO_2 supply and oxygen tension) were constant (Benemann and Oswald, 1996) . It might be possible to achieve a rapid light-dark cycling in systems where the light path is short, e.g. under 0.05 m, but open raceways are likely to provide cycle times of seconds with minimal or no stimulation of growth rate due to the frequent exposure to light and dark (Garcia Camacho et al., 2011, Grobbelaar, 2009b, Molina Grima, 2011). Early studies of stirred systems also concluded that the rapid light dark cycle or flashing light effect could only be achieved at "uneconomical stirring rates" (Tamiya, 1957).

2.2.5 Gaseous transfer zones and sumps in raceways

Photosynthesis can be simplified into two reactants (carbon dioxide and water) and two products (glucose and oxygen), represented by the equation:

Equation 9

$$6CO_2 + 6H_2O = C_6H_{12}O_6 + 6O_2$$

Stoichiometrically, this equation suggests that every 1 kg of algae produced will require 1.5 kg of CO_2. Values for the amount of CO_2 required to produce algal biomass are quoted at between 1.65 (Berg-Nilsen, 2006) and 2.2 (Weissman and Goebel, 1987) kg per kg dry weight with a typical figure of 1.8 kg of CO_2 per kg of dry algae (Chisti, 2007). Stoichiometric analysis suggests that the amount of CO_2 required increases as protein and especially lipid contents increases and carbohydrate decreases.

Algae rapidly deplete the CO_2 within a growth medium and to maximise algal growth additional CO_2, to that available via atmospheric transfer, should be supplied (Gouveia, 2011, McGinn et al., 2011). If 1.8 kg of CO_2 is consumed by photosynthesis this will produce 1.3 kg of O_2 and again it can be demonstrated by basic calculation that this will accumulate within the algal growth medium unless some additional means of gaseous transfer is used. Elevated oxygen levels of only a few times saturation in water have been shown to inhibit algal growth (Jimenez et al., 2003, Laws and Berning, 1991, Li et al., 2003, Marquez et al., 1995, Molina Grima et al., 2001).

CO_2 can be supplied as gas via bubbles or by the addition of carbonated water, but the supply of gaseous CO_2 has been shown to give higher growth (Laws and Berning, 1991) possibly due to removal of O_2. It has been suggested, that from an engineering point of view, gaseous transfer is the key issue in the design of algal growth systems (Lundquist et al., 2010).

Suggestions for gaseous exchange sumps range from simple open pits to more complex counter current units with depths ranging from under 1m to 3m deep and bubble sizes ranging from 1mm to 3mm (Putt, 2007, Weissman et al., 1989). Sump depth to achieve 90 % gaseous transfer of CO_2 from flue gas to the liquid medium in a raceway pond has been calculated at 3 m (Putt, 2007, Stephenson et al., 2010). The modelling of gas transfer in sumps is not highly accurate, in part due to uncertainties about bubble size and distribution, with the gaseous transfer rate in shallow sumps being greater than predicted by simple single-size bubble models (Weissman and Goebel, 1987), and the calculations used to arrive at a rough design of the carbonation sump being "fairly crude" (Stephenson, 2009). In a large-scale experimental micro-algal open growth pond 78 to 90 % of carbon dioxide gas in bubbles was transferred to the micro-algal growth medium in a sump 0.9 m deep, with results indicating that greatly increasing depth may not greatly increase the total transfer of carbon dioxide (Weissman et al., 1989). The cost of construction of a sump and the operational energy required increases with increasing depth, and it has therefore suggested that sump depths should be kept below 1.5 m (Weissman and Goebel, 1987). The reduction in depth of the sump from 3 to 1.5 m has been estimated to reduce the energy required by over 50 % (Stephenson, 2009).

Recent work by the University of Southampton, at the Cajamar Experimental Station in Southern Spain, has indicated that a simple 1.8 m deep sump can transfer 90 % of the carbon dioxide from flue gas containing 10 % CO_2 to water. The gas transfer efficiency of a sump was found to reduce as the depth of the sump reduced.

The head loss in a sump h_g can be represented by:

Equation 10

$$h_g = \frac{kv^2}{2g}$$

Only one head loss factor of 2 (Weissman et al., 1989) has been found in the literature for a simple open gaseous exchange sump in a raceway. Standard data on tees and bends (Idel'chik, 1986) indicate that more complex sumps will have higher head loss factors than horizontal bends. Results from a pilot scale pond (50 m long and 1m wide channels) at the Cajamar Experimental Station in Southern Spain suggest that that power required to circulate a fluid in a raceway is approximately doubled when a baffled counter current sump is used rather than a simple open sump. Mixing time was found to be more than doubled for a baffled counter current sump compared to a simple open sump (Mendoza et al., 2013a).

The power to pump CO_2 into the algal suspension could be considerable. The energy required to transfer 1 kg of oxygen into primary effluent in aerated activated sludge systems is 0.4 to 1.1 kWh (Green et al., 1995). There are some partially studies of the energy inputs required for gaseous sumps (Putt, 2007, Stephenson et al., 2010), and further work is being undertaken at the University of Southampton.

2.2.6 **Raceway size**

Work in Israel concluded that the maximum size of a raceway driven by a single paddlewheel is 1500 m^2 (Ben-Amotz, 2008b). The largest commercial raceway, however, is reported to be 5000 m^2 (Becker, 1994) and it has been proposed that raceways could be as large 10000 m^2 (Borowitzka, 2005).

The circuit length of the raceway pond is determined by the head drop permissible across the paddlewheel and/or the maximum variation of pond depth to allow consistent light penetration and mixing around the raceway. Oswald suggested that, in order to achieve consistent mixing and light penetration, the maximum head drop around a raceway should be half the mean depth (0.15 m for a depth of 0.3 m) and calculated the maximum length and area for 6m wide ponds as 5420 m and 32500 m^2 for a 0.3 m deep pond flowing at 0.15 m s^{-1} (Oswald, 1988). This calculation considering only frictional head losses in the raceway straights and took no account of the additional head losses in raceway bends and other features such as gaseous exchange sumps, and therefore overestimated the potential maximum size of a raceway.

The maximum head variation across a paddlewheel may be less than 0.15 m further limiting the maximum size of a raceway. A maximum head difference of 0.076 m has been recommended by a paddlewheel manufacture for commercial algal raceways (Waterwheel Factory Inc. private communication, 2010) and the head difference across a single paddlewheel in mixed crayfish ponds has been reported at only 0.031 m (Pfeiffer et al., 2007). Substituting a head of 0.076 m in Oswald calculation would halve the maximum length and area for a 0.3 m deep raceway, but still indicates that is feasible to build raceway ponds with an area greater than 1500 m^2 and raceways of one hectare could be practicable.

The head losses for power requirement to circulate growth medium around algal raceway are calculated and further discussed in Sections 4.2 & 6.3.1

2.2.7 Requirements for micro-algal growth

The growth of micro-algae requires water and a source of carbon together with both macro-nutrients such as nitrogen (N) and phosphorus (P) as phosphate and a range of micro-nutrients. Micro-algae have a high content of both N and P relative to land plants at 5 to 12 % and 0.3 to 1 % respectively (Hannon et al., 2010, Williams and Laurens, 2010). An extensive review of the of potential resource demands of micro-algal biofuel in the USA has concluded that carbon dioxide, nitrogen and phosphate are likely to emerge as the dominant constraints for scale-up in autotrophic systems (Pate et al., 2011).

2.2.7.1 Nutrients and water

Although some cyanobacteria have the ability to fix nitrogen, virtually all algal species require an exogenous source of fixed nitrogen with most using ammonia preferentially (Hannon et al., 2010, Lundquist et al., 2010). Nitrogen fertilisers are often made using the Haber-Bosch process which uses methane and nitrogen to produce ammonia (NH_3). This not only has a significant cost, at US$ 300 $tonne^{-1}$ of NH_3 (Williams and Laurens, 2010), but the embodied energy within the fertiliser also adds considerable energy inputs of 34 to 50 GJ $tonne^{-1}$ of nitrogen (Jenssen and Kongshaug, 2003). The embedded energy in the supply of nutrients and CO_2 could be greater than energy produced unless they are from ‘discounted inputs’ such as wastewater for nutrients or flue gas for

CO_2 (Beal et al., 2012a), and/or are recycled. Recycling of harvest water back to the growth system has been found, in a life cycle assessment of the production of micro-algal biodiesel, to reduce nutrient requirement by 55 % (Yang et al., 2011b), while anaerobic digestion of algal biomass could allow recycling of 90 % of nutrients (Lundquist et al., 2010, Williams and Laurens, 2010). If biogas or lipids for biodiesel are being produced there will be little or no export of N and P in the products and the nutrients are therefore available for recycling with potentially only a small top-up to cover processing losses.

Phosphorus, although required in smaller quantities than nitrogen, may present a greater challenge for micro-algal biofuel production. Agricultural fertilisers currently used in the USA contain less than optimal concentrations of phosphate due to limited supplies, as mined phosphate production peaked in the late 1980s (Hannon et al., 2010). Although LCAs have found very little impact from the use of phosphate on energy return and global warming potential, the effect on sustainability of micro-algal production could be significant due to the rapid depletion of mined phosphate reserves (Aquafuels, 2011, Ferrell and Sarisky-Reed, 2010). Both phosphate and nitrogen are found in municipal wastewater at typical concentrations of 30-40 mg l^{-1} N and 5-10 mg l^{-1} P (Williams and Laurens, 2010). *Chlorella* and *Scenedesmus* have both been found to grow in a wide range of wastewaters, often with sub-optimal ratios of N and P, with 'complete decomposition' of nitrogen and phosphate nutrients in 10 days (Ahrens and Sander, 2010). In what was termed a "realistic technology and engineering assessment of algae biofuel" it was found that there could be no favourable outcome for algal biofuel production unless wastewater treatment was the primary goal of the process (Lundquist et al., 2010). A mass balance for the EU FP7 All-Gas project has not only shown that sufficient N & P can be provided for micro-algal growth by wastewater, but also that with recycling valuable N & P can be exported from the system for other fertiliser uses (Banks, 2011, Banks et al., 2011a).

Beal (2011) stated that; "it does matter how much energy you produce per acre if it requires more water than is available". It will be vital to minimise freshwater use in micro-algal biofuel production not only to minimise cost, but to avoid future 'water versus fuel' debates (Hannon et al., 2010). Recycling can reduce water usage for micro-algal biofuel production by 84 % (Yang et al., 2011b). One of the main advantages of micro-algae for biofuel production is their ability to grow in water ‘unsuitable’ for land crops (Ferrell and Sarisky-Reed, 2010). The use of wastewater or sea water for micro-algal biofuel production can reduce the freshwater demand by 90 % (Gouveia, 2011). Freshwater is required to compensate for water loss and avoid salt build-up due to evaporation (Yang et al., 2011b).

Algae also have a requirement for other nutrients in small quantities. Twenty six of twenty seven micro-algae were found to require vitamin B_{12} for growth which can be produce by bacteria (Kazamia et al., 2012). Wastewater and seawater together with the bacteria found in them could provide the essential micronutrients, minerals and vitamins required.

The supply and reuse of materials for micro-algal growth will have significant influence on the economics, energy balance and sustainability of micro-algal biofuel systems. For micro-algal biofuel to be successful it will need low cost and sustainable sources of water and nutrients, and both will need to be recycled. Wastewater could supply both nutrients and water. Seawater or brackish water can provide not only a useable water source, but also some nutrients and could be used in combination with nutrients from wastewater or low cost digestate from other organic wastes.

The research will focus on operational energy inputs and technology of unit operations of micro-algal biofuel production systems, with embodied energy inputs in terms water and nutrients minimised; by recycling, using methods such as anaerobic digestion that allow return of nutrients and water to the growth system; and by the use of low embedded energy sources of water and nutrients, such as wastewater.

2.2.7.2 **Carbon dioxide**

Carbon dioxide is needed for the autotrophic growth of micro-algae. To maximise algal growth CO_2 should be supplied in addition to that available via atmospheric transfer (Section 2.2.4). The manufacture and supply of pure CO_2 for algal biofuel production is not viable energetically or economically (Aquafuels, 2011, Beer et al., 2007, Campbell et al., 2009). Most studies have generally considered flue gas from fuel combustion as the main source of CO_2 (Aquafuels, 2011, McGinn et al., 2011).

Flue gases from power plants account for over 7 % of the world's CO_2 emissions (Gouveia et al., 2010). They could be available at little or no cost and it has been suggested that micro-algae grown on CO_2 from flue gas should be eligible for financial support as a GHG mitigation technology (Parliamentary Office of Science & Technology, 2010). However the EU and UN do not currently considered the use of fossil fuel flue gas for the production of micro-algal biofuel eligible for financial support from climate change reduction funding, as the CO_2 taken up by the micro-algae is released back into the atmosphere when the biofuel is burnt (Parliamentary Office of Science & Technology, 2011). Power plants also normally produce CO_2 24 hours per day, but algae will only consume it during daylight, and are therefore unable to mitigate night-time CO_2 emissions without flue gas storage, which is probably prohibitively expensive.

The use of fossil fuel generated flue gas for the production of algae may not be considered sustainable, but use of flue gas from biomass or biofuel combustion, as in the EU FP7 All-Gas project (Banks et al., 2011a) could be sustainable. Although flue gas purchase costs are zero, or low, the costs of gas transport are not 'trivial' (Brune et al., 2009). Algal raceways should be located near to the source of flue gas allowing the use of a low pressure blower. If the transport distance increases a high pressure compressor will be required, increasing energy costs by an order of magnitude (Kadam, 2002).

Flue gas typically has a CO_2 content of 6 to 13 % (Brune et al., 2009, Douskova et al., 2009) with concentrations from gas-fired stations typically lower, at 7.4 to 7.7%, than from coal-powered stations at 12.5 to 12.8 % (Xu et al., 2003). In addition to CO_2 flue gas will contain nitrogen, water vapour and oxygen together with smaller amounts of potentially toxic gases: carbon monoxide, at 50 to 300 ppm; mono-nitrogen oxides (NO_x), at 60 to 420 ppm; and sulphur oxides (SO_x), up to 420 ppm (Xu et al., 2003). The emission of flue gases, which are low in oxygen and contain CO, NO_x and SO_x, at ground level could have Health and Safety implications, but 70 -80 % of the CO_2, NO_x and SO_x is readily removed from the flue gas by sparging the gas into a raceway pond (Downey, 2012), greatly reducing the potential hazard.

Flue gases have been found to have no negative effect on the growth of micro-algae (Aravanis, 2012, Laws and Berning, 1991) and growth rates for *Chlorella* were shown to be higher on flue gases with a CO_2 content of 11-13 % than air enriched to 12 % CO_2 (Douskova et al., 2009). NO_x in flue gases has been found to have little negative effect on micro- algae and can also provide a nitrogen source (Sheehan et al., 1998). In a study by MIT micro-algae were been found to be able to utilise not only the carbon dioxide from boiler emissions, but also 85 % of NO_x (Envirotech, 2004), potentially reducing nitrogen fertiliser demand.

Low cost carbon input appears to be vital for micro-algal growth, and flue gas could provide a source. The use of fossil fuel flue gas for micro-algal biomass production, although unsustainable, may be a valuable GHG mitigation strategy, as CO_2 is used in a second fuel before being released to the atmosphere, but it is eventually released and not sequestered. The use of flue gas from biomass could be sustainable. The current research will therefore focus on the use of flue gas as a carbon source that is assumed to have no embodied energy. Energy inputs for CO_2 supply can be reduced to those required to transport gas to the gaseous transfer sump.

2.3 **Algal harvesting**

Algal biomass can be ‘energy rich’, but the growth of algae in dilute suspension at around 0.02 to 0.05 % dry solids (Zamalloa et al., 2011) poses considerable challenges in achieving a viable energy balance from the process operations needed for biofuel production. The challenges of algae harvesting come from the small size of micro-algal cells, as most algae are below 30 µm (Molina Grima et al., 2003); the similarity of density of the algal cells to the growth medium (Reynolds, 1984); the negative surface charge on the algae that results in dispersed stable algal suspensions, especially during the growth phase (Edzwald, 1993, Moraine et al., 1979, Packer, 2009); and the algal growth rates which require frequent harvesting compared to terrestrial plants.

The cost effective harvesting of micro-algae is considered to be the most problematic area of algal biofuel production (Greenwell et al., 2010) and a key factor limiting the commercial use of micro-algae (Olguín, 2003). It has been suggested that 20 to 30 % of the costs of micro-algal biomass is due to the costs of harvesting (Mata et al., 2010, Molina Grima et al., 2003, Verma et al., 2010), but estimates as high as 50 % of micro-algal biomass cost have been given (Greenwell et al., 2010). It has been estimated that 90 % of the equipment cost for algal biomass production in open systems may come from harvesting and dewatering (the removal of water from the micro-algal biomass by mechanical methods, such as centrifugation or filtration) (Amer et al., 2011). The need for continuous harvesting of the dilute suspension makes the harvesting of micro-algae 'inherently more expensive' than harvesting land plants (Benemann et al., 1977), and the separation of micro-algae by settlement and centrifugation has a typical harvesting energy requirement of 1 MJ kg^{-1} of dry biomass (Sawayama et al., 1999). This is greater than the energy cost of harvesting wood at 0.7 – 0.9 MJ kg^{-1} (Sawayama et al., 1999). The cost of harvesting micro-algae, in both energy and economic terms, therefore needs to be reduced. A recent report by UK's Biotechnology and Biological Sciences Research Council (BBSRC) on algal research has concluded that: "hardly any commercial activity exists in downstream processing" (Schlarb-Ridley, 2011). Most work on micro-algal species selection for biofuel production has been focused on yield and composition rather than on ease of recovery (Brennan and Owende, 2010).

The final moisture content of the harvested algal biomass is an important criterion in the selection of the harvesting method (Molina Grima et al., 2003). Micro-algal biomass can spoil in hours if the moisture content is greater than 85 % (Mata et al., 2010), while high moisture content can have a substantial influence on the costs and methods of further processing (Molina Grima et al., 2003) and energy extraction from the biomass.

Algae can be harvested by a number of methods: sedimentation, flocculation, flotation, centrifugation and filtration or a combination of any of these. Despite the importance of harvesting to the economic and energy balance viability of micro-algal biofuel, there is no universal harvesting method for micro-algae (Mata et al., 2010, Shen et al., 2009). A recent extensive review of dewatering micro-algal cultures concluded that "currently there is no superior method of harvesting and dewatering" (Uduman et al., 2010). A summary of advantages and disadvantages of the various methods to harvest micro-algae is given in Table 5 (Milledge and Heaven, 2011)

Table 5 Comparison of micro-algal harvesting methods (Mohn, 1988, Molina Grima et al., 2003, Shen et al., 2009)

	Advantages	Disadvantages	Total solids output concentration
Centrifugation	Can handle most algal types with rapid efficient cell harvesting.	High capital and operational costs.	10-22 %
Filtration	Wide variety of filter and membrane types available.	Highly dependent on algal species, best suited to large algal cells. Clogging and fouling an issue.	2-27 %
Ultrafiltration	Can handle delicate cells.	High capital and operational costs	1.5-4 %
Sedimentation	Low cost. Potential for use as a first stage to reduce energy input and cost of subsequent stages.	Algal species specific, best suited to dense non-motile cells. Separation can be slow. Low final concentration	0.5-3 %
Chemical flocculation	Wide range of flocculants available, price varies, although can be low cost.	Removal of flocculants and chemical contamination	3-8 %
Flotation	Can be more rapid than sedimentation. Possibility to combine with gaseous transfer.	Algal species specific. High capital and operational cost.	>7%

2.3.1 **Sedimentation**

In sedimentation gravitational forces cause liquid or solid particles to separate from a liquid of different density, but the process can be extremely slow especially if density difference or particle size is small. Sedimentation can be described by Stokes' Law which assumes that sedimentation velocity is proportional to the square of the (Stokes') radius of the cells and the difference in density between the micro-algal cells and the medium as shown below:

Equation 11

$$\text{Settling velocity} = \frac{2}{9}\, g \frac{r_c^2}{\mu} (\rho_s - \rho_l)$$

where r_c is cell radius, μ is fluid dynamic viscosity and ρ_s and ρ_l are the solid and liquid densities.

The cytoplasm of marine micro-algae has a density between 1030 and 1100 kg m^{-3} (Smayda, 1970), the density of cyanobacteria is between 1082 and 1104 kg m^{-3} (Kromkamp and Walsby, 1990), marine diatom and dinoflagellates between 1030 and 1230 kg m^{-3} and the freshwater green micro-algae *(Chlorococcum)* between 1040 and 1140 kg m^{-3} (Van Lerland and Peperzak, 1984). The density of micro-algae is thus close to that of water and of salt water (998 and 1024 kg m^{-3} at 20° C respectively) (El-Dessouky and Ettouney, 2002, Kestin et al., 1978, Millero and Lepple, 1973) and therefore there is little density difference driving micro-algal settlement.

Stokes' law holds for spheroid shapes, but micro-algae are most often not spherical having a diverse range of shapes (Peperzak et al., 2003, Smayda, 1970, Sournia, 1978). The observed sinking rates of micro-algae have been found to deviate from calculated rates, being up to several times higher or lower than the calculated rate (Reynolds, 1984, Smayda, 1970). The settling velocity is very dependent upon the type of micro-algae present, but average settling velocities of 0.2 m day^{-1} for diatoms, 0.1 m day^{-1} for green micro-algae and 0.0-0.05 m day^{-1} for cyanobacteria have been suggested for water quality models (Cole and Wells, 1995).

A settlement velocity of 0.1 m day^{-1} was calculated using Stokes' Law (Equation 11) for a common spherical shaped micro-algae, *Chlorella* (density 1070 kg m^{-3} and average cell diameter 5 μm (Edzwald, 1993)), in freshwater (density at 20°C 998 kg m^{-3} and viscosity 1 x 10^{-3} Pa s^{-1} (Weast, 1985)). An experimental study found a considerably higher settling rate of 3.6 m day^{-1} (Collet et al., 2011), but in practice *Chlorella* does not normally settle readily (Nurdogan and Oswald, 1996). The calculated settlement velocity of *Cyclotella,* a similar sized alga to *Chlorella*, is 0.04 m day^{-1}, but the observed settlement rate was higher at 0.16 m day^{-1} (Smayda, 1970).

A study of the sinking rate of 24 autotrophic micro-algae (ranging in size from under 10 to 1000 μm) gave rates between -0.4 to over 2.2 m day^{-1} with an average of 0.6 m day^{-1}. No correlation was found between size and sinking rate and no relationship was found between cell size and sinking rates for diatoms (Peperzak et al., 2003). In a study of 20 micro-algae only four always settled readily, although 14 settled out occasionally (Peperzak et al., 2003). In another study of 30 species of micro-algae, found in wastewater, most were 'reluctant' to settle, with needle like or long cylindrical micro-algae being particularly resistant to settling (Choi et al., 2006). Filamentous algae *(Spirulina)* and colonial algae *(Micractinium, Scenedesmus)* with a cluster diameter of ~60 μm have been shown to be harvestable by settlement, but smaller algae *(Chlorella)* and motile micro-algae *(Euglena, Chlorogonium)* do not readily settle out of suspension (Nurdogan and Oswald, 1996). Dinoflagellates have been found to be able to swim at speeds of up to 0.03 m min^{-1} (Smayda, 1970) and many species of micro-algae have been shown to move upwards towards light (Kromkamp and Walsby, 1990, Smayda, 1970, Sournia, 1978).

Settlement behaviour of micro-algae varies between species, but can also alter within the same species. Settlements rates have been shown to vary with light intensity (Waite et al., 1992), nutrient deficiency has been shown to decrease settlement rate (Bienfang, 1981) and sinking rate increases in older cells, especially in senescent cells (non-dividing cells between maturity and death) (Smayda, 1970) and spore-producing cells (Bienfang, 1981). The average density of carbohydrate is 1500 kg m^{-3}, protein 1300 kg m^{-3} and lipid 860 kg m^{-3} (Reynolds, 1984), and micro-algae with a high lipid content are likely to settle less readily due to the lower density.

Sedimentation has not been widely used for separation of micro-algae (Uduman et al., 2010) and although settling has been demonstrated in pilot-scale wastewater treatment systems (Lundquist et al., 2010), it has not yet been achieved on a large scale. The sinking rate of small micro-algae 4-5 μm in the open ocean is 'insignificantly small' (Waite et al., 1992). A study of harvesting *Chlorella* by settlement found that only 30-35 % of the cells could be harvested and that concentration of the harvested algae was low, only 50 % higher than in the unsettled micro-algal suspension (Janelt et al., 1997). It was concluded that settlement may only be viable for micro-algal species with a high settlement rate such as *Spirulina* (Janelt et al., 1997).

Cell recovery and solid concentrations from micro-algal settlement are low (Mata et al., 2010, Shen et al., 2009) with cell recoveries of 60 to 65 % (Collet et al., 2011, Ras et al., 2011) and solid concentrations of up to 1.5 % total suspended solids (Uduman et al., 2010). Energy consumption of harvesting by settlement is generally low, with a lamella separators using 0.1 kWh m^{-3} to achieve an output concentration of 0.1 to 1.5 % dry micro-algal biomass (Uduman et al., 2010, Van den Hende et al., 2011).

Settlers have a simple construction, can be scaled up and have relatively low investment and operating expenses (Janelt et al., 1997). Many algal species do not readily settle and for those that do recovery rates and harvested concentrations are low, as already noted: but the settlement of colonial and larger micro-algae could be useful as a low energy input pre-concentration step for use with other harvesting techniques.

2.3.2 **Flocculation**

In flocculation dispersed micro-algal cells aggregate into larger clumps of cells, or flocs, with a higher sedimentation rate (Daintith and Martin, 2010, Gouveia, 2011, Knuckey et al., 2006). Flocculation can occur naturally in certain micro-algae, in a process known as auto-flocculation, and micro-algae may flocculate in response to environmental stress; changes in nitrogen, pH and dissolved oxygen (DO) (Schenk et al., 2008, Uduman et al., 2010). Auto-flocculation does not occur in all micro-algal species and can be slow and unreliable (Schenk et al., 2008). Flocculation can be induced by chemicals, both inorganic and organic, or by microorganisms; but flocculants may be species-specific and recovery and recycling of the flocculants can be problematic (Mohn, 1988, Molina Grima et al., 2003, Oswald, 1988, Shen et al., 2009). The shape, size and composition of flocs can be very diverse depending on micro-algal species and flocculant (Jago et al., 2007). An ideal flocculant should be inexpensive, non-toxic and effective in low concentrations (Molina Grima et al., 2003) and it should also preferably be derived from non-fossil fuel sources, be sustainable and renewable.

Flocculation has been suggested as a superior method to separate algae as it can handle large quantities of micro-algal suspension and a wide range of micro-algae (Uduman et al., 2010). Although flocculation can be a reliable and cost-effective method it is still 'quite expensive' (Benemann et al., 1980) and is normally used in conjunction with other harvesting methods (Brennan and Owende, 2010).

2.3.2.1 **Non-organic flocculants**

Lime (calcium hydroxide) has been used to remove suspended solids and micro-algae from wastewater since the 1920s (Oswald, 1988). Multi-valent metal salts, ferric chloride, ferric sulphate, and aluminium chloride (alum) are commonly used in water and wastewater treatment to remove algae, and alum has been found effective in flocculating both *Chlorella* and *Scenedesmus* (Molina Grima et al., 2003). Aluminium salts have been found more effective in the flocculation of *Chlorella* than ferric salts (Papazi et al., 2010). Ferric salts have also been found to be inferior to alum in the flocculation of micro-algae in respect of optimal dose, pH and the quality of the resultant water and slurry (Shelef et al., 1984a).

Cyclotella, a diatom with a density of 1114 kg m^{-3} and an average diameter 6 µm, does not settle rapidly and a floc of cells, at the same density, would need to be 88 µm in diameter to settle in a conventional settler (Edzwald, 1993). Alum flocculated *Cyclotella* has a lower density of 1001 kg m^{-3}, however, and the floc particle diameter would need to be 210 µm for settlement at 20 °C (Edzwald, 1993). Alum flocs typically range in diameter from 30 to 400 µm

(Hendricks, 2010), but despite this increased diameter of alum flocculated cells the low density of the floc can result in a slow rate of sedimentation (Edzwald, 1993).

Dosages of non-organic flocculants can be high at 1 g l^{-1} (Papazi et al., 2010) and although aluminium sulphate flocculated micro-algae have been used for aquaculture feed inorganic flocculants can be toxic (Harith et al., 2009). Inorganic flocculants can also have negative effects on micro-algal viability and can colour and modify micro-algal growth media, preventing recycling and reuse (Molina Grima et al., 2003, Papazi et al., 2010, Schenk et al., 2008). Although alum and other inorganic flocculants are relatively cheap compared to some synthetic organic flocculants, the higher dosage rates required can result in a higher cost per unit of micro-algae flocculated than more expensive organic flocculants (Mohn, 1988). It would appear that there is a need for alternatives to the traditional inorganic salt flocculants which are lower dose, less toxic and do not have adverse effects on growth medium recycling after flocculation.

2.3.2.2 **Organic flocculants**

Up to the late 1970s no polyelectrolyte flocculant effective for micro-algal effluent was available (Moraine et al., 1979), but effective polyelectrolytes were later found (Shelef et al., 1984a). Cationic polyelectrolytes are now considered as the most effective flocculants for recovery of micro-algae (Uduman et al., 2010). Recent research at the University of Almeria has found cationic polyelectrolytes more effective at flocculating freshwater micro-algae than metal salts, achieving high biomass concentration (concentration factor up to 35 times) at lower dosage rates of 2 to 25 mg l^{-1} (Granados et al., 2012).

Magnafloc LT25, a non-ionic polymer from BASF, has been found effective in flocculating a wide range of micro-algae at an addition rate of 0.5 mg l^{-1} of algal suspension in conjunction with pH adjustment to 10-10.6, with micro-algal concentrations in the settled floc 200-800 times higher than in the original suspension (Knuckey et al., 2006). Magnafloc LT25 has also been found effective in flocculating *Chaetoceros* at a dosage of less than 1mg l^{-1} while maintaining high micro-algal cell viability (over 75 %) (Harith et al., 2009).

Magnafloc 1957, a low molecular weight cationic resin was found to be as effective as ferric chloride in dewatering of sludge in post Auto-thermal Thermophilic Aerated Digestion (ATAD) and Magnafloc 1957 replaced ferric chloride due to lower health safety risk at no additional operating cost at a water reclamation plant in Bendigo, Australia (Elliott, 2006). Praestol, a cationic organic flocculant based on polyacrylamide, has been found to be effective at dosages of 1mg l^{-1} in flocculating both *Teraselmis* and *Spirulina* with 70 % recovery of biomass and no inhibitory effect on the micro-algal growth in the recycled growth medium after flocculation (Pushparaj et al., 1993).

Flocculants derived from renewable plant and animal materials could have environmental advantages over both inorganic flocculants and polyelectrolyte flocculants derived from fossil fuel. Chitosan, a cationic inorganic polymer derived from crustacean shells, has been used in the treatment of wastewater from the food industry (Harith et al., 2009). Chitosan has been shown to be effective on a wide range of freshwater micro-algae, but dosages are considerably higher than with synthetic organic flocculants at 20 to 150 mg l^{-1} (Harith et al., 2009, Molina Grima et al., 2003). Recent research at the University of Almeria has shown that chitosan was not efficient in producing flocs from *Muriellopsis,* with low biomass recovery and biomass concentrations, while requiring significantly higher dosage than synthetic polyelectrolytes (Granados et al., 2012). No efficient flocculation was observed using only chitosan for *Phaeodactylum*, but 'satisfactory' flocculation results were obtained using chitosan at a dosage of 20 mg l^{-1} if the pH was increased to 9.9 (Şirin et al., 2012). Although chitosan is considered non-toxic (Vandamme et al., 2010), there have been reports of reduced survival of oyster larvae fed chitosan flocculated micro-algae (Molina Grima et al., 2003). The costs of chitosan and the higher dosages compared to synthetic polyelectrolytes would appear to make it uneconomic for harvesting of micro-algae for biofuel production (Mohn, 1988, Vandamme et al., 2010).

Starch and modified starch can settle micro-algae (Mohn, 1988). Cationic starch is increasingly being used as an alternative to inorganic and synthetic organic flocculants in the wastewater and paper mill industries and has been found to flocculate *Scenedesmus* and *Parachlorella,* but at higher dosages than chitosan and with a large degree of variation between effectiveness of the cationic starches tested (Vandamme et al., 2010). Starch and modified starches do not appear to affect the viability of micro-algae and have been used in the treatment of drinking water (Vandamme et al., 2010). Modified starches could be more cost-effective than both inorganic and synthetic organic flocculants (Mohn, 1988, Vandamme et al., 2010), but current cationic starches are not specifically designed for micro-algae and their modification to improve micro-algal performance could dramatically increase costs (Vandamme et al., 2010).

The majority of research work on flocculation has been on freshwater algae (Uduman et al., 2010). Although many species of freshwater micro-algae can be successfully flocculated using organic cationic polymers, salinity levels above 5 g l^{-1} have been shown to inhibit flocculation (Knuckey et al., 2006, Molina Grima et al., 2003), while sea water typically has a salinity of ~35 g l^{-1} (Millero and Lepple, 1973, Speight, 2005). At high ionic strengths it is believed that polyelectrolytes tend to fold tightly and are unable to bridge between micro-algal cells to form a floc (Molina Grima et al., 2003). In marine systems the use of polyelectrolytes in conjunction with inorganic flocculants, ferric salts, alum and lime has been found to be effective (Knuckey et al., 2006, Sukenik et al., 1988), but the dosage of flocculants to flocculate marine micro-algae has been found to be 5 to 10 times higher than that for freshwater micro-algae (Knuckey et al., 2006, Uduman et al., 2010). The flocculant dosage required for the removal of 90 % of micro-algae from suspension has been found to increase linearly with salinity as expressed in ionic strength (Shelef et al., 1984b, Sukenik et al., 1988). A very high total energy usage, 14.8 kWh m^{-3}, has been reported for a suspension of *Tetraselmis*, a marine micro-alga, using a synthetic cationic polyelectrolyte polymer; a greater energy use per cubic metre than centrifugation or filtration (Danquah et al., 2009)

2.3.2.3 **Flocculation by pH and environment modification**

Flocculation of some micro-algae can be achieved by adjustment of pH (Molina Grima et al., 2003, Shelef et al., 1984a). Increasing pH to 11-12 has been shown to induce flocculation in *Chlorella* (Ras et al., 2011), but *Chlamydomonas* did not flocculate readily with the addition of alkali (Schlesinger et al., 2012). Extreme pH may cause micro-algal damage and death and could be unreliable and uneconomic on a commercial scale

(Benemann and Oswald, 1996, Lee et al., 2009). The amount of alkali required to cause flocculation of micro-algae can be lower than those normally found in high density micro-algae suspensions, greater than normally found in micro-algal growth. This may make alkaline flocculation economically viable (Schlesinger et al., 2012), but a low energy pre-concentration settlement technology is required before flocculation, adding extra complexity and cost. It is possible that flocculation could be achieved through other forms of environmental modification, such as nitrogen limitation; however the exact mechanisms behind environmental modification induced flocculation have not yet been fully investigated and more research is needed in this area (Park et al., 2011). As with extreme pH, flocculation induced by environmental modification may cause micro-algal damage and death and could be unreliable and uneconomic on a commercial scale (Benemann and Oswald, 1996, Lee et al., 2009).

2.3.2.4 **Bio-flocculation**

Bio-flocculant can be produced by bacteria and can cause the flocculation of micro-algae (Shelef et al., 1984a). Bio-flocculants produced from bacteria have been shown to be effective in the flocculation of *Chlorella* (Molina Grima et al., 2003). Bacteria have also been found able to flocculate *Pleurochrysis carterae*, but a relatively high organic carbon content in the growth medium (0.01 %) is required to grow the bacteria to flocculate the micro-algae; approximately 20 % of the carbon content in the growth media from the micro-algae (0.05 %) (Lee et al., 2009). Bacteria can make up to 30 % of the biomass in the photic zone of open waters (Sournia, 1978), and a large proportion of the mixed micro-algal biomass grown in wastewater. Many micro-algal species dominant in wastewater treatment HRAPs often form large colonies (Park et al., 2011). Effective separation of algae by sedimentation due to their incorporation into biomass flocs has been demonstrated in symbiotic algal-bacterial wastewater treatment (Medina and Neis, 2007). The use of bacteria grown on waste or wastewater could hold the possibility of a low fossil fuel input method of separating micro-algae, especially if the energy within the bacterial biomass could be recovered with that of the micro-algae. Micro-algal bacterial floc from the secondary treatment of sewage supplemented by flue gas from a coal power plant has recently been shown to settle readily, removing 97.5 % of the biomass from the growth medium within 30 minutes and producing a sediment of 2 % bacterial/micro-algal dry biomass (Van den Hende et al., 2011).

2.3.2.5 **Alternative methods of flocculation**

Electro-coagulation-flocculation using sacrificial aluminium or iron anodes has been shown to be effective at a 1-litre bench scale in the flocculation of *Chlorella* and *Phaeodactylum,* with aluminium anodes being superior to iron (Vandamme et al., 2011). Power consumption was favourable in comparison to centrifugation, at between 0.3 and 2 kWh kg^{-1} with the lowest energy consumption in salt water, suggesting that electro-coagulation may be a "particularly attractive method for harvesting of marine micro-algae" (Vandamme et al., 2011). Aluminium concentration in the micro-algal biomass and growth medium for recycling was lower than with the use of alum. Although electro-coagulation-flocculation may be a promising technology there are concerns about increased power consumption in scale-up as the distance between electrodes greatly influences power consumption (Vandamme et al., 2011).

In electrolytic flocculation non-sacrificial anodes are used and negatively charged algae move towards the anode where the negative charge is lost enabling flocs to be formed (Poelman et al., 1997). This has the advantage that flocculants are not always required, but the electrodes are prone to fouling (Uduman et al., 2010). Electrolytic flocculation has been shown to be effective at a bench scale, removing 95 % of the original micro-algae in suspension with an energy consumption of 0.3 kWh m^{-3} (Poelman et al., 1997). Ultrasound has also been found to flocculate algae, but concentration factors are lower than for other methods with a maximum increase of twenty times the feed concentration (Bosma et al., 2003). Electro-coagulation-flocculation, electrolytic flocculation and ultrasonic flocculation have been shown to flocculate micro-algae, but there are disadvantages with each method and none yet appears to have been demonstrated on a commercial scale.

Another method that could be considered for increasing of the particle size to be harvested is the use of micro-algal predators. Larger predators could consume micro-algae and be more easily harvested than the micro-algae. The conversion of plant biomass to animal biomass is inefficient, however, due to energy losses from respiration and other metabolic processes, and it appears unlikely that this method will be a viable commercial option for micro-algal biofuel.

A wide range of flocculants are available, but there is currently no single flocculant or flocculation method suitable for all types of micro-algae, and the flocculation of marine micro-algae on an industrial scale has yet to be satisfactorily resolved. Sedimentation and flocculation appeared to offer potentially the lowest energy input.

2.3.3 **Flotation**

Flotation of micro-algae can be promoted by addition of air bubbles (Singh et al., 2011). Flotation processes are classified according to the method of bubble production: dissolved air flotation, electrolytic flotation and dispersed air flotation (Shelef et al., 1984a).

Flotation can be a relatively fast technique for the harvesting of micro-algae compared to sedimentation for a number of micro-algal species (Edzwald, 1993, Oswald, 1988, Singh et al., 2011). The addition of flocculants is required in most cases for flotation to be effective (Edzwald, 1993, Mohn, 1988). Flocculation and froth flotation has been found to be effective in the removal of micro-algae from wastewater using fine air bubbles (no dimensions given) generated by a sparger with gas pressure of 3-atmospheres (Moraine et al., 1979). Flocculation flotation was found to be superior to sedimentation for the separation of a marine micro-alga, *Isochrysis galbana*, but only when large strong flocs were formed by the addition of a combination of organic and inorganic polymers (Shelef et al., 1984b). The reduced density of micro-algal flocs compared to micro-algal cells could favour flotation over sedimentation as a method of separation. The concentration of micro-algae in the separated suspension from flotation separation (7 %) is generally higher than micro-algal suspensions from sedimentation (Mohn, 1988, Oswald, 1988).

Dissolved air flotation (DAF) is a process where small bubbles are generated, with a mean size of 40 μm and ranging from 10 to 100 μm (Edzwald, 1993).

Most wastewater treatment lagoons in the USA do not harvest algae, but at plants that do, chemical coagulation followed by dissolved air flotation (DAF) is the most common method; the micro-algae removal is for purification of effluent, however, rather than for micro-algal biomass production (Christenson and Sims, 2011). DAF has been found to be effective for harvesting of micro-algae grown on pig slurry, but a high dosage of alum (0.3 g l^{-1}) was required (Goh, 1984). Unfortunately DAF although an efficient flotation option, is energy intensive due to the high pressure required (Hanotu et al., 2012).

Electro-flotation has been found to be effective at a bench scale on a range of micro-algae, but as with DAF it is energy intensive (Shelef et al., 1984a) and not the 'best choice for micro-algal recovery' (Uduman et al., 2010). Oswald (1988) suggested that it could be more useful in salt rather than fresh water. OriginOil developed a process called Quantum Fracturing™, in which pulsed electromagnetic fields and pH modification fracture the micro-algal cells with lipid floating to the surface and the remaining micro-algal biomass settling out (Gouveia, 2011), but there appears to be little independent published information on energy consumption.

Micro-bubble generation by fluidic oscillation is a technique for generating small bubbles using less energy than traditional methods, developed at the University of Sheffield (Zimmerman et al., 2009). Micro-bubbles generated by fluidic oscillation have recently been shown to be effective in the recovery of algal biomass from growth medium (Hanotu et al., 2012). Considerably more research is required, however, to establish whether an energy-efficient large-scale fluidic oscillation micro-bubble method for micro-algae harvesting is practicable.

Flotation can have high investment and operational costs and high energy usage (Mohn, 1988) especially if small bubbles are required. It has been suggested that the cost of flotation can be as great or greater than centrifugation when the cost of flocculants are included (Mohn, 1988) and a recent review concluded that there is little evidence of the technical or economic feasibility of flotation (Brennan and Owende, 2010).

2.3.4 **Filtration**

There is a wide a variety of filter designs, but membrane filters can be simply classified by pore or membrane size: macro-filtration >10 μm, micro-filtration 0.1 to 10 μm, ultra-filtration 0.02 to 0.2 μm and reverse osmosis <0.001μm. The pressure to force fluid through a membrane, and therefore the operational energy required, generally increases with reducing membrane pore size. Micro-algae are typically between 2 and 30 μm (Brennan and Owende, 2010, Edzwald, 1993, Molina Grima et al., 2003). Filtration of *Isochrysis galbana* has shown that a pore size of less than 1.5 μm is required to remove 'most' marine micro-algal cells from suspension, but on flocculation a pore size of 25 μm was found to be effective (Shelef et al., 1984b). Micro-filtration would appear to be the most appropriate filtration method for the majority of common species while macro filtration is the most appropriate for flocculated cells and larger cells.

2.3.4.1 **Micro-filtration**

Micro-filtration has been used for the recovery of micro-algal cells for aquaculture, but membrane filtration has not been widely used for producing micro-algal biomass on a large scale and could be less economic than centrifugation at commercial scale (Molina Grima et al., 2003).

2.3.4.2 **Ultra-filtration**

Ultra-filtration is a possible alternative for the recovery of very fragile cells, but has not been generally used for micro-algae (Mata et al., 2010, Molina Grima et al., 2003). Operating costs are high and maintenance costs very high (Mata et al., 2010, Purchas, 1981). It has been suggested that ultra-filtration of micro-algae will develop in a similar way to reverse osmosis for the desalination of sea water, and that the energy input of an optimised micro-algal ultrafiltration plant could be 3 kWh m^{-3}, equivalent to the lowest current energy usage in reverse osmosis desalination (Gouveia, 2011). Extracellular organic matter, however, can lead to rapid clogging of ultrafiltration membranes in the filtration of *Spirulina* (Rossi et al., 2004). An ultrafiltration membrane with 0.03 μm pore size has been used to harvest micro-algae grown on carbon dioxide emissions from a semi-conductor manufacturing plant (Avanti

Membrane Technology, Inc. private communication, 2012). Average permeate flux was 70 l m^{-2} $hour^{-1}$, but although 95 % of the micro-algae were recovered the concentration factor was only 20 and additional means of concentration are required for further processing. Energy consumption is believed to range between 1 to 3 kWh m^{-3} (Avanti Membrane Technology, Inc. private communication, 2012).

2.3.4.3 **Macro-filtration**

A wide range of macro-filtration units are available and have been used for water treatment. Many types have been used to harvest algae and found satisfactory at recovering relatively large algal cells (Molina Grima et al., 2003), but they can be hampered by low throughput and rapid clogging (Mohn, 1988, Oswald, 1988).

Vibrating screens were able to separate *Coelastrum* and *Spirulina*, although not considered to be the optimum method for *Spirulina* (Mohn, 1988). The energy cost to produce 6 % dry weight of micro-algae has been estimated at 0.4 kWh m^{-3} (Van den Hende et al., 2011).

Filter presses are an assembly of perforated, filter plates alternating with hollow frames which are compressed in a framework to form a series of filter chambers with filtered liquid (filtrate) passing through the filter medium on the filter plates and solids being retain in the filter press. They have found wide application in industry due to the simple design, flexibility and capability to handle a wide range of slurries, and have been used to reduce the number of bacteria and yeast in wine (Brennan et al., 1969, Richardson et al., 2002). Although the equipment is relatively cheap, labour costs can be high and cake washing is not always effective (Brennan et al., 1969, Richardson et al., 2002). A modified filter press with plastic diaphragms that inflate to remove the micro-algae from the filter membrane has been found to be effective in the filtration of *Scenedesmus*, but capital costs were approximately one third higher than conventional filter presses and pre-coating of the membrane with starch was required to prevent clogging (Mohn, 1988).

Rotary vacuum filters consist of a cylindrical drum covered in a filter medium. The drum is partially submerged in the liquid to be filtered, with the filtrate being drawn through the filter medium by a vacuum applied to the interior of the drum. Filters of this type are common (Brennan et al., 1969, Richardson et al., 2002) and have been used to dewater organic sludge from anaerobic digestion (Bailey and Ollis, 1977, Srinivas, 2008). *Coelastrum,* a micro-alga that forms small colonies, can be filtered to a cake containing 18 % dry weight solids without a filter pre-coat, but filtration rates fall rapidly and high energy inputs are required, with the result that this filtration method is not being recommended for micro-algal recovery (Mohn, 1988). Filter aids have also been required for filtration of *Penicillium* and *Streptomyces* mycelia by rotary vacuum filter presses (Bailey and Ollis, 1977). Vacuum belt filters can filter larger or colonial micro-algae, but investment and energy costs are very high (Mohn, 1988). Larger species of micro-algae such as *Spirulina* and *Micractinium* have been found to filter on a rotary vacuum filter with a 12 µm pore diameter yielding a 1-3 % dry weight micro-algal slurry, but smaller species of micro-algae such as *Chlorella* did not filter effectively even when the pore size was reduced to 5 µm (Goh, 1984).

Belt filters consist of two filter belts that are compressed through rollers, with water being squeezed through the belts and the solids retained on the belts for subsequently separation. They are widely used in the water treatment industry and have been suggested as suitable for separation of *Spirulina* (Mohn, 1988). Large micro-algae have been reported as readily filtered to a concentration of 18 % dry weight if the belt filter press is fed with pre-concentrated algae at 4 %, at an energy consumption of 0.5 kWh m^{-3} (Molina Grima et al., 2003). A typical three-belt filter is used by Thames Water, UK to remove sludge from an activated sludge wastewater treatment plant. The sludge suspension is first settled in a large conical settler to 0.6 % dry solids and then feed to the belt filter press together with a low dose of polyelectrolyte flocculant, and then gravity filtered to over 6 % dry solids and further dewatered in the rotary belt filter to up to 25 % (Thames Water private communication, 2012). Such a process could be envisioned for harvesting micro-algae. The price of a three belt Klampress is approximately £360,000 to process 80 m^3 $hour^{-1}$ with estimated power consumption of 17 to 21 kW (Ashbrook Simon Hartley private communications, 2009 and 2012), equivalent to an energy input of ~0.25 kWh m^{-3}.

Two extensive reviews of the filtration of micro-algae have concluded that filtration methods are suitable for micro-algae with larger cells, but inadequate to recover micro-algal species with diameters of less than 10 μm (Molina Grima et al., 2003, Uduman et al., 2010). Filter aids and flocculants would both appear to assist filtration and reduce equipment operational energy requirements; but additional materials increase costs and may need to be removed from the micro-algal biomass and the spent micro-algal growth medium. Ultrafiltration is capable of the removal of small micro-algae, but its use is limited by high energy input and the low concentration of output micro-algal suspensions. Flocculation and belt filtration has been successfully used in the water treatment industry as an effective low-cost separation method for microbial biomass, and could be a viable method for the large scale separation of micro-algae, but requires further investigation.

2.3.5 **Centrifugation**

In centrifugation, gravity is replaced as the force driving separation by a much greater centrifugal force, thus greatly reducing separation time. Almost all types of micro-algae can be separated by centrifugation (Mohn, 1988). There are many designs of centrifuge, but they can be roughly classified into three groups (Porteous, 1983):

a. Disc-stack
b. Simple bowl
c. Scroll conveyor bowl (decanter)

2.3.5.1 **Disc-stack centrifuges**

Disc-stack centrifuges are the most common industrial centrifuge and are widely used in commercial plants for high value algal products and in algal biofuel pilot plants (Molina Grima et al., 2003). A disc-stack centrifuge consists of a relatively shallow cylindrical bowl containing a number (stack) of closely spaced metal cones (discs) that rotate with the bowl (Figure 13). The mixture to be separated is fed to the centre of the stack of discs and the dense phase travels outwards on the underside of the discs while the lighter phase is displaced to the centre. The centrifugal force applied may be from 4000 to 14000 times gravitational force. Materials of different densities are thus separated into thin layers, and the narrow flow channel of 0.4 mm to 3 mm between the closely-spaced discs means that the distance materials must travel for this separation to occur is small (Mannweiler and Hoare, 1992, Perry and Chilton, 1973). Disc-stack centrifuges are ideally suited for separating particles of the size (3 -30 μm) and concentration (0.02 to 0.05 %) of algal cells in a growth medium, as shown in Figure 14. They can separate not only solid/liquid, but also liquid/liquid or liquid/liquid/solid on a continuous basis.

2.3.5.1.1 **Energy requirements for disc-stack centrifugation**

Disc-stack centrifuges generally have high energy consumption (Uduman et al., 2010). As an example, a Westfalia HSB400 disc-bowl centrifuge with intermittent self-cleaning bowl centrifugal clarifier has a maximum capacity of 95 m^3 $hour^{-1}$, but is limited to 35 m^3 $hour^{-1}$ for algae harvesting (Cawdery, D, GEA Westfalia, personal communication, 2009). The maximum power of the motor is 75 kW, but normal operating demand is probably around 50 kW, giving an energy cost for separation of 1.4 kWh m^{-3} (Cawdery, D, GEA Westfalia, personal communication, 2009). A value of 1 kWh m^{-3} has been reported for concentrating *Scenedesmus* from 0.1 to 12 % using a Westfalia self-cleaning disk stack centrifuge (Molina Grima et al., 2003) and an energy consumption of 1.4 kWh m^{-3} has been reported for the disc bowl centrifuge harvesting of micro-algae grown on pig waste (Goh, 1984). If a HSB400 centrifuge is fed with a suspension of 0.02 % dry weight of micro-algae having an oil content of 20 %, this would yield the equivalent 7 kg of dry algal material per hour and 1.4 kg of algal oil. If 90 % of the algal oil is converted to methyl ester biodiesel then 1.26 kg is produced with a calorific value of 13 kWh, assuming a net calorific value 10.33 kWh kg^{-1} (DEFRA, 2010, Milledge and Heaven, 2011). The operating energy for centrifugation is thus approximately four times the energy available in the algal biodiesel. Although this calculation is based on the data from one manufacturer, similar information for Alfa-Laval models (Ord, D., Alfa Laval, personal communication, 2009) also indicates that more energy is used in centrifugation than is available in the biodiesel produced.

This simple calculation together with other studies (Ferrell and Sarisky-Reed, 2010, Molina Grima et al., 2003) indicates the high energy usage of disc-stack centrifuges. The energy return using centrifugation could be improved by: pre-concentration using a combination of separation techniques; use of the entire algal biomass rather than just the lipid fraction for energy production; or the use of the centrifuge to eliminate other energy consuming unit operations in algal biofuel production process. Pre-concentration, by settlement or other low energy methods, to 0.5 % (algal dry weight) could improve the energy balance, but would still require 15 % of the energy in the biodiesel product for centrifugation (Milledge and Heaven, 2011). The energetic position of using a disc-stack centrifuge for the production of biofuel could also be improved by the use of the entire algal biomass (Milledge, 2010a). A kilogram of dry algal biomass containing 20 % oil would yield around 1.9 kWh of biodiesel, but the calorific value of the entire biomass is around 6 kWh (Milledge, 2010b) and the exploitation of the entire biomass could thus be a key factor in a positive energy balance in the production of biofuel (Heaven et al., 2011, Milledge, 2010a, Sialve et al., 2009, Stephenson et al., 2010).

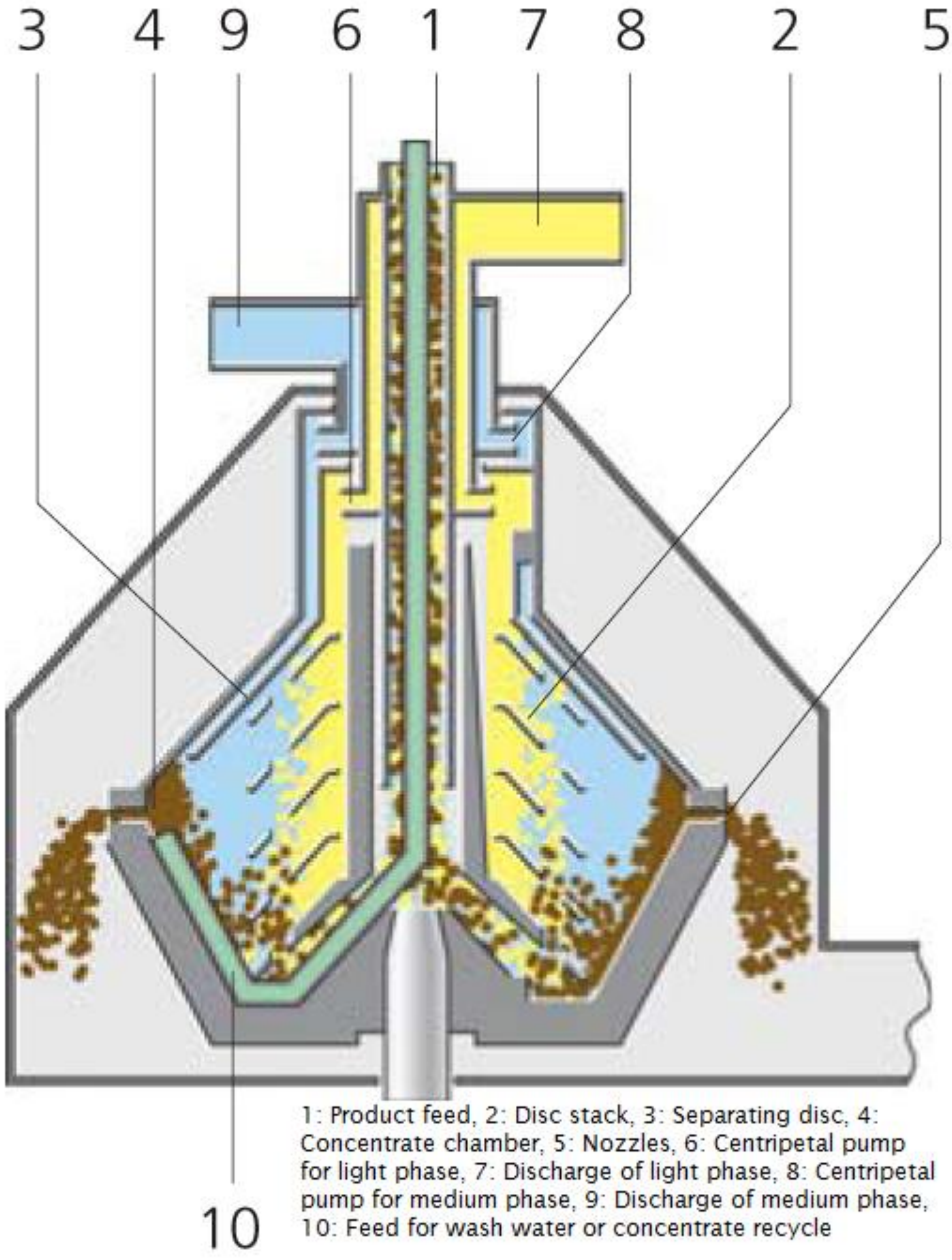


Figure 13 Disc-stack centrifuge for liquid/liquid/solid separation. Courtesy GEA Westfalia.

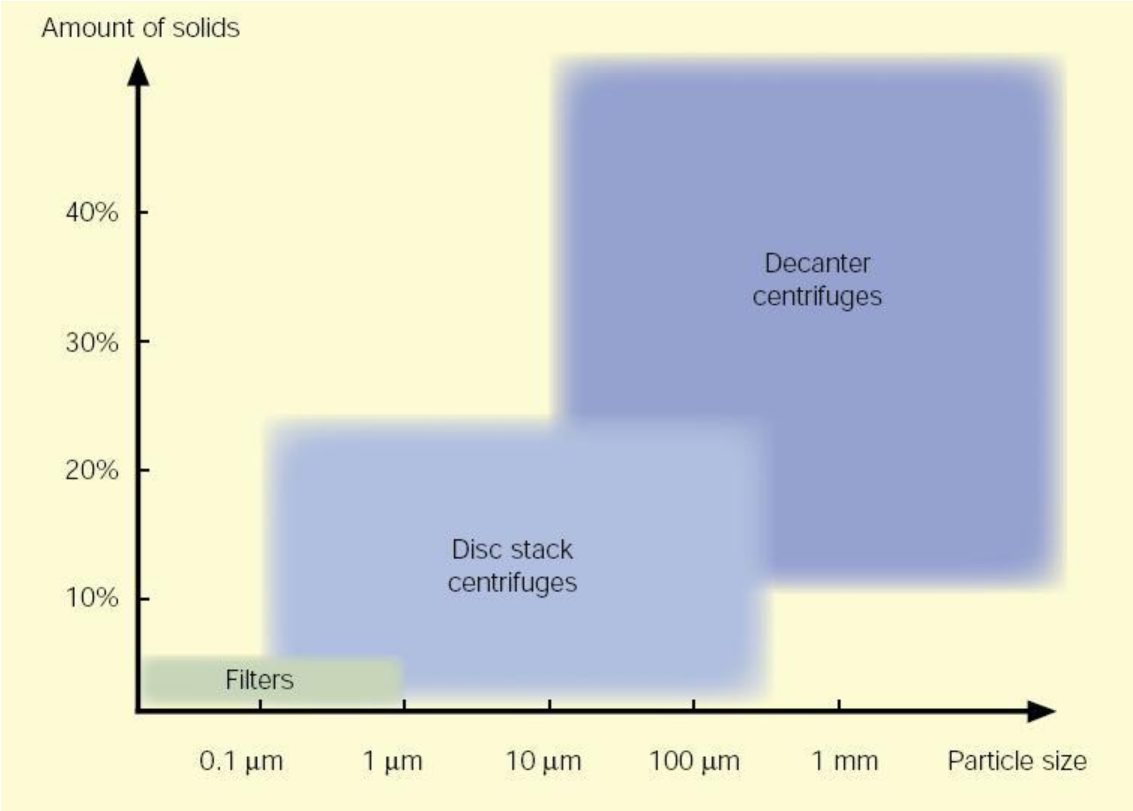


Figure 14 Centrifuge application graph; Particle sizes and concentration range. Courtesy Alfa Laval

2.3.5.2 Disc-stack centrifugation for combined cell separation and disruption

In the production of micro-algal biodiesel cells must be harvested and disrupted to release triglycerides for trans-esterification (Section 2.4.8). The breaking of cell walls can require large amounts of energy, and disc-stack centrifuges, although effective in harvesting a wide range of micro-algae, are also energy intensive. If these processes could be combined a considerable reduction could potentially be made in the overall operational energy requirements needed to produce algal biodiesel. As a result of this work the novel concept of use of a disc-stack centrifuges to rupture and harvest micro-algal cell was suggested and examined further (Milledge and Heaven, 2011)

2.3.5.2.1 Micro-eddies and algal cell disruption

Many algae are sensitive to hydrodynamic forces and cells may be damaged in mixing, pumping and gaseous transfer (Garcia Camacho et al., 2011, Hondzo et al., 1997, Joshi et al., 1996). If the hydro-mechanical forces are sufficient they can fracture cells, but lesser forces may cause reduced growth and cell death without any obvious physical damage. Although information exists on the effect of hydrodynamic forces on a wide range of bacterial, animal and plant cells in defined flow experimental systems, much less is known about the effect of hydrodynamic forces in process equipment (Chisti, 2001). It has been suggested that the micro-algal cells are damaged when the size of the micro-eddies is of the same order as or smaller than the algal cell (Molina Grima et al., 2010). Eddies with scales larger than a cell simply carry the cell from place to place, but eddies of similar size or smaller than a cell exert mechanical forces on the cell wall, and if these are greater than cell wall strength the wall is fractured (Doulah, 1977). Disruption of the cells may occur as a result of localised velocity gradients within an eddy or between eddies (Rodriguez et al., 1993), although the exact nature of the of the micro-eddy hydrodynamic forces acting on the cells; shear, compression, torsion or impact, causing cell disruption are not fully understood (Clarke et al., 2010). The size of a micro-eddy may be estimated using Kolomogrof's theory (Davidson, 2004, Molina Grima et al., 2010).

Equation 12

$$\lambda = \left(\frac{\mu}{\rho}\right)^{\frac{3}{4}} \xi^{-1/4}$$

where λ is the micro-eddy length and ξ is the energy dissipation per unit mass.

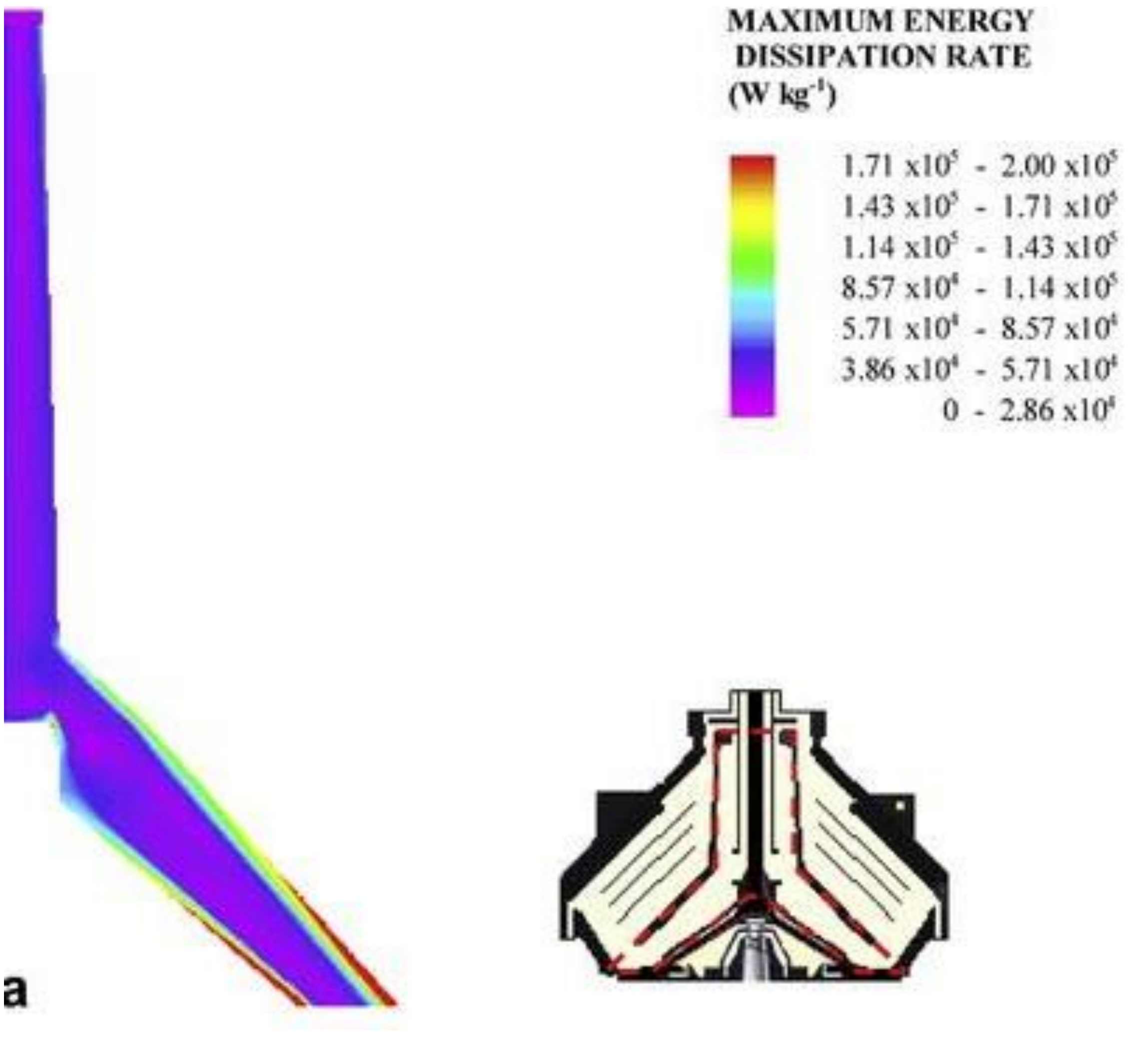
2.3.5.2.2 **Cell disruption in disc-stack centrifuges**

Damage to yeast cells has been demonstrated in disc-stack centrifuges in the brewing industry (Chlup et al., 2008). If sufficiently high hydrodynamic forces could be generated in a disc-stack centrifuge to provide cell disruption, with simultaneous lipid separation through liquid/liquid/solid separation of the type in Figure 13, considerable energy could be saved in the production of algal oil. Areas of high shear stress have been demonstrated in disc centrifuges as shown in Figure 15 (Boychyn et al., 2004)). Using Equation 12 with a maximum energy dissipation per unit of 2.00×10^5 W kg^{-1}, viscosity 9×10^{-3} Pa s and density 1115 kg m^{-3}, the minimum size of micro-eddies is estimated at 7 μm which is the size of many micro-algae. This calculation is based on one manufacturer's disc-stack centrifuge, but the similarity in the general design of disc centrifuges and similar or higher maximum hydraulic energy dissipation rates occurring in an alternative type of centrifuge (Boychyn et al., 2004) indicates that damage to algal cells could occur during disc-stack centrifugation.

Disc-stack centrifuges although suited to the separation of the particle sizes and concentrations found in micro-algal suspensions, have too high an energy consumption to be suitable for the production of algal biodiesel rather than higher value commercial algal products. The energy balance could be improved by combination with other separation methods and by the exploitation of entire biomass to produce energy.

Disc-stack centrifuges have been shown to cause cell damage to yeast, and calculation of micro-eddies sizes indicates that micro-algal cells could also be damaged. If the algal cell fracture was sufficient to liberate oil it is possible that a disc-stack centrifuge operating as a liquid/ liquid /solid separator could achieve or be designed to achieve cell destruction, oil separation and algal biomass separation in a single operation. Considerable energy could be saved by eliminating the energy requirement in the process operations of cell fracture and lipid extraction. Although this is an intriguing prospect, it is unlikely that current disc-stack centrifuges will cause sufficient algal cell disruption in a single pass. Work on yeast has shown some cell damage and viability reduction on a single pass through the centrifuge, but 9 passes were required to achieve 92.4 % decrease in cell viability (Chlup et al., 2008). If cells are fractured the smaller solid particles may also reduce centrifugation efficiency and reduced algal solids recovery as has been shown with both yeast and mammalian cells (Chlup et al., 2008, Hutchinson et al., 2006).

It would appear that current disc-stack centrifuges can cause damage to algal cells, but their use to achieve combined algal cell fracture and oil separation will require some redesign and extensive further research.



Greatest energy dissipation rates are indicated in red, while the lowest ones are in purple.

Figure 15 CFD analysis of the feed zone of a pilot disc-stack centrifuge (Boychyn, et al., 2004)

2.3.5.3 **Solid bowl and other centrifuge types**

Solid bowl centrifuges were found not to be superior to disc-bowl centrifuges when used for the recovery of micro-algae grown on pig waste (Goh, 1984). Decanter centrifuges have been found to be as effective as solid bowl centrifuges for separating micro-algae (Goh, 1984), but the energy consumption of decanter centrifuges is higher than that of disc bowl centrifuges at 8 kWh m^{-3} (Molina Grima et al., 2003). Decanter centrifuges can produce a more concentrated output than disc bowl centrifuges, but as can be seen in Figure 14 they are better suited to higher solid suspensions than those generated by micro-algal growth ponds (Brennan et al., 1969, Mohn, 1988, Perry and Chilton, 1973, Purchas, 1981). It is suggested that decanter centrifuges could be useful for the further concentration of micro-algal slurries from other harvesting methods (Goh, 1984, Molina Grima et al., 2003).

Hydro-cyclones, although relatively low energy (0.3 kWh m^{-3}) compared to other harvesting methods, are reported to be an unreliable means of concentrating micro-algae as they only achieve a maximum concentration 0.4 % with a concentration factor of 4 (Molina Grima et al., 2003). The advantage of hydro-cyclones for micro-algal separation has been given as low capital costs, but the disadvantages are that they can only process a limited number of micro-algal strains and efficiency is highly dependent on solids concentration (Origin Oil, 2010). Hydro-cyclones have been found to disrupt natural flocs of the marine micro-algae *Phaeocystis* (Veldhuis et al., 2006), and may also break up other micro-algal flocs, increasing subsequent harvesting difficulties. It would appear that if hydro-cyclones have a role in the harvesting of micro-algae it will be limited to pre-concentration of micro-algae prior to another harvesting method.

In a new type of spiral plate centrifuge, manufactured by Evodus, the suspension flows outwards in thin films over vertical plates with the solid sediment or micro-algae being forced to collect on the outer bottom edge of the vanes. Work by Evodus with the James Cook University in Australia suggests a 0.025 % suspension of *Nannochloropsis* can be harvested to a micro-algal paste of 31.5 % dry weight for an energy usage of 1.9 kWh kg^{-1} dried algae, equivalent to 34 % of the total energy within the micro-algae (Evodus private communication, 2011). This energy requirement is considerably below that calculated for the harvesting of algae by disc-stack centrifugation (Milledge and Heaven, 2011), but the discharge of the harvested algae is not continuous and current maximum throughput is limited to 4 m^3 $hour^{-1}$. Evodus report that trials are continuing in both commercial and research organisations (Evodus private communication, 2011).

Centrifuges and disc-stack centrifuges in particular are effective at removing a wide range of micro-algal cells, but at a high operational energy cost. Pre-concentration prior to centrifugation could reduce energy costs and allow centrifugation to be part of the harvesting operation for micro-algal biofuel.

2.3.6 **Materials handling**

The harvesting of micro-algae is one stage in the process of the production of micro-algal biofuel and the harvesting operation must be linked to both a growth system and a method of exploiting the energy within the micro-algal organic matter. The energy costs of moving materials between process operations could be considerable, especially for the flow of the dilute micro-algal suspension from the growth system and for the recycling of the growth media after harvesting. In an outline design developed for Pure Energy Fuels for the production of micro-algal biodiesel the energy required for the movement and recycling of material between major unit operations was estimated to be as great as, or greater than, the operational energy for the mixing and gaseous transfer in micro-algal raceway growth ponds (Section 7.1.5).

The physical properties of the micro-algal suspension vary with concentration and may influence subsequent treatment and handling. A 1 to 2 % suspension is milk-like (Oswald, 1988) with a viscosity of 1.1 to 1.7 mPa-s similar to water (Adesanya et al., 2012, Bolhouse, 2010). Algal suspensions behave as Newtonian fluids up to concentrations of between 4 and 8 % depending on algal species (Bolhouse, 2010, Wileman et al., 2012). Differences in viscosity in low concentration algal suspension (<5 %) are considered not to be ‘significant’ for the design and operation of algal growth systems (Adesanya et al., 2012).

At concentrations above about 7 % the micro-algal suspensions are non-Newtonian, exhibiting shear thinning (Adesanya et al., 2012, Wileman et al., 2012) and are described as cream-like (Oswald, 1988). The increase of viscosity to 30 mPa-s (Wileman et al., 2012) may increase the energy to pump and mix algal suspensions and non-Newtonian behaviour could contribute to handling and pump problems.

At 15-20 % micro-algal suspensions are cheese-like (Oswald, 1988) and may no longer be fluid (Greenwell et al., 2010), further increasing handling difficulties and costs.

2.3.7 **Drying**

Drying may be required prior to energy extraction as many processes, such as direct combustion, pyrolysis and current commercial biodiesel production, require dry feedstock. The removal of water from the algal biomass by evaporation can be very energy intensive. To heat and evaporate water at atmospheric pressure from a temperature of 20 °C, requires an energy input of approximately 2.6 MJ kg^{-1} or over 700 kWh m^{-3} (Mayhew and Rogers, 1972, Weast, 1985).

A variety of methods have been used to dry micro-algae prior to further processing or energy extraction: these include solar drying, roller drying, spray drying and freeze drying. Solar drying does not require fossil fuel energy, but is weather dependent and can cause considerable denaturisation of organic compounds. It is the least expensive drying option (Brennan and Owende, 2010), but large areas are required as only around 100 g of dry matter can be produced from each square metre of sun-drier surface (Oswald, 1988). Roller, spray and freeze driers have been widely used in the food industry and have all produced satisfactory result in the drying of *Dunaliella* (Molina Grima et al., 2003). Spray drying has been the preferred method of drying high value micro-algal products, but is expensive (Brennan and Owende, 2010, Molina Grima et al., 2003, Oswald, 1988) and probably uneconomic for the production of micro-algal biofuels. Although spray drying can produce a dark green powder (Oswald, 1988) it can cause significant deterioration of micro-algal pigments (Brennan and Owende, 2010, Molina Grima et al., 2003). Freeze drying tends to cause less damage to organic materials than spray drying, but is more expensive (Brennan et al., 1969) and is typically used for products such as premium instant coffee to give a better flavour than spray dried coffee. The use of freeze dying is considered too expensive for the large-scale commercial recovery of micro-algae and its use is confined to research (Molina Grima et al., 2003).

Dewatering during harvesting uses less energy than evaporation to remove water and it would appear preferable to minimise the water content of the harvested micro-algae prior to drying and to select energy extraction methods that do not necessitate drying of the micro-algae.

2.4 Energy extraction from micro-algal biomass

Energy may be extracted from micro-algae by:

a. direct combustion
b. pyrolysis
c. gasification
d. liquefaction
e. hydrogen production by biochemical processes in certain algae
f. fuel cells
g. fermentation to bioethanol
h. trans-esterification to biodiesel
i. anaerobic digestion

Table 6 Methods of energy extraction from micro-algal biomass

	Utilises entire organic biomass	Requires drying of biomass after harvesting	Primary energy product
Direct Combustion	Yes	Yes	Heat
Pyrolysis	Yes	Yes	Primarily liquid by flash pyrolysis
Gasification	Yes	Yes [b] (conventional)	Primarily Gas
Liquefaction	Yes	No	Primarily Liquid
Bio-hydrogen	Yes	No	Gas
Fuel Cells	Yes	No	Electricity
Bioethanol	No [a]	No	Liquid
Biodiesel	No	Yes [c]	Liquid
Anaerobic digestion	Yes	No	Gas

[a] Currently restricted to fermentable sugars as no large-scale commercial production of fuel bioethanol from lignocellulosic materials

[b] Supercritical water gasification (SCWG) an alternative gasification technology can convert high moisture biomass

[c] No current commercial process for the wet trans-esterification of wet micro-algal biomass

Table 6 summarises the energy outputs (heat, electricity, liquid, gaseous or solid fuel) of the various potential methods of producing useful energy from micro-algae together with the need for drying the biomass after harvesting and the capacity to exploit the entire biomass.

2.4.1 **Direct combustion**

Direct combustion is, historically and currently, the main method by which biomass energy is utilised (Demirbas, 2001), but does not appear to have been greatly explored for the production of energy from micro-algae. Biomass direct combustion can provide heat or steam for household and industrial uses or for the production of electricity (Demirbas, 2001). Many industries devote a considerable amount of energy to the production of steam, with the pulp and paper industry using 81 % of its total energy consumption for this purpose (Saidur et al., 2011). The lumber pulp and paper industry uses biomass to provide 60 % of its energy needs (Demirbas, 2001). The efficiency of biomass direct combustion for the production of electrical energy is between 20 to 40 % with the highest efficiencies being achieved in large plants of 100 MW and in the co-combustion of biomass and coal (Demirbas, 2001, McKendry, 2002). The co-combustion of biomass in coal fired plants is considered an especially attractive option for biomass exploitation due to its higher efficiencies (Demirbas, 2001, McKendry, 2002, Saidur et al., 2011). The co-generation of heat and electricity can significantly improve the economics of biomass combustion, but requires that there is a local demand for heat (Demirbas, 2001). The co-firing of a power plant with micro-algae grown using the carbon dioxide emissions of the power plant appears conceptually to be an elegant method of reducing the GHG emissions per unit of electrical power generated by the plant, but unfortunately may not be practicable. A Life Cycle Assessment of co-firing of coal and solar dried micro-algae found that, although GHG emissions and air acidification could be reduced by co-firing with micro-algae grown on the power plant emissions, the depletion of natural resources and eutrophication potential increased and the use of micro-algae is "obviously more expensive than coal" (Kadam, 2002).

The moisture content of biomass can reduce the heat available compared to that from dry biomass by 20 % (Demirbas, 2001) and the direct combustion of biomass is feasible only for biomass with a moisture content of less than 50 % (McKendry, 2002, Varfolomeev and Wasserman, 2011). Large biomass plants can be as efficient as fossil fuel plants, but the higher moisture content of biomass results in additional costs (Demirbas, 2001). Micro-algal biomass will need considerable further drying after harvesting before it could be used for direct combustion. Generally high moisture content biomass is considered better suited to biological conversion, by anaerobic digestion or fermentation, to other more readily useable fuels (McKendry, 2002).

Ash content can be a considerable problem in direct combustion of biomass due to fouling of the boilers restricting the use of high ash content biomass (Demirbas, 2001). Wood has a typical ash content of 0.5 to 2 % (Misra et al., 1993, Saidur et al., 2011), but the ash content of micro-algae can be high: *Spirulina* having an ash content 7 to 10 % (Tokusoglu and Unal, 2003), diatoms containing 9 to 35 % ash due to the silica outer wall (Brown and Jeffrey, 1995) and *Pleurochrysis carterae* a coccolithophorid, containing 10 % calcium carbonate (Moheimani and Borowitzka, 2006, Moheimani, 2005).

The fine particulate nature of dry micro-algae may be advantageous in co-combustion with pulverised coal as it will not require additional particle size reduction. Non-micro-algal biomass particles are normally much bigger than pulverised coal and the amount of energy required for the grinding of the biomass to a diameter of less than 1 mm (2-3 % of the heating value) is almost double that compared to the energy required for coal pulverisation (0.9-1.2 % of the heating value). The energy requirements for biomass pulverisation increase significantly (>20 % of the heating value) for fibrous and or moist biomass (Belosevic, 2010). The combustion of biomass can generate fine particle emissions which can be harmful to health, with the amount, type and quantity of particle emissions being influenced by biomass, combustion technology and emission control equipment (Sippula, 2010). If micro-algal biomass is to be a used in direct combustion extensive research will be required to optimise combustion, co-combustion and reduce emissions.

Using direct combustion of micro-algae it is possible to produce both heat and electrical energy; however, the problem of drying the micro-algae together with challenges of ash and emission control may make the direct combustion of micro-algae impractical on a commercial scale.

2.4.2 **Pyrolysis**

Pyrolysis is the thermal decomposition of the organic component of dry biomass by heating in the absence of air, producing, as its primary product, a hydro-carbon rich liquid (bio-oil or bio crude) (McKendry, 2002, Saidur et al., 2011). The exploitation of pyrolysis products dates back to ancient times with the ancient Egyptians using the pyrolysis of wood to produce tars for use in embalming (Demirbas, 2001). Pyrolysis can produce high volumes of fuel relative to biomass feed and the process can be modified to favour the production of bio-oil, syngas or solid char (Miao et al., 2004). Bio-oil is perhaps a more attractive end product than char or syngas as it has a higher energy density and is easily transport and stored (Jena and Das, 2011).

2.4.2.1 **Methods of pyrolysis**

Pyrolysis processes can be classified by temperature and process time as; slow, fast and flash (Ghasemi et al., 2012) . Slow pyrolysis results in higher yields of char rather than the liquid or gaseous products (Brennan and Owende, 2010, Ghasemi et al., 2012). Fast pyrolysis is considered a better process than slow pyrolysis (Ghasemi et al., 2012), with higher temperatures and the capability of achieving greater liquid product and gas yields of around 70- 80 %, compared to 15 to 65 % for slow pyrolysis (Brennan and Owende, 2010, Varfolomeev and Wasserman, 2011). The properties of bio-oil from fast pyrolysis have also been reported to be more suitable for refining to liquid fuels (Miao et al., 2004). Flash pyrolysis covers a range of newer technologies with temperatures above 500 ˚C and short residence times of a few seconds or less (Ghasemi et al., 2012, McKendry, 2002). These are considered as having future potential for the commercial production of biofuel from biomass (Brennan and Owende, 2010). The optimum pyrolysis reaction range for *Chlorella* has been found to be close to the conditions found in flash pyrolysis (Bhola et al., 2011). A new experimental microwave assisted method of pyrolysis has been laboratory tested on *Chlorella* yielding up to 22 % bio-oil (Du et al., 2011).

2.4.2.2 **Pyrolysis of micro-algae**

The pyrolysis of dry *Chlorella* has been found to give higher yields and better quality bio-oil (higher calorific value) than from macro-algal or moss biomass.

(Demirbas, 2010). The maximum bio-oil yield from pyrolysis of dry *Chlorella* was found to be between 50.8 and 57.9 % of the weight of the original dry biomass (Ozkurt, 2009) and (Demirbas, 2010, Miao and Wu, 2004, Miao et al., 2004, Ozkurt, 2009).The Higher Heating Values (HHV) of *Chlorella* and the micro-algal bio-oil were 23.6 MJ kg^{-1} and 39.7 MJ kg^{-1} respectively (Demirbas, 2010). Therefore the yield of bio-oil energy is 85 % of the initial energy in the micro-algal biomass in the conversion of 50.8 % w/w of biomass to bio-oil. The lipid content of micro-algae is believed to influence the energy balance of pyrolysis with higher lipid content micro-algae having an improved energy balance and producing bio-oil that requires less refining (Bhola et al., 2011).

It is suggested that the energy required to produce bio-oil by pyrolysis from micro-algae would be similar to the narrow range 200-400 kJ kg^{-1} reported in the published literature for a 'diverse' range of dry biomass feedstocks (Maddi et al., 2011). An energy input for pyrolysis has been quoted as 1.3 to 2.7 % of the calorific value of the micro-algae, but no estimate of the energy to dry the micro-algae, which can be very significant, was given. The major challenge for pyrolysis of micro-algal biomass is that it requires dry biomass. When drying of algal biomass was included pyrolysis used more energy than was produced as usable solid, liquid and gaseous fuels (Jena and Das, 2011). The Energy Consumption Ratio for the production of bio-oil from micro-algae, a ratio of the energy input for thermochemical treatment to the energy in the bio-oil, was found to be 0.44 to 0.63 for hydrothermal liquefaction and 0.92 to 1.24 pyrolysis due to the requirement for the moisture to be evaporated prior to pyrolysis (Vardon et al., 2012). Indicating again that pyrolysis can use as much, or more, energy than is generated as biofuels.

2.4.2.3 **Refining of pyrolysis bio-oil**

The bio-oils from pyrolysis are normally highly oxygenated complex mixtures of organic compounds that results in a mixture than can be polar, viscous, corrosive, unstable and unsuitable for use in conventional fuel engines unless refined (Peng et al., 2000). Bio-oil from micro-algae pyrolysis has been reported to have a lower oxygen content and viscosity and higher heating value than bio-oil from wood or other terrestrial plant material (Du et al., 2011, Miao and Wu, 2004, Miao et al., 2004), and could be more suitable for refining into liquid fuels. However bio-oil from both macro-algae and micro-algae may contain nitrogen compounds that bring additional fuel refining costs (Maddi et al., 2011). It is probable that bio-oil from micro-algae will need to be refined requiring additional energy input prior to the production of a readily useable energy source. There may be a difference between the optimum process for energy return from pyrolysis alone and for a process that produces the best energy return after refining. The use of a catalyst (sodium carbonate) during pyrolysis has been suggested to upgrade bio-oil from pyrolysis, reducing or eliminating the need for further refining and has been found to improve the bio-oil quality, reducing acidity and oxygenation, from the pyrolysis of *Chlorella* without a reduction in yield (Babich et al., 2011).

Although pyrolysis is carried out at atmospheric pressure and is a well-established and 'simpler' process than hydrothermal liquefaction (Babich et al., 2011), the ability of hydrothermal liquefaction to use wet biomass would appear to give it an advantage over pyrolysis. As with direct combustion the need to dry the micro-algae prior to pyrolysis may preclude it as an energetically and economically viable method of producing bioenergy.

2.4.3 **Gasification**

Gasification is the conversion of organic matter by partial oxidation at high temperature (800 -1000 °C) mainly into a combustible gas mixture (syngas) (Demirbas, 2001, McKendry, 2002, Saidur et al., 2011). The syngas has a calorific value of 4-6 MJ m^{-3}, around half that of natural gas (McKendry, 2002), and is a mixture of hydrogen (30-40 %), carbon monoxide (20-30 %) methane (10-15 %), ethylene (1 %), nitrogen and water vapour (Demirbas, 2001, Saidur et al., 2011). The gas can be burnt to produce heat or converted to electricity in combined gas turbine systems that can achieve an electric energy output of 50 % of the heating value of the incoming gas (Demirbas, 2001, McKendry, 2002). Conventional biomass gasification processes require dry feedstock (Guan et al., 2012a), but supercritical water gasification (SCWG) is an alternative gasification technology for the conversion of high moisture biomass and it is suggested it can be net energy positive in a well-engineered systems (Guan et al., 2012b). The enthalpy change needed to take ambient liquid water to a low-density supercritical state (400 °C and 250 bar) is similar to that required to vaporise liquid water at ambient temperature, but the advantage of the SCWG process is that much of the energy invested in reaching a supercritical state can be captured and used again, with the hot effluent from the gasification reactor being used to preheat the wet biomass feed stream (Guan et al., 2012a).

The syngas from gasification can be used to produce methanol and hydrogen as a fuel for transport and other uses (McKendry, 2002, Saidur et al., 2011). Methanol from the gasification of biomass could also be used as a renewable alternative to methanol derived from fossil fuel for the production of biodiesel (including micro-algal biodiesel). This would make the biodiesel eligible for renewable fuel credits such as the Renewable Fuel Obligation Certificates (ROC) in the UK. ROCs may be grated for electricity generated from biodiesel and glycerol manufactured from biomass and biomass-derived alcohols, but if the fossil derived methanol is used in the production of the biodiesel it is ineligible for ROCs (Office of Gas and Electricity Markets, 2009).

2.4.3.1 **Gasification of micro-algae**

Experimental laboratory studies of the steam gasification of dry *Chlorella,* macro-algal and moss biomass found yields of syngas were higher for the micro-algae than for both moss and macro-algal biomass, with maximum syngas yields of 40.6 % of the weight of the original biomass and a hydrogen gas content of up to 48.7 % by volume (Demirbas, 2010). The syngas yield increased with increasing temperature from 302 to 652 ˚C, in agreement with a recent model of the kinetics of supercritical water gasification that indicate that higher temperatures favour production of intermediates which are more easily gasified together with the production of gas at the expense of char (Guan et al., 2012b). Unfortunately insufficient data was presented by Guan et al. (2012) to calculate the energy yield of the syngas and the energy inputs into the process.

Gasification of *Spirulina* to a methane-rich syngas using supercritical water and ruthenium catalysts has been predicted to yield up to 60-70 % of the heating value contained in the algal biomass (Stucki et al., 2009). Experimental studies on supercritical water gasification at 550 ˚C of *Nannochloropsis* found energy conversion of biomass to syngas of up to 60 % (Guan et al., 2012a). A theoretical study of the production of methanol from *Spirulina,* using the maximum theoretical yield of 0.64 g methanol g^{-1} dry micro-algal biomass, gave a ratio of produced energy to required energy of 1.1, with the gasification and methanol synthesis process using some 25 % of the total process energy (Hirano et al., 1998). As the energy content of methanol at 23 MJ kg^{-1} is similar to that of micro-algae this would equate to an energy conversion of 60-70 %. Although the energy balance is slightly positive, the evaluation assumes the maximum theoretical yield and that wet micro-algal biomass with 79 % moisture content can be successfully treated using gasification. Using a novel method of catalytic gasification of wet *Chlorella* biomass (87 % water), at lower temperature (350 ˚C) and higher pressure (18 MPa) than conventional gasification, up to 70 % of maximum theoretical syngas was produced (Minowa and Sawayama, 1999). The study also attempted a brief energy evaluation comparing gasification to direct combustion of micro-algae. Although the energy evaluation showed gasification to be superior to direct combustion the assumptions made included: the maximum theoretical yield of syngas, recycling of nutrient only in gasification and a halving of the calorific value of algal biomass for combustion, but no similar allowance for combustion of the syngas. The energy for gasification was given as 5.95 MJ kg^{-1} of dry cells, or approximately 28 % of total energy in the original biomass. It has been suggested for every 4.5 J of energy in the syngas gas, 1.0 J of unrecovered heat energy is required for supercritical water gasification (Guan et al., 2012a), equivalent to an energy input of 22 % of the syngas or 21 % of the calorific value of the original algal biomass.

A recent review has concluded that there is little data available on the gasification of micro-algae and in particular on the energy balance and the need for drying of micro-algae prior to gasification (Brennan and Owende, 2010). This work would endorse this conclusion. If gasification of micro-algae can be achieved with wet biomass it would possibly become more economic and energetically attractive.

The economics and energy balance of gasification could also be improved by recycling of nutrients in the micro-algal biomass for the growth of new micro-algae. It has been found that nitrogen recovered from the aqueous phase after gasification can be used as part of a medium to successfully grow micro-algae (Minowa et al., 1995).

Combustible gas can also be produced from wet micro-algal biomass by anaerobic digestion (Section 2.4.9) at much lower temperatures than gasification. Both gasification and anaerobic digestion have been suggested as promising methods for exploiting bioenergy from biomass in India (Singh and Gu, 2010). Anaerobic digestion of algal residues, however, has been shown to have a higher net energy return and much lower GHG emissions than gasification (Delrue et al., 2012). Although gasification is generally a more rapid process than anaerobic digestion it would appear than the energy input needed to achieve the temperatures required will make it uncompetitive in terms of energy ratio with anaerobic digestion, unless a much higher yield of combustible gas can be achieved than from anaerobic digestion.

2.4.4 **Liquefaction and hydro-thermal upgrading**

Liquefaction is a low temperature high pressure process where biomass is converted into a stable liquid hydrocarbon fuel (bio-oil) in the presence of a catalyst and hydrogen (Demirbas, 2001, McKendry, 2002). In hydro-thermal upgrading the biomass is converted to partially oxygenated hydrocarbons at high pressure in the presence of a catalyst in a wet environment (Demirbas, 2001, McKendry, 2002). In practice it would appear that the terms liquefaction, hydro-liquefaction and hydro-thermal liquefaction are used for processes where wet biomass is converted to bio-oil by temperature and pressure in the presence of a catalyst, with and without the presence of gaseous hydrogen. Reviews of thermal treatments of biofuel have concluded that commercial interest in liquefaction is low due to the more complex feed systems and higher costs than pyrolysis and gasification (Demirbas, 2001, McKendry, 2002); but hydro-thermal upgrading has the advantage of the conversion taking place in an aqueous environment and drying of biomass after harvesting may not be required prior to liquefaction and hydro-thermal upgrading (Brown et al., 2010, Minowa et al., 1995, Sawayama et al., 1999). The ability of hydrothermal treatments to handle wet biomass make them some of the most interesting methods of producing biofuel from micro-algae (Torri et al., 2012). *Dunaliella* with a moisture content of over 78 % has been treated by hydro-thermal upgrading (termed liquefaction by the authors) on laboratory scale (20 g of wet algae) yielding 37 % oil based on the volatile solids content of the micro-algal biomass (Minowa et al., 1995), but hydrothermal liquation of biomass with a moisture content above 90 % is believed to have an unfavourable energy balance (Vardon et al., 2012).

Hydrothermal carbonisation is a process in which biomass is heated in water under pressure to create char rather than liquid products. In an experimental study of various micro-algae a char with an energy content similar to bituminous coal and containing 55 % of the carbon from the original biomass was produced by hydrothermal carbonisation (Heilmann et al., 2010). The hydrothermal carbonisation process requires a 10 % solids concentration and drying of algal biomass may not be required prior to conversion. The hydrothermal carbonisation process can produce char with a calorific value of 12.01 MJ from 1kg of dry algal biomass, but to heat a system contain 1kg of dry algal biomass and 9 kg of water from ambient to 203 °C will require, 7.31 MJ. With insulation and temperature control, however, no significant additional energy was needed to maintain reaction temperature for 2 hours (Heilmann et al., 2010). It is suggested that hydrothermal carbonisation gives a better return on energy investment than direct combustion as the micro-algae do not need to be dried after harvesting and heat recovery employed in any industrial process would result in additional energy improvement. Lack of production of char is generally considered to be an advantage of hydrothermal treatment as bio-oil is a more useful product to both char and syngas (Jena and Das, 2011), and the application of hydrothermal carbonisation for biofuel production may therefore be more limited than hydrothermal liquefaction.

Although biomass liquefaction has been extensive researched, micro-algae have seldom been studied despite the fact that they should decompose and hydrolysed more easily than biomass containing lignin (Yang et al., 2011a). The cell wall of *Desmodesmus* has been found to be resistant to hydrothermal liquefaction (Alba et al., 2012). The yield of bio-oil from the liquefaction of *Dunaliella* has been reported to be as much as 87 % of the original micro-algal volatile solids (VS) on a weight basis (Yang et al., 2011a), but this appears to be unrealistically high. The calculated energy is over 140 % of that in the original biomass and there appears to be over 24 % more carbon in bio-oil than in the original biomass. This may be due to a simple error in assessment methods and calculations, or may be due to a reaction between the algal biomass and the large quantity of ethanol (9 times the weight of wet biomass) used in the liquefaction. The maximum bio-oil yield from the liquefaction of *Microcystis* has been reported as 33 % of the weight of biomass VS and 40 % of the energy in the micro-algal biomass. (Yang et al., 2004). Bio-oil yield from the liquefaction of wet *Nannochloropsis* (79 % moisture content) has been found to be a maximum of 43 % of the biomass dry weight with 80 % recovery of the carbon and 90 % of the energy in the *Nannochloropsis* organic material (Brown et al., 2010). Bio-oil yields from hydrothermal liquefaction, as percentage mass of original dry micro-algal biomass, have been reported as: up to 41 % for *Spirulina* (Jena and Das, 2011), between 24 and 45 % for *Scenedesmus* (Vardon et al., 2012) and up to 49 % for *Desmodesmus* or 75 % recovery of the energy in the micro-algal biomass as bio-oil (Alba et al., 2012).

The energy needed to heat the wet algal biomass to operating temperature for liquefaction has been estimated at between 65 and 85 % of the total energy available in the bio-oil produced (Minowa et al., 1995). A more recent estimate reported the energy required for hydrothermal liquefaction as a percentage of bio-oil energy produced at between 44 and 63 % for micro-algal biomass with a moisture content of 80 % (Vardon et al., 2012). Although the liquefaction of micro-algal biomass can be a net energy producer and the bio-oil has a high calorific value and can be readily refined into a variety of liquid fuels, it would appear that a complete process for the production of micro-algal biofuel from liquefaction using existing micro-algal growth and harvesting techniques may use more energy than is produced.

The ability of liquefaction to use wet biomass and to convert the vast majority of the chemical energy into readily refined liquid fuels may make it worthy of further study, but the heat energy required for the process is the major challenge. The enthalpy of compressed water at 200 °C and 15.55 bar, the least energetic conditions found in the literature (Brown et al., 2010), is 852 kJ kg^{-1} (Mayhew and Rogers, 1972) an increase of around 768 kJ kg^{-1} from water at 20°C and atmospheric pressure. As outlined in the review of harvesting a moisture content of 75 % appears to be the minimum achievable by current techniques without drying. A kilogram of micro-algal slurry with 75 %

moisture content would contain 0.25 kg of dry micro-algal biomass: if this algal biomass had 20 % lipid content its calorific value would be approximately 1.5 kWh. The enthalpy change from atmospheric temperature and pressure to the minimum requirements for micro-algal liquefaction (200 ˚C and 15.55 bar) would be approximately 0.16 kWh or under 11 % of calorific value of the micro-algal slurry. The enthalpy change of the dry algal biomass is not known, but if it is assumed to be similar to that of water the total energy required to increase the enthalpy of the entire slurry would still be below 15 % of the calorific value of the algal biomass. It may therefore be possible to considerably reduce the energy requirements of liquefaction and liquefaction may be viable if the energy input can be reduced to closer to minimum theoretical input.

The hydro-thermal liquefaction of micro-algae can produce bio-oil of similar or higher calorific value to that from pyrolysis, but the chemical composition is different and hydrothermal liquefied oil can have higher viscosity and percentage of higher boiling point compounds (Jena and Das, 2011, Vardon et al., 2012). As with bio-oil from pyrolysis, bio-oil from hydrothermal liquefaction will probably need further refining to produce a commercial useable biofuel. The lower quantity of low boiling point compounds in hydrothermal bio-oil will make it less desirable for light fuel applications (Vardon et al., 2012).

The recycling of nutrients particularly nitrogen and phosphorus, could reduce the net energy inputs for micro-algal biofuel production, and the nitrogen and phosphorus dissolved in the aqueous phase from hydrothermal liquefaction is believed to be capable of reuse as a growth medium for micro-algae (Alba et al., 2012)

Liquefaction and hydro-thermal upgrading can handle wet biomass eliminating the need for drying after harvesting, but the process is more complex and has higher costs than pyrolysis and gasification. Large amounts of energy are required to heat and compress the wet biomass and processes that require lower temperatures such as anaerobic digestion (Section 2.4.9) will have a lower energy input and potentially higher return on energy investment.

2.4.5 **Bio-hydrogen**

Hydrogen is considered a particularly attractive replacement for fossil fuel as it combustion produces water vapour rather than greenhouse gases. Fuel cells and other technologies are commercially available to exploit hydrogen, and vehicles using hydrogen as fuel are already in operation, for example Honda's FCX. Although, hydrogen is believed to be beginning to move from a "fuel of the future to an energy carrier of the present" (Benemann, 2000), the major challenge remains producing renewable hydrogen at an affordable and competitive cost remains.

The production of hydrogen by cyanobacteria has been known since the late 19th century (Benemann, 2000). Gaffon is generally credited with the first scientific study of bio-hydrogen from micro-algae in research on *Scenedesmus* in the late 1930s and early 1940s (Benemann, 2000, Kruse et al., 2005, Varfolomeev and Wasserman, 2011). A variety of metabolic pathways for the production of bio-hydrogen have been studied (Kruse and Hankamer, 2010, Ljunggren, 2011, McKinlay and Harwood, 2010) with hydrogenase, the enzyme mainly responsible for hydrogen production, being widely found in both prokaryotic organisms and eukaryotic plants and with all five major taxonomic groups of cyanobacteria containing hydrogenase genes (Kruse and Hankamer, 2010).

Considerable research on bio-hydrogen has been carried out since the 1970s, and over a hundred million dollars had been spent on research up to 2000, but with "little progress toward the goal of a practical and commercial process" (Benemann, 2000). Research has been continued since 2000, but yields of energy from bio-hydrogen production systems are low at 0.3 % up to 1.3 % of the total light energy arriving at the surface of the reactor and 5 % is required for an economically viable system (Kruse and Hankamer, 2010). It is has been suggested, however, that the maximum practical efficiency of conversion of solar energy is 1 %, with cyanobacteria only able to achieve this conversion rate for a short period in a pure argon atmosphere, and outdoor systems achieve an average of only 0.05 % (Sorensen, 2012). The considerable challenges of oxygen inhibition and scale-up also still need to be overcome (Kruse and Hankamer, 2010, McKinlay and Harwood, 2010, Varfolomeev and Wasserman, 2011). Substantial research effort appears to be being directed at the genetic modification of micro-algae (Gressel, 2008, McKinlay and Harwood, 2010, Varfolomeev and Wasserman, 2011), but although this may overcome the challenges of yield and oxygen inhibition it may produce fresh issues of containment and public perception.

A major advantage of bio-hydrogen production is that hydrogen does not accumulate in the culture (Ghasemi et al., 2012) and harvesting and energy extraction costs could be reduced; but despite extensive research a commercially viable production appears to be some way off.

2.4.6 **Micro-algal fuel cells**

In microbial fuel cells (MFCs) electrical current is generated from oxidation-reduction reactions that occur within living microorganisms with the oxidation of an organic compound at the anode generating electrons that produce an electric current (De Schamphelaire and Verstraete, 2009, Powell et al., 2011). Micro-algae have been used as a source of organic material for bacterial oxidation at the anode. With marine plankton as a substrate 80 % of organic carbon was removed in a microbial fuel cell (De Schamphelaire and Verstraete, 2009). Using *Chlorella* as a substrate around 60 % of the chemical oxygen demand was removed, but conversion of chemical energy to electrical energy was low at between 10 and 25 % (Velasquez-Orta et al., 2009). The electrical yield from microbial fuel cells has been quoted at 2.5 kWh kg^{-1} of dry *Chlorella* biomass (Velasquez-Orta et al., 2009), but this appears to be over stated by a factor of 10 due to the use of an incorrect conversion factor of 2.77 kWh per MJ rather than 0.277 kWh per MJ (Perry and Chilton, 1973). However when the data presented is corrected, the energy produced from dry micro-algal biomass by fuel a cell is approximately a quarter of that achieved by both direct combustion and anaerobic digestion. The maximum power generation of microbial fuel cells oxidising biomass is currently only up to 1 W m^{-2} (Howe, 2012, Thorne et al., 2011) and could only produce the power equivalent to 3.8 tonnes ha^{-1} (Howe, 2012) considerable below that anticipated from growth of micro-algae for the production of biodiesel.

A recent development in microbial fuel cells is a photo-microbial fuel cell or bio-photovoltaic fuel cell where photosynthetic algae growing at the anode generate the electrons, thereby removing the need for an organic substrate (Howe, 2012, Thorne et al., 2011). Bio-photovoltaic fuel cells currently have very low efficiency and energy production is between 1 and 1.5 orders of magnitude lower than microbial fuel cells oxidising biomass (Howe, 2012).

Photosynthetic micro-algae can also act as electron acceptors at the cathode (Powell et al., 2011). A power density 0.95 mW m^{-2} was achieved in a coupled microbial fuel cell, with *Chlorella* growing at the cathode and yeast growing heterotrophically on glucose as an electron donor (Powell et al., 2011): this is considerable below the best output of MFCs. In a unique design combining anaerobic digestion and a coupled microbial fuel cell, bacteria growing on the waste from the anaerobic digestion of micro-algae biomass act as the electron donor, with micro-algae, being grown for the biomass for anaerobic digestion, acting as the electron acceptor, but power output is low at an average of 12 mW m^{-3} (De Schamphelaire and Verstraete, 2009).

The energy output of microbial fuel cells can be low and is currently many times lower when growing micro-algae are used either as an electron donor or acceptor. It is clear that there is a need for a considerable improvement in micro-algal fuel cell efficiencies before they can be considered as a commercial option for exploiting micro-algae for biofuel. There will also be considerable problems in the scale-up of MFCs, in particular those requiring the growth of photosynthetically active micro-algae (Rosenbaum et al., 2010). Micro-algal fuels cells, if they have a future in the production of micro-algal biofuels, may be limited to exploiting addition energy production opportunities generated by their use in conjunction with other methods of micro-algal biofuel production, principally anaerobic digestion and fermentation where micro-algal fuel cells may have the capability of giving incremental energy output gains.

2.4.7 **Bioethanol**

First generation bioethanol, such as that produce from corn in the USA and sugarcane ethanol in Brazil, is now widely produced and used (Yang et al., 2011b) and there is considerable interest in producing second generation bioethanol from cellulosic biomass (Balat et al., 2008). Bioethanol can be readily used in current technology, with 86 % of cars sold in Brazil in 2008 capable of using ethanol or a mixture of ethanol and fossil fuel petroleum (Walker, 2010). Bioethanol accounted for more than 94 % of global biofuel production in 2008, with the majority coming from sugarcane (Balat et al., 2008). Bioethanol has been suggested as having a better development potential than conventional biodiesel (Lee, 2011). However, there are disadvantages with bioethanol; "lower energy density than gasoline, corrosiveness, low flame luminosity, lower vapour pressure (making cold starts difficult), miscibility with water, and toxicity to ecosystems" (Balat et al., 2008). The energy balance of corn ethanol is probably marginal (Beal, 2011) and it has been suggested that "at present, bioethanol produced from sugarcane in Brazil is the only credible example of a biofuel that exhibits a significant net energy gain" (Walker, 2010). The growth of crops such as sugarcane and corn for the production of sugar for bioethanol will be considerably constrained, as these compete directly with food production.

In grain crops about half of the above ground biomass, straw, is ‘wasted’ and worldwide 2 billion tonnes of cereal straw are produced annually (Gressel, 2008) and there is now considerable interest in exploiting straw and other lignocellulosic materials for ethanol production (Balat et al., 2008). In sugar and ethanol production from sugarcane considerable quantities of bagasse are produced which can be burnt to produce heat to distil bioethanol, but there are concerns about the environmental effects of this and it may be beneficial to convert bagasse to bioethanol (Gressel, 2008). The total potential worldwide bioethanol production from crop residues and wasted crops has been estimated at 491 billion litres year^{-1}, about 16 times higher than the current world bioethanol production (Balat et al., 2008). Cellulosic ethanol was expected to play a large role in meeting the goals of the US Energy Independence and Security Act of 2007 for renewable biofuels (Ferrell and Sarisky-Reed, 2010); but despite extensive research and the availability of low cost lignocellulosic biomass there is, as yet, no large-scale commercial production of fuel bioethanol from lignocellulosic materials (Balat et al., 2008). One of the problems encountered with production of bioethanol from straw is that biodegradation of hemicelluloses and cellulose by cellulases can be inhibited by lignin, found in many terrestrial sources of second generation biofuel biomass (Gressel, 2008). Micro-algae do not normally contain significant quantities of lignin and therefore hold out the prospect of the cellulosic components of micro-algae being more readily converted to sugars by cellulases.

2.4.7.1 Micro-algal bioethanol

Micro-algae can contain significant quantities of carbohydrates and proteins that can be converted to bioethanol via fermentation (Harun et al., 2010) with *Chlorella* containing up to 50 % w/w of starch under favourable growth conditions (Doucha and Lívanský, 2009). Other micro-algae are also known to contain up to 50 % w/w of carbohydrates that can be fermented to bioethanol (Singh and Olsen, 2011). The yield of ethanol from the fermentation of micro-algal biomass has been found to be up to 38 % of the dry micro-algal biomass (Harun et al., 2010). An advantage of using fermentation of micro-algae could be that wet biomass could be used, but a recent study has found that the sugar released from fermentation of dried biomass was 55 % higher than that from wet micro-algal biomass (Miranda et al., 2012). If the micro-algal biomass needs to be dried prior to conversion to fermentable sugars for bioethanol production it is likely that bioethanol production will not be energy efficient or economic.

Ethanol yield has been found to be improved by the removal of lipid from micro-algae prior to fermentation (Harun et al., 2010), raising the prospect of bioethanol production being combined with biodiesel production.

In addition to simple carbohydrates and sugars within the cell, more complex carbohydrate associated with the cell wall will need to be broken down into fermentable sugar if the entire micro-algal biomass is to be exploited for bioethanol (Harun et al., 2011b). Most micro-algal species have cell walls based on cellulose (Harun and Danquah, 2011), however there is considerable diversity in algal structural polysaccharides. Many algae lack cellulose and have other polymers that provide structure to the cell while some lack cell walls entirely (Ferrell and Sarisky-Reed, 2010, Tarchevsky and Marchenko, 1991).

Cell wall disruption is considered essential to release the carbohydrates and sugars to maximise bioethanol yields. A sugar extraction efficiency of 96 % has been achieved by acid hydrolysis of dried *Scenedesmus* biomass (Miranda et al., 2012). Ultrasonic cell disruption followed by enzymatic saccharification released 64 %, of the dry biomass of *Chlorococcum,* as glucose that could be fermented to ethanol (Harun and Danquah, 2011). Enzyme pre-treatment of *Chlamydomonas,* liquefaction by amylase followed by enzymatic saccharification, yielded 23.5 % w/w ethanol after fermentation (Choi et al., 2010). The alkaline pre-treatment of cells to release fermentable sugars increased the ethanol production from *Chlorococcum* biomass containing 33 % carbohydrate, with a maximum yield of 26 % w/w of the dry biomass (Harun et al., 2011b).

Some micro-algae contain cellulases and it may be possible to recover these and other industrial enzymes from the micro-algal biomass (Ferrell and Sarisky-Reed, 2010). A possible process could be to use extracted micro-algal enzymes to produce fermentable sugars from micro-algal biomass for bioethanol production, but although elegant it will require considerably more research and is probably currently uneconomic.

Like yeast, some micro-algae such as *Chlorella* and *Chlamydomonas* are capable of producing ethanol and other alcohols through heterotrophic fermentation (Ferrell and Sarisky-Reed, 2010), but micro-algae are probably of more interest as feedstock rather than as biological means of converting biomass to ethanol as yeast fermentation is an established and extensively researched process.

Continuous fermentation of glucose can produce a yield of ethanol of 51 % w/w (McGhee et al., 1982) an energy conversion of 98 % (glucose HHV 15.6 kJ g^{-1} and ethanol HHV 29.8 kJ g^{-1}). Reported yields of ethanol from disrupted micro-algal cells range 23.5 to 38 % w/w with the higher figure giving a maximum energy yield of 57 % assuming a HHV of 5.5 kWh kg^{-1} for low lipid content micro-algae (Milledge, 2010b). The challenge of producing bioethanol from micro-algae, as with lignocellulosic biomass, will be to convert the entire organic micro-algal biomass to fermentable sugars economically and energy efficiently. There appears to be considerably less research on the production of bioethanol than biodiesel, but bioethanol has been suggested as one of three top targets for future micro-algal biofuel production (Gouveia, 2011). The fermentation of micro-algae has the potential advantage of exploiting the entire biomass, if an economic method of producing fermentable sugars from complex organic material is found. However there are considerable quantities of complex waste organic matter from agriculture that could potentially provide a lower cost feedstock for fermentation than purpose grown micro-algae.

2.4.8 **Biodiesel and trans-esterification**

"Biodiesel is a fuel that is obtained from a manufacturing process that converts plant oils or animal fats together with alcohol into a fuel that can be used in an internal combustion engine" (Office of Gas and Electricity Markets, 2009). Chemically it is the alkyl esters of fatty acids which are produced by trans-esterification of triglycerides of fatty acids using an alcohol, normally methanol or ethanol (Knothe et al., 2005).

The higher lipid content of some micro-algae has focused much of the published research work on the production of biodiesel from the micro-algal lipids via trans-esterification. The most often quoted and long running study by the NREL (Sheehan et al., 1998) focused almost entirely on biodiesel with relatively little discussion of alternative methods of exploiting the energy within the micro-algal biomass.

There are a number of challenges for the production of micro-algal biodiesel:

a. In conventional commercial trans-esterification processes the biomass needs to be dry (Hidalgo et al., 2013)
b. The cell wall may require disruption to release the lipid which is energetically demanding (de Boer et al., 2012).
c. Lipid needs to be extracted from micro-algal biomass by solvents and other methods (Rawat et al., 2013).

Drying of the biomass prior to oil extraction can use considerable energy and can be the main energy input in the production of biodiesel (de Boer et al., 2012). Wet solvent extraction of lipid from micro-algae biomass which eliminates the need for drying and potentially reducing energy input may be possible, but has yet to be proven at industrial scale (de Boer et al., 2012, Lardon et al., 2009, Sills et al., 2012), but for algal biofuels to yield net gains in energy lipid extraction methods for wet biomass must be developed (Delrue et al., 2012, Sills et al., 2012).

Intact cell walls hamper lipid recovery and the most effective methods of recovery are from disrupted algal cells (Greenwell et al., 2010). Mechanical pressing is the industry standard for oil recovery from oilseeds for both food and biofuel production, but it is ineffective for micro-algae (de Boer et al., 2012). A number of cell disruption techniques have been applied to micro-algae, but mechanical disruption is generally considered preferable to chemical disruption as it avoids chemical contamination and preserves the functionality of the cell contents (Chisti and Moo Young, 1986). The breaking of cell walls can require large amounts of energy, and can be achieved by ultrasound, milling, autoclaving or homogenisation (Mata et al., 2010). Homogenisation can be very efficient, with between 77 and 96 % of algal cells ruptured per pass (GEA Process Engineering, 2011), but to homogenise 10 l of algal suspension with algal cell concentrations between 100 and 200 g l^{-1} requires 1.5-2.0 kWh (Greenwell et al., 2010) or 0.75 to 2 kWh kg^{-1} of algal cells disrupted. It has been suggested that cell disruption and subsequent oil extraction represent the largest energy input in the production of micro-algal biodiesel (Razon and Tan, 2011). If cell disruption processes could be combined with algal harvesting then a considerable reductions could be made in operation energy requirements (Section 2.3.5.2).

The high energy demands from micro-algal biomass drying, cell disruption and lipids extraction have led to interest in in-situ or direct trans-esterification, where the biomass is directly in contact with the alcohol and catalyst (Hidalgo et al., 2013, Velasquez-Orta et al., 2012), thus reducing the number of unit operations, simplifying the process and potentially reducing energy inputs (Rawat et al., 2013, Velasquez-Orta et al., 2012). Direct trans-esterification of oilseeds using an alkaline catalyst has a high tolerance for water (Velasquez-Orta et al., 2012), but increasing the biomass water content decreases trans-esterification efficiency (Hidalgo et al., 2013). The amount of methanol required for biodiesel production by in-situ trans-esterification is ‘extremely high' and will need to be reduced for direct trans-esterification to become economic (Velasquez-Orta et al., 2012). In-situ trans-esterification also faces energetic hurdles, due to the large volumes of water, solvents or reactant that need to be evaporated from the biomass, and these must be overcome if it is to be energetically viable (de Boer et al., 2012).

Although the lipid content of micro-algae can be high this is not the case for all species and generally the production of lipids, as an energy storage compounds, occurs under nutrient stress where the growth rate is reduced. The NREL and others (Bhola et al., 2011, Illman et al., 2000) have shown a considerable reductions in algal yield under nutrient stress condition that promote high lipid content. Despite the higher lipid content , the actual lipid yield can be lower under nutrient stress than in nutrient replete conditions due to a much lower growth rate (Liu et al., 2013, Sheehan et al., 1998). Unfortunately not all micro-algal lipids are suitable for conversion to biodiesel by trans-esterification (Chisti, 2007). The presence of lipids other than triglyceride may require energy intensive pre-treatment steps before the alkaline trans-esterification (de Boer et al., 2012).

A considerable number of Life Cycle Assessments (LCAs) have been carried out on the production of biodiesel and it has been concluded that the process may be marginal in terms of energy balance, global warming potential (GWP) and economics. Only in the best case scenarios was algal biodiesel found to be comparable to first generation biodiesel and algal biodiesel was not "really competitive under current feasibility assumptions" (Lardon et al., 2009). A reworking of the data from 6 LCAs, in what was termed a Meta-model of Algae Bio-Energy Life cycles (MABEL), found that the energy return on energy invested (EROI) ranged from one, no return on the energy invested to two, twice the energy invested (Liu et al., 2011). A recent extensive review and LCA using a Monte Carlo approach to estimate ranges of expected values found that nearly half of all the LCA results had an EROI of less than one (Figure 16)(Sills et al., 2012). The Sills' (2012) study also showed that methane from anaerobic digestion of defatted algae is required for net gains in energy and must be an integral part of algal biodiesel production process to yield EROI values that are greater than one.

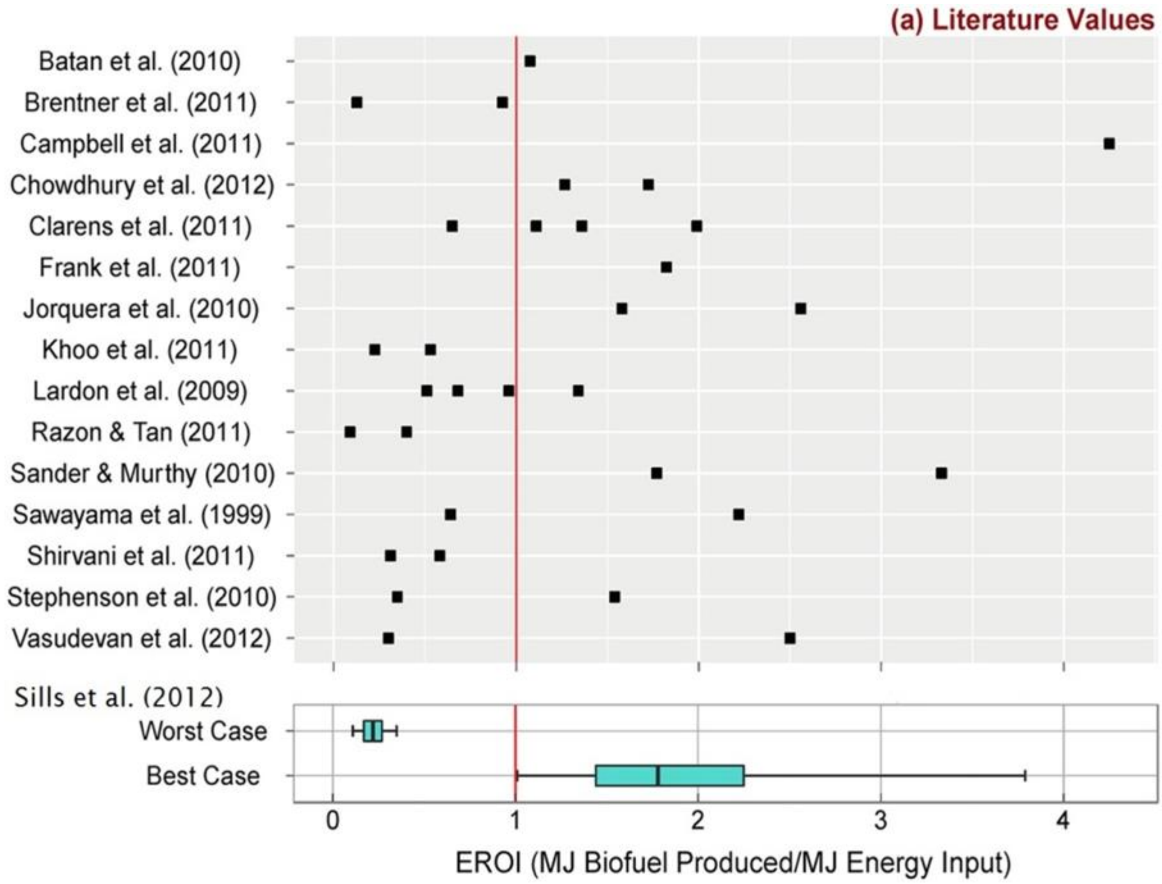


Figure 16 Energy Return on Energy Invested (EROI) values from published LCA studies (Sills et al., 2012)

The anaerobic digestion of micro-algal biomass to produce biogas following lipid removal for biodiesel production has been suggested in order to reduce the cost of biodiesel production by over 40 % due to the use of biogas to power parts of the micro-algal biofuel process (Harun et al., 2011a). The partial extraction of energy from the algae in the form of biodiesel does not appear to be energetically or commercially viable on its own and anaerobic digestion appears vital to any efficient and economic process for producing biodiesel from algae (Delrue et al., 2012, Milledge, 2010a, Stephenson et al., 2010, Zamalloa et al., 2011). A study has shown that when the lipid content is below 40 % the anaerobic digestion of the entire biomass without lipid extraction may be the optimal strategy for energy recovery (Sialve et al., 2009). The Sialve (2009) study contains errors and if lipid is digestible then energetically it is always better to digest algae (Heaven et al., 2011).

Micro-algal biodiesel production has been shown, in many studies, to produce negative net energy output and where there is a positive output it depends on technology that is not available on industrial scale and/or the exploitation of the defatted biomass to produce biogas from anaerobic digestion. Anaerobic digestion of the entire algal biomass is thus more energy efficient as it utilises the entire wet algal biomass and can exploit algae with a wide range of lipid contents (Milledge, 2010a).

2.4.9 Anaerobic digestion

The advantages of producing biogas from the anaerobic digestion of micro-algae are that wet biomass can be used and there is the potential to exploit the entire organic biomass for energy production. Considerable research has been carried out on the anaerobic digestion of a variety of organic materials and some of the earliest studies on extracting bioenergy from micro-algae examined anaerobic digestion (Golueke et al., 1957). However relatively few studies have been carried on the anaerobic digestion of freshwater micro-algae and almost none on marine micro-algae (Gonzalez-Fernandez et al., 2012a, Zamalloa et al., 2011). Micro-algae have been successfully digested to produce methane at a concentration of 1 % dry weight, but higher concentrations are considered more practicable (Oswald, 1988). Anaerobic digestion may also allow the recycling of nutrients back to the micro-algal growth system (Singh and Olsen, 2011), reducing costs and embodied energy inputs, but concerns have been expressed about pigmentation of micro-algal digestate inhibiting light penetration if it is used in growth ponds (Oswald, 1988).

The theoretical yield of biogas, calculated from the chemical composition of micro-algae using the "Buswell equation" (Buswell and Mueller, 1952, Symons and Buswell, 1933), can be high. The proportions of carbohydrates, proteins and lipids affect the potential of micro-algae as a substrate for anaerobic digestion (Park and Li, 2012) with lipid yielding higher volumes of biogas per gram of feed material than both carbohydrate and protein (Heaven et al., 2011, Weiland, 2010, Zamalloa et al., 2011). Practical yields from the anaerobic digestion of micro-algae are considerably below the theoretical maximum. The destruction of organic volatile solids from micro-algae was found to be only 60 to 70 % of that found in raw sewage (Golueke et al., 1957). Methane yields from the anaerobic digestion of micro-algae have been reported in the range of 0.09 to 0.34 l g^{-1} of volatile solids (Gonzalez-Fernandez et al., 2012a, Zamalloa et al., 2011). There is considerable conjecture about the reason for the relatively low practical methane yields compared to the theoretical values. Is it; the cell wall protecting the contents of the cells from digestion, the relative high amount of cell wall to contents, the nature of the cell wall or cell contents?

The low methane production rates from anaerobic digestion have been attributed to the resistance of the micro-algal cell wall to digestion even after death (Zamalloa et al., 2011). Micro-algal cells walls typically make up 13-15 % of the weight of the cell, but some species may contain up to 40 % (Tarchevsky and Marchenko, 1991). The largest variation of cell wall composition is found in micro-algae, as if "nature decided to conduct a vast experiment with algae to select from the numerous polysaccharides the one that suits a cell wall best" (Tarchevsky and Marchenko, 1991). The degree of polymerisation of cellulose and the diameter of cellulose fibres is higher in algae than bacteria and terrestrial plants (Klemm et al., 2005). Could the nature of micro-algal cellulose be a factor in low biogas yields? Others have suggested glycoproteins in the cell wall may be a possible factor in poor biogas yield from micro-algae as these are highly resistant to bacterial degradation. (Banks & Heaven private communication, 2012). The cell walls of micro-algae have been shown in many cases not to be a simple micro-fibre cellulose structure, but a complex structure comprised of several distinct layers, some of which form a highly ordered crystalline lattice (Roberts, 1974).

The tough cell wall of some species of micro-algae may also prevent the contents being digested to produce biogas and the low conversion of micro-algae to methane has been attributed to the resistance of intact micro-algal cells to bacterial invasion and destruction (Golueke et al., 1957) Rupturing the cell wall by thermal, chemical and mechanical methods prior to anaerobic digestion has been shown to improve methane yields (Park et al., 2011). Thermal pre-treatment of *Scenedesmus* biomass doubled its anaerobic biodegradability (Gonzalez-Fernandez et al., 2012b). However the energy required for breaking the cell wall will negatively impact on the energy balance unless more energy is released as additional methane than is used to fracture the cells.

The bacteria involved in the production of methane by anaerobic digestion are sensitive to the chemical composition of the feedstock in particular the carbon to nitrogen ratio of the substrates (Gonzalez-Fernandez et al., 2012a, Park et al., 2011, Samson and LeDuy, 1983). The low methane yield has been attributed to ammonia toxicity derived from high concentrations of protein found in many micro-algae (Golueke and Oswald, 1959, Park and Li, 2012, Samson and LeDuy, 1983). The co-digestion of organic nitrogen rich micro-algae with low nitrogen/ high carbon substrates, such as sewage sludge or glycerol, has been found to produce a synergistic effect with methane yields higher than from either substrate (Gonzalez-Fernandez et al., 2012a, Samson and LeDuy, 1983). Methane yields of double that of micro-algal biomass alone have been achieved by co-digestion with low nitrogen wastes (Samson and LeDuy, 1983).

One of the advantages of growing micro-algae for biofuel is that many species can be grown in salt water. Low salt concentrations can stimulate microbial growth, but high salt concentrations ($\geq$10 gl^{-1}) are known to inhibit anaerobic systems through an increase of osmotic pressure or dehydration of methanogenic micro-organisms (Hierholtzer and Akunna, 2012, Lefebvre and Moletta, 2006). The toxicity of salt is predominantly determined by the sodium cation and other light metal ions, such as potassium, have also been found to be toxic to methanogens at high levels (Chen et al., 2008). An optimal sodium concentration for mesophilic methanogens in waste treatment processes of 230 mg Na l^{-1} has been recommended (Chen et al., 2003). Mesophilic methanogenic activity is halved at 14 g Na l^{-1} (Chen et al., 2003, Ramakrishnan et al., 1998), the approximate level of sodium found in sea water (El-Dessouky and Ettouney, 2002). Anaerobic digesters can be acclimatised to higher salt levels if they are continuously exposed to gradually increasing salt concentration rather than salt shock (Lefebvre and Moletta, 2006). Adaptation of methanogens to high concentrations of sodium over prolonged periods of time can allow the anaerobic digestion of high salt concentration wet biomass with the sodium concentration to halve methanogenic activity increasing to 37.4 g Na l^{-1} after acclimation (Chen et al., 2003). It may therefore be possible to produce biogas from micro-algae grown in sea water and research is being undertaken at the University of Southampton on the effect of salt concentration on the anaerobic digestion of various micro-algae.

Micro-algal biomass has shown its potential for the production of various biofuels, although it is clear that there are significant technologically hurdles to be overcome before micro-algal biofuel is energetically and commercially viable. It is probably too early, at the current stage of biofuel development, to definitively select which method or combinations of methods for exploiting energy from micro-algae will be commercial exploited. However anaerobic digestion is relatively simple, has the potential to exploit the entire organic carbon content of micro-algae and can utilise wet biomass. It is likely to play a leading role in combination with other methods and could be the major method of biofuel production from micro-algae.

2.4.10 **Co-production and biorefineries**

Current commercially viable exploitation of micro-algae products is limited to products other than fuel, and the immediate future for the commercialisation of micro-algae may be with non-fuel products (Milledge, 2011a, Milledge, 2012a). However the lessons learned from non-fuel products, together with their potential for co-production with fuel, may lead to the more rapid commercial realisation of micro-algal biofuel. Micro-algal co-products "have potential to provide a ‘bridge’ while the economics of algal biofuels improve" (Hannon et al., 2010).

The cultivation of micro-algae simply for biofuels may not be currently profitable and the micro-algal industry must take advantage of markets for additional high-value products such as ‘nutraceuticals’, pigments and vitamins (Hannon et al., 2010, Milledge, 2010a, Milledge, 2012b). Co-production of micro-algal bioenergy with high-value products is currently more economically viable than the production of just micro-algal biofuel alone (Jonker and Faaij, 2013, Subhadra and Grinson, 2011). A recent economic model of the production of algal biofuel found that oil for biofuel production could represent a relatively small portion of algae related revenue opportunities (Brown, 2009).

The term 'biorefinery' has been used in the literature since the 1980s, and refers to the co-production of a spectrum of high value bio-based products (food, feed, nutraceuticals, pharmaceutical and chemicals) and energy (fuels, power, heat) from biomass (Gonzalez-Delgado and Kafarov, 2011, Olguin, 2012, Taylor, 2008, Wageningen University, 2011). The biorefinery concept is an 'emerging research field' (Rawat et al., 2013) and in December 2009 the US Department of Energy announced a US$ 100 million grant for three organisations to research algae biorefineries (Singh and Ahluwalia, 2013).

Biorefineries could allow the exploitation of the entire micro-algal biomass. *Dunaliella salina* is grown for the production of β-carotene (Milledge, 2011a) and is also a source of glycerol for potential use as biofuel and a green chemical feedstock. A recent study has concluded, however, that the production of glycerol for use as biofuel would be currently uneconomic without high value co-products (Harvey et al., 2012). The growth of *Dunaliella* could provide the biomass for a biorefinery. Laboratory studies suggest that *Dunaliella tertiolecta* could also be potentially used as a source; of high value lipids; extracellular polysaccharide, for polymer production; and glucose for bioethanol production (Geun Goo et al., 2013). This type of biorefinery, that produces a variety of products from a single biomass source, may be termed a vertical biorefinery (Milledge, 2013). Although biorefineries could improve the economics of biofuel production (Pires et al., 2012) they are likely to be energy intensive (Olguin, 2012, Rawat et al., 2013), and will involve increased energy inputs, process complexity and possibly reduce energy outputs. A biorefinery plant should operate sustainably with its energy met by biofuels produced (Cherubini, 2010). Despite increasing interest, however, it has yet to be established whether micro-algal biorefineries can produce more energy than is required by the processes within them.

Although high value algal products may allow the commercialisation of algae in the short term, the immense potential scale of algal fuel production could result in the creation of such large quantities of algal non-fuel materials that the market price is dramatically reduced. Therefore, this study will concentrate on the maximisation of fuel production and minimisation of operational energy inputs.

2.5 Conclusions

There is a wide range of combinations of growth, harvesting and energy extraction unit operations that could form a micro-algal biofuel production system, but as yet there is no successful economically viable commercial system producing biofuel. The literature review has shown that the overwhelming balance of the published information indicates that the energy inputs and costs of producing micro-algal biomass in open systems are lower than PBRs. One of the drawbacks of open systems is the difficulty of species control and contamination. The algal species that currently form the vast bulk of commercially-grown micro-algal biomass (i.e. *Chlorella, Spirulina and Dunaliella*) are grown in highly selective environments, which means that they can be grown in open air cultures and still remain relatively free of contamination by other algae. Another approach is to grow mixed micro-algal cultures that grow naturally in the local environment, recently referred to as "synthetic ecology" (Kazamia et al., 2012) or 'grow what grows'. This latter approach may not be suited to the production of biodiesel that exploits only triglycerides found in the micro-algae, as "synthetic ecology" may not yield high volumes of lipid; but may be a viable process when coupled to an energy extraction process, such as anaerobic digestion, that can potentially exploit the majority of organic materials in micro-algal biomass. A recent meeting organised by InCrops and EnAlgae in 2012 in Bury St Edmunds, titled "AD and Algae", highlighted the dearth of information on the anaerobic digestion of micro-algae, but also noted the complete lack of information on full process design and energy balances. Therefore this research will concentrate on assessment of the energy balance for micro-algal biofuel processes based on open raceway systems, with biogas production from anaerobic digestion of micro-algal biomass as the preferred fuel conversion technology.

Harvesting of micro-algal biomass is a critical issue in the development of a commercially viable process for production of micro-algal biofuel due to the dilute nature of micro-algal suspension. The optimum post-harvest micro-algal concentration for anaerobic digestion may not be the optimum for the overall process. The research will examine changes in harvesting method and the degree of micro-algal concentration, or concentration factor, and their effects not only on harvesting energy requirements, but also on the downstream energy requirements for transfer and recycling together with the mixing and heat energy required by an anaerobic digester.

3. Software evaluation

Before commencing on the development of a tool for the assessment of energy balances, commercially available software packages were assessed for their potential suitability for this purpose. When flow-sheets consist of more than a few unit operations, using purpose-built software may be an effective solution in the production of mass and energy balances (Gosling, 2005). Specialist process flow-sheet software could allow the development not only of a mass and energy balance, but of a dynamic model that could describe the 'behaviour' of the system over time. A previous review of process simulation and modelling for industrial bioprocessing suggested that Aspen (Aspen Technology Inc., Burlington, MA, USA and SuperPro (Intelligen, Scotch Plain, NJ, USA) are capable of producing mass and energy balances for flow-sheet simulations containing both continuous and batch processes (Gosling, 2005). UNISIM from Honeywell (Process Solutions, Honeywell, Phoenix, AZ, USA) has been used to evaluate 15 integrated processes including biodiesel and micro-algae biomass production (Monteiro et al., 2010). It was concluded that SuperPro, Aspen, UNISIM together with Life Cycle Assessment (LCA) Software, which have been widely used for GHG and environmental impact assessment of algal biofuel, should be evaluated for their ability to produce a dynamic mass and energy balance for algal biofuel flow-sheets.

3.1.1 FP7 All-Gas project

The FP7 All-Gas project aims to produce biogas from the anaerobic digestion of algal biomass grown in open high rate ponds using nutrients from wastewater. Carbon dioxide is supplied from the combustion of local agricultural biomass with a proportion provided from upgrading or combustion of the micro-algal biogas (Banks et al., 2011a). The carbon needs of the system may also be partly supplied from pre-treatment of the wastewater by anaerobic digestion and the growth of bacteria on the residual organic carbon entering the micro-algal growth ponds. The process is a mixture of continuous and discontinuous (batch) processes. At the heart of the process are the open high rate algal growth ponds (HRAPs), with the outputs (algal biomass and oxygen) and required inputs (N, P and C) controlled by the photosynthetic growth and respiration of the algae. The photosynthetic growth of the algae will, in nutrient-replete conditions (N, P and C), be controlled by the level of solar insolation and temperature and therefore will vary throughout the day and year and will be very different between night and day and winter and summer. Preliminary flow-sheets and mass balances (Figure 17) had been developed for the process by Aqualia SA and the University of Southampton and were used as the basis for software evaluation. The development of a dynamic model of the process flow-sheet, with respect to time, might allow; the identification and resolution of potential 'conflicts' between unit operations; the optimisation of the process; and the production of a comprehensive energy balance. It was decided to use the All-Gas flow-sheets as a test case for the suitability of the software.

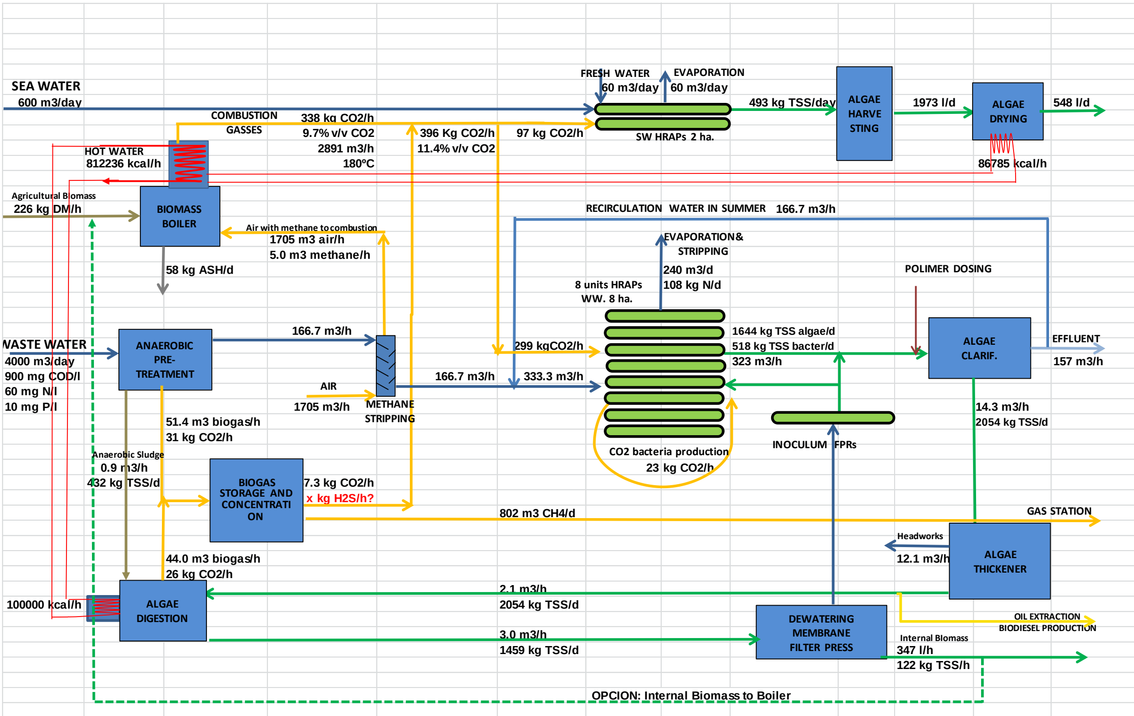


Figure 17 All-Gas initial flow-sheet. Courtesy of Aqualia SA.

3.1.2 Life Cycle Assessment software

There have been a number Life Cycle Assessments (LCAs) on algal biofuels production (Liu et al., 2011, Sills et al., 2012, Stephenson et al., 2010). SimaPro (SimaPro, Carshalton, UK) has been used to carry out a net energy analysis for the production of algal biogas (Razon and Tan, 2011) and is the most widely used LCA software (Simapro UK, 2011). Unfortunately, it is not suitable for a mass balance, but it was suggested that Umberto (IFU Hamburg GmbH, Germany) may be capable of producing a dynamic mass balance (SimaPro private communication, 2011). On further investigation, however, it was established that while Umberto could be capable of producing a combined material and energy balance of the entire process system, it cannot provide a 'live' or dynamic model (IFU Hamburg private communication, 2011).

LCAs depend on modelling assumptions and this is a problem with a micro-algal biofuel system where there is a lack of long-term full-scale operational data. Although it is important to distinguish between modelling assumptions and modelling errors (Clarens et al., 2011b) the assumptions used in many LCAs have been and can be challenged leading to a considerable degree of uncertainty in the validity of LCAs.

Pfromm et al. (2011) has suggested that algal biofuel LCAs focus on materials rather than processes. They are essentially an inventory that lack the rigorous checks on data consistency offered by an engineering mass balance and the expansion of LCAs to energy balances therefore cannot succeed (Pfromm et al., 2011). Although LCAs are an accepted methodology for assessing the potential of algal energy production systems (Razon and Tan, 2011), it appears that current LCA software is unlikely to produce a rigorous dynamic mass and energy balance model.

3.1.3 **SuperPro software**

SuperPro software is a flow-sheet modelling program that is claimed by the manufacture, Intelligen, as facilitating modelling, evaluation and optimisation of integrated processes in a wide range of industries including wastewater treatment. The University of Southampton has had some past experience of SuperPro and the evaluation of software for the production of a dynamic model for the FP7 All-Gas micro-algal biogas production began with SuperPro. An evaluation copy was downloaded from Intelligen, but was limited to only two unit operations. To evaluate SuperPro software a simple flow-sheet for the separation of micro-algal cells was used consisting of a settlement tank followed by a centrifuge.

An aspect of the SuperPro software is that it allows cells together with intra-cellular and extra-cellular contents to be defined. The ability to model cells was thought to be valuable in subsequent unit operations, such as oil extraction, where cells may be disrupted and cell contents released and in particular allow the tracking of nutrients (N & P) for recycling. For the evaluation the parameters for the micro-algae were set to those of yeast, the nearest of the standard inputs to micro-algae, although cell size was increased. Problems were encountered with the software with the cell contents function, however; the algal suspension input was set with 0 % extra cellular content, but the output to the settler was changed to 100 % extra-cellular content, despite it being highly unlikely that there is complete cell disruption of algal or yeast cells within a settlement tank. It was recommended by Intelligen not to use the cell function (Intelligen private communication, 2011).

The examination of SuperPro confirmed a previous review (Gosling, 2005) that, although SuperPro can produce mass balances for both continuous and batch processes, it is unable to produce a dynamic model. Intelligen confirmed that SuperPro cannot develop a fully dynamic model, but suggested that a dynamic model would be overkill for an algal biofuel process and that such a model would be difficult to interpret and make work reliably (Intelligen private communication, 2011). The Joint Bioenergy Institute in California (a US DOE Research Centre) is reported to be using SuperPro for multiple models of a micro-algal production process, rather than one dynamic model, to assess daily and seasonal variations in a batch reactor.

SuperPro can produce excellent graphics and could be used to produce multiple mass balances of an algal process for night and day and a variety of seasons, but it was unable to develop a dynamic model and further investigation of the SuperPro software was halted.

3.1.4 UNISIM software

A copy of UNISIM Design was provided by Honeywell, Bracknell, Berkshire. It is described as intuitive and interactive process modelling software that enables the creation of steady-state and dynamic models for plant design, performance monitoring, troubleshooting and operational improvement and uses unit operation models mainly from the oil and gas refining and chemical process industries (Honeywell, 2011).

A section of the proposed FP7 All-Gas flow-sheet consisting of the methane stripper, biomass boiler and algal high rate growth pond (HRAP) or raceway was selected, and an outline model was developed using UNISIM for evaluation of the software (Figure 18). Discussions were then held with Honeywell to explore the applicability and limitations of the model.

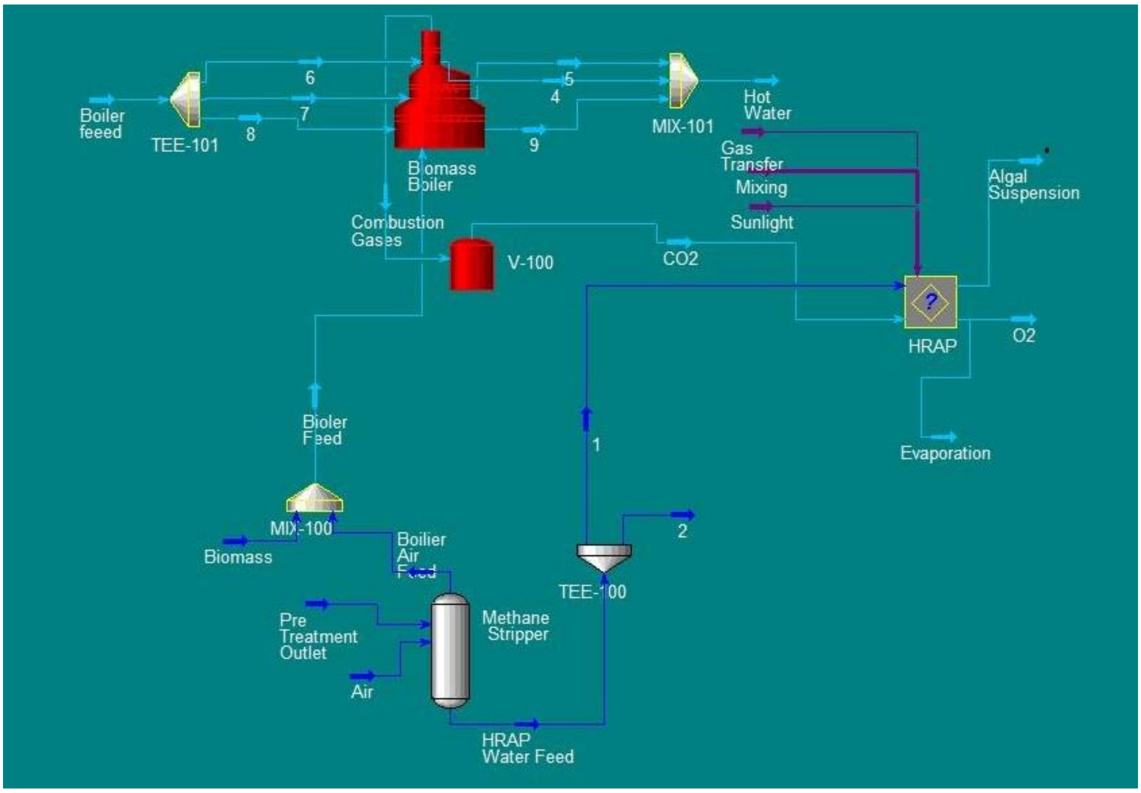


Figure 18 UNISIM flow-sheet of part of the proposed FP7 All-Gas Micro-algal biogas production plant

After examination of the initial UNISIM flow-sheet, Honeywell commented that; "it is unusual kind of flow-sheet to be modelled within UNISIM Design and is innovative and challenging, involving unusual unit operations and components that are not among the default unit operations available" (Niemirowski, Honeywell private communication, 2011). UNISIM Design does not have all the unit operations required for FP7 All-Gas flow-sheet, but the user may create custom unit operations, property packages and kinetic reactions.

UNISIM, unlike SuperPro, does not have a "cell function", but the components of a fluid within a stream can be distributed among 3 phases; gas, liquid and solid. The modelling of an algal cell as complex of water, carbohydrate, proteins lipid and other organic and non-organic compounds would appear impractical. Algae could be modelled as a solid using an average chemical composition for algae, and this approach was applied to the woody biomass feed for the biomass generators using the chemical composition of cellulose ($(C_6H_{10}O_5)_n$). Unfortunately, a problem was encountered as no combustion of the woody biomass occurred in the biomass boiler using the standard UNISIM fired heater unit operation model. The UNISIM operation manual supplied with software states, "the combustion reaction in the burner model of the fired heater performs pure hydrocarbon combustion calculations only". A simulated fuel containing elements other than C and H does not appear to run in standard fired boiler model and an alternative unit operation must be modelled.

UNISIM appears to be capable of producing dynamic flow-sheet mass and energy balance models for the oil, gas and related industries using a standard suite of component materials and unit operations. It also is capable of customisation with the ability to produce additional customised unit operations. It was always envisioned that a customised model of the HRAP based on algal growth unit operation would need to be produced for UNISIM, but it was hoped that many of the unit operations surrounding the HRAP could be simulated using the standard options available. Unfortunately, from initial experience it would appear that a considerable number of unit operations will need to be developed in the customisation of UNISIM for an algal biofuel flow-sheet and this view is supported by Honeywell (Niemirowski, Honeywell private communication, 2011). Although UNISIM could be capable of producing dynamic mass and energy balances for an algal biofuel production process a very considerable amount of effort and time, greater than 3 years, could be needed to produce them and it was decided that Aspen should be evaluated before further time was spent on UNISIM.

3.1.5 **Aspen Plus software**

Aspen Plus is the most widely used commercial process simulation software (Jana, 2009). It appears capable of producing batch, continuous and dynamic flow-sheet mass balances, but requires customisation for bioprocesses (Gosling, 2005). Custom unit operations can be generated using Excel workbooks or Fortran subroutines (Jana, 2009, Schefflan, 2011). A considerable database of potential process stream materials is available within Aspen. In a project to develop a physical property database for biofuel components for Aspen Plus, focusing on production of ethanol from lignocellulose, it was found, however, that many key components were not available in the standard Aspen database and many properties of biological materials needed for successful process simulation were not available anywhere (Wooley and Putsche, 1996).

Aspen has been used for a variety of analyses of parts of processes for the production of bioenergy from micro-algae and other organic materials, but does not, as yet, appear to have been used for a dynamic model of an entire micro-algal biofuel flow-sheet. Aspen has been used for a techno-economic assessment for the production of hydrogen, methane and ethanol from potato processing waste and barley straw (Ljunggren, 2011) and for four studies on 'biodiesel' production from micro-algae (Carlson et al., 2010, Davis et al., 2011, Peralta et al., 2010, Pokoo-Aikins et al., 2010).

A senior design report has been carried out on Algae to Alkanes, at the University of Pennsylvania. The study looked at growth, harvesting and oil extraction; however Aspen was only used for the 'catalytic hydro-treatment' of triglycerides to produce n-alkanes for blending in mineral diesel (Carlson et al., 2010). Another study used Aspen Plus to assess the economic impact of oil content, oil conversion and selling prices of glycerol and biodiesel from the alkali trans-esterification of algal oil from heterotrophic growth of *Chlorella,* but focused on the downstream processing following micro-algal growth, harvesting and oil extraction (Pokoo-Aikins et al., 2010). Some thermodynamic data was available in Aspen for fatty acids, but some crucial properties were not present for fatty acids and data for most components was not present. Data and properties had to be entered using the user definitions or estimated using Aspen Plus from the molecular structure, although the program does not automatically distinguish between cis and trans compounds. Little information is given on the production of mass and energy balances or the ability of Aspen Plus to produce dynamic models. Aspen Plus has also been used for an exergy analysis for the trans-esterification, methanol recovery, washing and FAME production in the conversion of *Chlorella* algal oil to biodiesel with the physical and chemical exergy of each stream being "calculated with the help of the thermodynamic properties calculated by Aspen Plus" (Peralta et al., 2010)

A detailed techno-economic analysis of the growth of autotrophic algae, in both open ponds and photo-bioreactors, for biofuel production has been carried out using Aspen Plus (Davis et al., 2011). Aspen software was used; "in order to obtain more accurate mass balance information than is typically assumed", to estimate key requirements (such as hydrogen demand in hydro-treating the algal oil to produce biodiesel) and to estimate the power requirements of the process. Although this study indicates that accurate mass balances can be produced from Aspen for algal fuel process flow-sheets, it does not comment on the ability of the software to produce dynamic models, but does add a caveat concerning process power calculations. Additional assumptions had to be made for estimation of the power required by the process which did not affect the mass balance, but "the data produced should be viewed as a qualitative estimate rather than an absolute quantitative basis" (Davis et al., 2011).

There have recent reports that a complete non-dynamic techno-economic model has been developed for production of liquid biofuel from micro-algae, but no details of the model have been published and it is not freely available for evaluation and adaption (Aspen Technology Inc., 2012, Dunlop, 2012). The model took over three years to develop and a dynamic energy balance model, although considered possible, would take considerably longer (Dunlop private communication, 2012).

Evaluation of Aspen Plus to model micro-algal biofuel flow-sheets was carried out based on the section of the proposed FP7 All-Gas flow-sheet consisting of the methane stripper, biomass boiler and algal high rate growth pond also used to evaluate UNISIM. The evaluation indicated that, as with UNISIM, Aspen is capable of producing dynamic mass and energy balances, but much of the data and many of the unit operations required for micro-algal biofuel production are not available within the program. Some of the unit operations would need modification and adaption to be used: for example, the biomass burner model produced heat output, but no carbon dioxide and an additional model would need to be added to produce the combustion products.

Aspen Plus may be capable of producing dynamic mass and energy balances for an entire algal biofuel production process. However, the production of a dynamic model for a complete flow-sheet is probably currently impractical. Work as part of another project at the University of Southampton using Aspen Plus to model part of a micro-algal biofuel process, anaerobic digestion, rather than a full dynamic model of an entire micro-algal biofuel process has confirmed the difficulties of producing a functioning steady state model for even one part of a micro-algal biofuel process and the need for more data.

3.1.6 **Pinch analysis**

Pinch analysis has been used successfully in process optimisation for over 30 years, initially for the efficient use of energy for heating and cooling and subsequently for reducing water and hydrogen usage (Klemes et al., 2010, Knopf, 2011, Miller et al., 2010). When heating and cooling curves of a process are plotted the point where the two curves are at their closest is known as the 'pinch' and defines the minimum rate of heat exchange. There are two process design problems in bio-chemical processes; unit operation design and the design of the whole system. Pinch analysis addresses the whole system with the application of targets and begins with the most constrained point in the network or pinch (Kemp, 2007).

Pinch analysis has been used to analyse the process streams that are available for thermal integration and energy consumption optimisation of bioethanol production process from sugarcane juice (Dias et al., 2009). A pinch analysis has also been carried out to optimise energy and water demands for ethanol from corn (Franceschin et al., 2008). There currently appears to be very little reported use of pinch analysis for parts of a micro-algal biofuel process and none for a complete process from growth of micro-algae to biofuel production. Pinch analysis has been used to optimise heat transfer integration in the trans-esterification of micro-algal oil in a model using Aspen software and simulated potential micro-algal oil based on nine triglycerides and fatty acids (Sanchez et al., 2011).

Carbon Emissions Pinch Analysis has been developed from ‘traditional’ pinch analysis for optimisation of the power generation mix based on demand/emissions targeting and has been further extended to include carbon capture and storage (Atkins et al., 2010, Shenoy and Shenoy, 2012). The use of a novel carbon pinch analysis may be useful in examining and optimising use of carbon dioxide and the conversion of inorganic to organic carbon and organic carbon to carbon dioxide in a micro-algal biofuel production process (Milledge, 2012a).

Pinch analysis cannot produce energy balances, but could be useful in optimising heat transfer and carbon and water usage; however it is probably not appropriate at this early stage of micro-algal biofuel process development.

3.1.7 **Conclusions**

UNISIM and Aspen Plus may be capable of producing dynamic mass and energy balances for an entire algal biofuel production process. However, the production of a dynamic model for a complete flow-sheet, from either software, for micro-algal biofuel, although innovative and challenging, is probably currently impractical. A very considerable amount of further effort and time (greater than 3 years) will be needed to produce algal biofuel dynamic models as many unit operations and components are not among the defaults available in either UNISIM or Aspen Plus. The lack of long-term full-scale operational data for micro-algal biofuel production may also require a large number of modelling assumptions for each unit operation within the processes, leading to a model that is at best easily criticised and probably inaccurate.

Pinch analysis, although useful for the optimisation of heat and water usage, cannot produce energy balances. There have been a limited number of energy balance assessments of micro-algal biofuel production and a greater number of LCAs that have some element of energy balance assessment, but these generally lack the rigor of an engineering mass and energy balance and none have assessed the interaction of growth system, harvesting and biomass exploitation on operational energies and net recovered energy.

There is currently no ready-made platform that can be used to produce an energy balance model for micro-algal biofuel, and there thus is an overriding need to develop a simple fundamental energy balance model of the micro-algal biofuel process. As noted at the end of Chapter 2, the proposed model should focus on open raceway ponds and anaerobic digestion; the following chapters described the process of development of the model and the results of scenario analysis carried out with it.

4. Initial investigations

As a preparatory step in the development of an algal biofuel energy balance model, initial investigations were carried out to establish the algal biomass and oil growth yields as a basis for the estimation of maximum theoretical and 'achievable' yields; and to define the operational energy returns on paddlewheel mixing in a raceway.

4.1 Micro-algal yield

Many authors believe that the productivity of micro-algal systems could potentially exceed that of land-based agriculture (Chisti, 2007, Rawat et al., 2013, Sheehan et al., 1998), but as early as the 1950s there were complaints of 'far-fetched estimates' (Tamiya, 1957) and very optimistic assessments of potential algal production have continued to appear. It has been suggested that "some proponents of algae mass culture would appear to go far beyond exaggeration into the realms of science fantasy" (Walker, 2009). A recent report on algal research in the UK has suggested unrealistic claims threaten the credibility of algal biofuels (Schlarb-Ridley, 2011). It thus appears vital for any energy assessment of micro-algal biofuel that the maximum and potential operational micro-algal yields are established.

4.1.1 Photosynthesis and photosynthetic efficiency

In autotrophic micro-algae, energy from solar radiation is converted into stored biomass by photosynthesis. Not all of the solar energy arriving at the cell can be used in photosynthesis. The photosynthetic efficiency (PE) is the percentage fraction of total light energy (solar insolation) converted into chemical energy (higher heating value (HHV) of biomass) during photosynthesis by algae or plants. Only light within the wavelength range of 400 to 700 nm (photosynthetically active radiation, PAR) corresponding to ~45 % of total solar energy can be utilised by plants, (FAO, 1997, Goldman, 1979b, Weyer et al., 2010).

Fixation, the process by which photosynthetic organisms convert carbon dioxide into organic compounds, has a minimum quantum requirement of between eight and ten photons of PAR to fix a CO_2 molecule (Brennan and

Owende, 2010, Kruse et al., 2005, Williams and Laurens, 2010), although over twelve photons are required to produce complex organic molecules (Wilhelm and Jakob, 2011). 1W m^{-2} of PAR is equivalent to 4.56 μmol m^{-2} of PAR photons (Thimijan and Heins, 1983) and therefore 1 mol m^{-2} is equivalent to 220 kJ m^{-2}. If eight photons are required to fix one atom of carbon then eight moles of PAR photons are required to fix one mole of carbon, equivalent to 1760 kJ of energy (8 * 220 kJ).

Photosynthesis can be simplified by regarding it as two reactants (carbon dioxide and water) and two products (glucose and oxygen) (Section 2.2.3). Six moles of carbon are required to produce one mole of glucose which has an enthalpy of combustion, higher heating value (HHV) or calorific value, of 2805 kJ (Wooley and Putsche, 1996). It requires 10560 kJ (6*1760 kJ) of energy from PAR to carry out the conversion via the photosynthesis. The maximum utilisation of PAR is therefore only 26.5 % (2805 kJ/10560 kJ). The theoretical maximum efficiency of total solar energy conversion by photosynthesis is therefore 11.9 % (45 % PAR in Solar radiation * 26.5 %). Other authors have suggested values of 12.4 % (Tredici, 2010), 11.7 % (Williams and Laurens, 2010) and 11 % (FAO, 1997) for the maximum theoretical photosynthetic efficiency (PE), based on similar calculations.

In practice, the photosynthetic efficiency (PE) observed in the field may be further decreased by factors such as: poor absorption of sunlight due to its reflection; respiration; and the need for optimal solar radiation levels (Tredici, 2010, Weyer et al., 2010, Williams and Laurens, 2010). Although micro-algae are often said to be more photosynthetically efficient than land plants (Wilhelm and Jakob, 2011) it is generally agreed there is little or no difference in the maximum efficiency (Tredici, 2010). If algal biomass yields are higher this it is a result of: a) every micro-algal cell being capable of photosynthetic activity, unlike terrestrial plants (Chisti, 2010); b) the lack of "waste" (no roots, stem or leaves that are not the target material); c) the more intimate contact between the algal cells and their nutrient medium (Tredici, 2010, Walker, 2009, Williams and Laurens, 2010). Sugarcane has been reported to achieve up to 8 % photosynthetic efficiency, but generally most land plants achieve 1 % or less (Kruse and Hankamer, 2010, Packer, 2009).

The conversion efficiency of photosynthesis can be compared with that of other light-based energy systems. Photo-voltaic cells can have conversion efficiencies of over 30 % (Green et al., 2011, NREL, 2011, Stephenson et al., 2011), with commercial systems having efficiencies of 10-22 % (Twidell and Weir, 2006) and experimental organic systems having an efficiency over 8 % (Green et al., 2011, NREL, 2011). Photosynthesis has the advantage that it can synthesise high energy molecules for liquid fuels.

4.1.2 **Solar insolation**

The solar insolation or solar energy reaching the earth surface varies with latitude and local climate conditions, as illustrated in Figure 19.

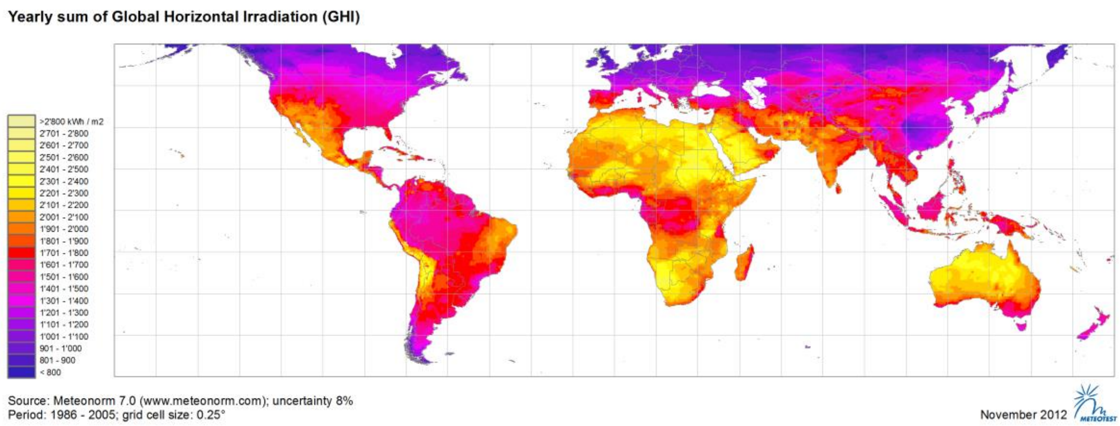


Figure 19 Total annual solar insolation. Courtesy METEOTEST www.meteonorm.com

In areas commonly considered suitable for commercial growth of micro-algae, such as the south-western USA (Sheehan et al., 1998), southern Spain (Banks et al., 2011a), Israel (Ben-Amotz, 2008a) or costal Namibia (Harvey et al., 2012, Milledge, 2013), solar energy reaching the ground is approximately 2000 kWh m^{-2} $year^{-1}$ (Maryland, 2007, Meteotest, 2009, Ministry of Environment and Tourism Namibia, 2002, NASA, 2009, NREL, 2009). Levels in Southern England are about 50 % of this, and the maximum in places such as north-west Australia and central Sahara, is of the order of 25 % higher.

Using an annual total solar insolation of 2000 kWh m^{-2} $year^{-1}$ typical of areas suggested for micro-algal biofuel production, and a maximum 11 % efficiency of solar energy conversion by photosynthesis, a potential maximum production biomass energy can be calculated as 220 kWh year^{-1}.

4.1.3 Micro-algal calorific value and calculated biomass yield

To estimate the maximum micro-algal biomass yield the calorific value of the biomass must be established. The potential lipid yield is also of interest for production of biodiesel, the focus of most the research on micro-algae (Chisti, 2007, Sheehan et al., 1998, Sills et al., 2012) (Section 2.4.8). The calorific value of food products can be estimated from the typical calorific values of carbohydrate, protein and lipid (Merrill and Watts, 1955). A similar simplified approach could thus be used to estimate the calorific value of micro-algae for a range of lipid contents as shown in the following paragraphs.

A typical calorific value for carbohydrate is 4.9 kWh kg^{1} and for lipid is 10.9 kWh kg^{1} with value for protein approximately similar to that of carbohydrate (Merrill and Watts, 1955, Platt and Irwin, 1973). Based on the above values the calorific value of algae for a range of oil contents (10 to 90 %) was calculated. Using the calculated calorific values of the micro-algae, the potential caloric value of biomass derived from the photosynthetic efficiency (11 %) and solar insolation it is possible to estimate the maximum yields of micro-algal biomass and oil. The same approach could be used with alternative calorific value data such as that generated from typical empirical formulae using the Du Long equation (IFRF. International Flame Research Foundation, 2004) (Equation 13).

4.1.4 Results and discussion

Table 7 shows calculated micro-algal biomass and oil yields for a range of oil contents (10-90 %) at a solar insulation of 2000 kWh m^{-2} year^{-1} the level typical of the south-west USA, Spain, south-west Africa, western Australia and many other suggested micro-algal growth locations.

4.1.4.1 Calorific value of micro-algae

An estimated calorific value for algae with 20 % oil content is 6.0 kWh kg^{-1}. This is in agreement with:

a. the calorific values of marine algae of 4.16 - 6.36 kWh kg^{-1} (Paine and Vadas, 1969)
b. a range of reported literature values of 20 - 23.75 kJ g^{-1} (5.56 - 6.6 kWh kg^{-1}) for algae with an average lipid content of up to 20 % (Tredici, 2010, Weyer et al., 2010)
c. a reported calorific value of *Chlorella* at 6.3 kWh kg^{-1} (Goldman, 1979b).

The value from Table 7 for 50 % oil content algae of 7.9 kWh kg^{-1} is in good agreement with the 26.9 kJ g^{-1} (7.5 kWh kg^{-1}) calculated for 50 % oil content algae *Nannochloropsis* (Weyer et al., 2010).

Table 7 Calculated maximum theoretical algal biomass and oil yields

Algae oil Content	Calorific value	Yield Algae	Yield Algae	Yield Algal Oil
	kWh kg^{-1}	Tonnes Ha^{-1} $year^{-1}$	g m^{-2} day^{-1}	Tonnes Ha^{-1} $year^{-1}$
10%	5.5	401	110	40
20%	6.0	361	99	72
30%	6.7	328	90	99
40%	7.3	301	83	120
50%	7.9	278	76	139
60%	8.5	258	71	155
70%	9.1	241	66	169
80%	9.8	226	62	181
90%	10.4	213	58	192

4.1.4.2 **Maximum theoretical micro-algal biomass yield**

Maximum theoretical algal dry biomass yield was estimated at 401 tonnes ha^{-1} $year^{-1}$ or a daily average of 110 g m^{-2} day^{-1} (Table 7) for10 % oil content, but algae with negligible oil content could have an estimated maximum yield of 123 g m^{-2} day^{-1}.

Grobbelaar (2009b) (2010) calculated a maximum algal biomass dry weight yield of 179 g m^{-2} day^{-1} assuming 40 % carbon content of the VS and 1104 µmol quanta m^{-2} s^{-1} of solar radiation reaching the earth's surface over a 12 hour diurnal cycle (equivalent to 2482 kWh m^{-2} $year^{-1}$). The carbon content is low. The carbon content of glucose is 40 %. and its calorific value is 4.3 kWh kg^{-1}, (Merrill and Watts, 1955, Wooley and Putsche, 1996). If the calorific value of glucose is assumed for 40 % carbon content algae a yield of 140 g m^{-2} day^{-1} can be calculated (in the same way as the data in Table 7 was calculated), but if adjusted for the higher solar insolation (equivalent to 2482 kWh m^{-2} $year^{-1}$) (Grobbelaar, 2009b) the result is 174 g m^{-2} day^{-1}. Although reducing the calorific value of algae to that of glucose increases the maximum calculated yield, and into agreement with that calculated by Grobbelaar (2009), the low carbon content of 40 % is not realistic and leads to an overestimation of maximum yield.

A typical carbon content of 50 % has been suggested for micro-algae (Aquafuels, 2011, Chisti, 2007) and a carbon content of 59 % has been calculated from a suggested stoichiochemistry of algal growth (Buhr and Miller, 1983). The CE-QUAL-W2 water quality model gives a range of reported literature values for micro-algae carbon content between 46 and 56 % (Cole and Wells, 1995). The carbon content as a percentage of algal VS has been calculated as 53 % for low lipid content algae and 60 % for high lipid content algae (Williams and Laurens, 2010) and measured at 51 and 56 % for *Chlorophyceae* (green micro-algae) (Ketchum and Redfield, 1949). The typical carbon contents of algal carbohydrate, protein and lipid are: 44 %, 46 % and 77 % based on a review of reported results (Heaven et al., 2011). Carbon content and calorific value increase with increasing lipid content and it can be estimated using stoichiometry that algae with oil content of 20 % would have a carbon content of ~50 %.

Williams and Laurens (2010) estimated the dry biomass yield of algae at a range of latitudes, based on an overall photosynthetic efficiency (PE) of 10 %, reduced from 12 % to allow for respiration. For algae with an oil content of 25 % and calorific value of 24.7 kJ g^{-1} (6.9 kWh kg^{-1}), estimated annual yields were approximately 300 tonne ha^{-1} $year^{-1}$ at latitude 30° N (corresponding approximately to the south-west USA). Daily rates varied from 50 g m^{-2} day^{-1} in winter to 117 g m^{-2} day^{-1} in mid-summer. When corrected for the lower photosynthetic efficiency and higher estimated calorific value this result is similar to those in Table 7.

4.1.4.3 Maximum theoretical micro-algal oil yield

A figure for algal oil production of 324 tonne ha^{-1} $year^{-1}$ (196 g m^{-2} day^{-1}) was calculated for the theoretical best case growth of algae with a 50 % oil content (Weyer et al., 2010), but a median literature calorific value of 21.9 kJ g^{-1} (6.1 kWh kg^{-1}) was used that significantly underestimate the calorific value of 50 % oil content algae. This together with very high solar insolation value of 11616 MJ m^{-2} $year^{-1}$(3227 kWh m^{-2} $year^{-1}$) result in a significant overestimate of potential oil yield. Zemke et al. (2010) calculated a maximum yield based on the assumption that the algal cell consisted entirely of triglyceride and obtained a value of 58 g m^{-2} day^{-1}, in close agreement with the 90 % oil content algae calculated in Table 7 .

4.1.4.4 Potential achievable micro-algal biomass and oil yield

Realistic achievable annual average daily yields are substantially lower than the calculated theoretical maximum (Milledge, 2010b). An early review of the growth of algae found yields of 5 - 21 g m^{-2} day^{-1} with photosynthetic efficiencies of between 1.2 - 3.2 % and suggested that 20 - 28 g m^{-2} day^{-1} is achievable in open algal growth systems (Tamiya, 1957). Other reviews, summarised in Table 8, have found algal yields ranging from 3 - 30 g m^{-2} day^{-1} and suggested achievable yields of 20 - 30 g m^{-2} day^{-1}. Yields of *Spirulina* from commercial open ponds for nutritional supplements are reported between 3 – 8 g m^{-2} day^{-1} equivalent to a photosynthetic efficiency of 0.25 % - 0.75 % (Reijnders, 2009).

The NREL in its extensive study reported single day productivities as high as 50 g m^{-2} day^{-1}, (equivalent to ~5% of overall solar radiation energy), but continuous production levels were substantially below this level with biomass productivities equivalent to a total solar energy conversion efficiency of about 2 % (Sheehan et al., 1998). Seambiotic in Israel have reported yields of 20 g m^{-2} day^{-1} equivalent to 73 tonne ha^{1} $year^{-1}$ (Ben-Amotz, 2008a) and Auburn University also suggested economically practical rates of 20 g m^{-2} day^{-1} for the south-eastern region of the USA (Putt, 2007),in both cases probably of the order of 2 - 2.5 % total solar energy conversion efficiency. Other published experimental data (Table 8) has shown growth rates between 2 - 35 g m^{-2} day^{-1} and an overall photosynthetic efficiency (PE) of 2.8 % for the growth of *Chlorella* in an open air lift photo-bioreactor (Strik et al., 2008).

Table 8 Algal dry weight yields and photosynthetic efficiencies from published sources

Reviews			
Yield g m^{-2} day^{-1}	Photosynthetic Efficiency %	Suggested Achievable Yield g m^{-2} day^{-1}	Reference
5-21	1.2 -3	20-28	(Tamiya, 1957)
15-25	0.25	30	(Goldman, 1979a)
3-8			(Reijnders, 2009)
		20	(Brune et al., 2009)
10-40			(Singh and Olsen, 2011)
Published Experimental Data			
Yield g m^{-2} day^{-1}	Photosynthetic Efficiency %	Suggested Achievable Yield g m^{-2} day^{-1}	Reference
25 -29			(Johnson et al., 1988)
16	1.1 – 3.15	20	(Weissman et al., 1989)
15			(Laws and Berning, 1991)
16-35			(Moheimani and Borowitzka, 2006)
	2.3		(Bosma et al., 2007)
	2.8		(Strik et al., 2008)

As noted above the maximum theoretical photosynthetic efficiency is around 11 %, but losses from reflection, photo-respiration, respiration and photo-saturation reduce the overall value to 5 - 6 % (Tredici, 2010, Walker, 2009, Williams and Laurens, 2010). Photons of different wavelengths have different energy contents and photosynthesis is unable to use the additional energy in blue light relative to red light, resulting in a loss of 6.6 % of the available energy in PAR (Martinez, 1996, Stephenson et al., 2011). Although yields based on a photosynthetic efficiency of 5 % are possible, the published data suggest that current practical photosynthetic efficiencies for the growth of micro- algae are 2 to 3 %. In land plants grown for biofuels a photosynthetic efficiency of 1 % is considered to be the best currently achieved in commercial production, but a doubling to 2 % on a large scale could reasonably be aspired to in the future (Walker, 2009). It would appear to be unjustified to use algal yield data based on photosynthetic efficiencies above 5 % for energy balance, techno-economic or life cycle assessment models, and a photosynthetic efficiency of 2 - 3 % appears more appropriate. Other sources indicate that it is now becoming generally accepted that the upper practical limit for micro-algal overall photosynthetic efficiency (PE) is 3% (Ben-Amotz, 2013). Recalculation of the data in Table 7 for a photosynthetic efficiency of 2.5 % and a solar insolation of 2000 kWh m^{-2} $year^{-1}$ at micro-algae oil contents of 10- 30 % gives average daily micro-algal dry biomass yields of 20 - 25 g m^{-2} day^{-1} and this would appear to be a realistic yield for assessment of output in algal growth system models. Micro-algal yields in temperate climates, such as SE UK, for the same photosynthetic efficiency will be half of that of areas with solar insolation 2000 kWh m^{-2} $year^{-1}$.

4.1.5 **Conclusions**

The simple calculation used in Table 7 allows calculation of maximum theoretical dry algal biomass and oil yields that can be used to counter some of the extreme yield values suggested in the ‘grey literature’. It takes into account the increase in calorific value with increasing oil content, unlike other studies such as Weyer et al. (2010). By reducing the maximum photosynthetic efficiency to take account of losses, such as photo-respiration, respiration and photo-saturation, it can be used to produce a realistic, practical or pragmatic yield, and by varying the value for solar insolation it can be adapted for a wide range of locations.

The literature appears to indicate that the maximum realistic or achievable overall photosynthetic efficiency is 3 %. The method of calculation used, however, does not make allowances for the additional energy losses in the light independent reactions in the algal cell to produce complex carbohydrates, proteins and lipids, or the reduced growth rates that are commonly found under nutrient stress. Further reductions below the pragmatic overall photosynthetic efficiency of 3% should be made in setting micro-algal yield targets for oil contents above ~30 % lipid to adjust for the reduced growth rate that occurs when nutrient stress is used to promotes lipid accumulation.

The estimation of yields could be further improved by the use of separate calorific values for protein and carbohydrate, and of calorific values based on the chemical composition of micro-algal components rather than typical food component values. These improvements will be integrated into the final energy model.

The establishment of maximum and realistic algal yields allows an energy balance and an energy return over operational energy invested (EROOI) to be postulated for individual growth systems, algal biofuel process operations and proposed complete algal biofuel production systems. Algal yield and energy contents figures will also permit the setting of appropriate input energy targets for processes operations.

4.2 Operational energy returns on paddlewheel mixing

Mixing in algal raceways is normally achieved by paddlewheels (Section 2.2.3), but how much energy of the potential calorific yield of micro-algae is used to maintain fluid flow?

4.2.1 Energy return on operational energy investment EROOI

Energy return on energy investment (EROEI or EROI) is the ratio of the energy produced compared to the amount of energy invested in its production. However as previously discussed (Section 1.2) problems arise in deciding which inputs and outputs count.

It is often unclear in the literature what has been included in a particular calculation. Reported EROIs often exclude the embodied energy in process equipment, but the embodied energy in the equipment does not appear to have been reported as major factor in the production of micro-algal biofuels. The embodied energy within process equipment is not considered in the energy balances of this work. The major energy inputs in the production of micro-algal biofuel are operational energy and the embodied energy of nutrients (Aquafuels, 2011). The use of low embedded energy sources of water, nutrients and CO_2, such as wastewater and flue gas have been assumed through-out. The embodied energy of materials has been excluded from the energy balances calculated subsequently unless specifically mentioned in any results such as that for flocculation harvesting (Section 6.9).

The term energy return on operational energy invested (EROOI) is ratio of the energy output to the operational energy input. The output will be the HHV of the biomass or where biogas is the end product the HHV of the methane in the estimated biogas production. The input will be the operational energy requirement, heat and electricity, of the process equipment. EROOI is used throughout this section and subsequent sections to measure energetic viability and net energy return.

4.2.2 **Basis of calculation**

Section 4.1 indicated that a pragmatic yield is 25 g m^{-2} day^{-1} for 20 % lipid micro-algal biomass having a calorific value of 6.0 kWh kg^{-1}. The total potential energy produced per day in micro-algal biomass is therefore 0.15 kWh m^{-2} day^{-1}.

The total hydraulic power required to move a micro-algal suspension around a raceway can be calculated from the head losses as previously discussed in Section 2.2.2.3 together with Sections 2.2.2.6 & 2.2.4

4.2.3 **Results and discussion**

Table 9 shows the estimated head losses in lined and unlined raceways of different dimensions based on Equation 3, 7 & 10 and assuming a friction factor of 0.01 for lined and 0.02 for unlined.

Table 9 Head losses in lined and unlined raceways [a].

		Head loss m			
	Velocity m s^{-1}	0.15		0.3	
	Friction Factor	0.01	0.02	0.01	0.02
Raceway Dimension	Area m^2				
Width / Straight Length	$w*2L + \pi w^2$				
1m / 50 m	103	0.010	0.016	0.040	0.065
10 / 100 m	2314	0.010	0.018	0.041	0.070
20 m / 200 m	9267	0.013	0.026	0.050	0.106
[a] Calculated from Equations 3, 7 & 10					

The head loss in lined raceways is approximately half that of unlined, halving the power and energy required for maintaining fluid flow within a raceway. Liners, therefore, are valuable in reducing raceway operational energy in addition to reducing seepage, improving cleanability and reduced water clouding problems due to the suspension of soil particles (Section 2.2.1).

For a pilot-scale raceway with a smooth liner surface, of the type used in Spain by the University of Southampton (Manning friction factor 0.01, 1 m wide, 0.3 m deep, with straights 50 m long and a mean fluid velocity of 0.3 ms^{-1}) a hydraulic power requirement of 35 W can be calculated (Milledge, 2011b). If a raceway is mixed for 24 hours the hydraulic energy required is 0.84 kWh day^{-1}. The hydraulic power required to mix the raceway is thus over 5 % of the total energy available in the biomass production of 25 g m^{-2} day^{-1}. Typically, quoted paddlewheel efficiencies are 10 to 20 %, and therefore, from one quarter to over half of the total potential higher heating value of the algal biomass could be used in mixing such a raceway. Optimised paddlewheels can have higher efficiencies, but a 40 % efficient paddlewheel would still use the equivalent 13 % of the energy in algal biomass to mix a small raceway.

Table 10 shows the calculated paddlewheel energy (assuming 40% overall efficiency), expressed as a percentage of micro-algal biomass energy content (based on a yield of 25 g m^{-2} day^{-1} and a calorific value of 6.0 kWh kg^{-1} for various size lined raceways at fluid velocities of 0.1 m s^{-1} and 0.3 m s^{-1}.

Table 10 Paddlewheel energy consumption as percentage of micro-algal biomass calorific value

		Paddlewheel Energy as % of Algal Calorific Value	
	Velocity $m\ s^{-1}$	0.15	0.3
Raceway Dimensions m	Area m^2		
Width / Straight Length			
1 m / 50 m	103	1.70%	13.50%
10 m / 100 m	2314	0.70%	6.20%
20 m / 200 m	9267	0.50%	3.80%

Larger raceways give a better energy return on mixing investment and reducing the fluid velocity also significantly improves energy return. The lower mixing energy requirement as a percentage of the potential micro-algal biomass energy yield in larger ponds compared to smaller ones is a result of the higher proportion of straight sections with a relatively low head loss. The eight-fold reduction in power requirement from halving of fluid velocity is due to hydraulic power being a function of the cube of fluid velocity.

The results in Table 10 are in agreement with the recalculated experimental data from Green et al. (1995), for an 0.1 hectare raceway in California, with a mixing energy requirement of 0.12 to 0.25 kWh to produce 1 kg of algae in an open pond operating at fluid velocity of 0.15 $m\ s^{-1}$, equivalent to an energy requirement for mixing of between 2 - 4 % of the calorific value of the micro-algae produced. LCA s have used figures between 0.05 and 3.3 % (Collet et al., 2011).

The power required to give a flow velocity of 0.15 $m\ s^{-1}$ in a 0.3 m deep unlined raceway of one hectare using a paddlewheel with 40 % efficiency has been estimated at 18 kWh $day^{-1}\ ha^{-1}$ (1 horsepower) (Benemann and Oswald, 1996).This is equivalent to an energy requirement for mixing of 1.2 % of the calorific value of micro-algae and is again in good agreement with the results in Table 10.

4.2.4 **Conclusions**

The spreadsheet developed to calculate the mixing energy requirement for raceways produced results that are in agreement with other published data. A simple EROOI has been produced, using this calculation and a pragmatic yield. This will be further developed and used in calculations of operation energy returns for entire micro-algal bioenergy production systems.

The required energy inputs relative to the biomass calorific value decreases with the addition of a liner, reduction in fluid velocity and increase in raceway size. For maximise EROOI ponds should, therefore, be lined and as large as practicable. The fluid velocity should be the minimum to provide mixing and regular exposure of the cells to light. It is possible that small gains in micro-algal yield due to improved mixing and exposure to light through increased fluid velocity may not be energetically or economically efficient, and sub-optimal micro-algal yields at lower flow-rates could offer a better energy return on energy investment. The effect of fluid velocity on EROOI is investigated further in Sections 5.1.2.1, 6.2.1 & 6.3.2.4, and the results of the work in this chapter were used in the development of a more extensive energy balance model, in the next chapter

5. Model construction

This chapter describes the construction and validation of the model, and some associated studies carried out to support the model development.

5.1 Model structure and assumptions

An operational energy and mass balance process integration model for micro-algal biogas production was developed, and implemented in a Microsoft Excel spreadsheet. The model was divided into three main operational areas based on those identified and explored in Section 2: growth, harvesting and energy extraction. The three areas were considered to be linked by a requirement for pumping power (Figure 20), which has not been fully accounted for in many studies (Dunlop, 2012). The model was comprised of nine worksheets, and was built up from fundamental equations such as those for fluid flow in pipes (Moody, 1944) and literature data such as that for fresh and salt water densities and viscosities (El-Dessouky and Ettouney, 2002, Kestin et al., 1978).

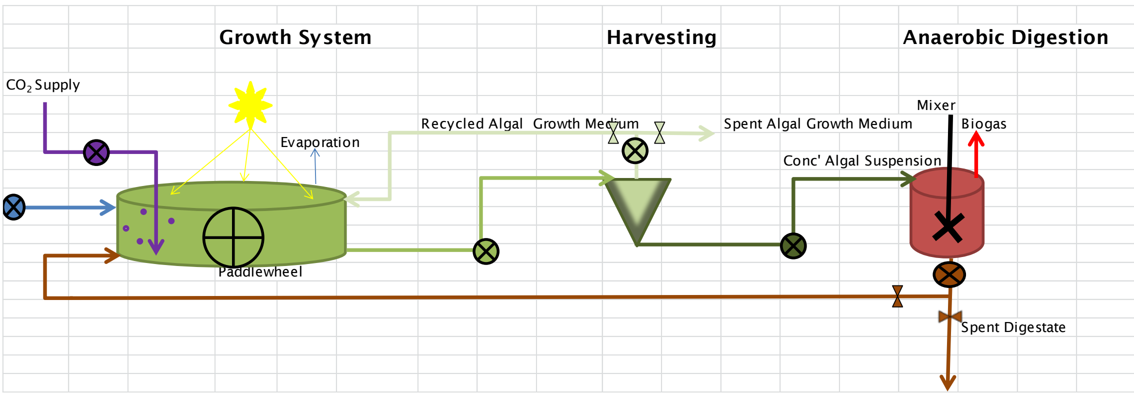


Figure 20 Excel model simplified process flow diagram

The construction of the model is described below with additional detail and specific assumptions discussed in the appropriate result and discussions sections.

5.1.1 Micro-algal biomass and biogas yields

The calorific yield of algal biomass was calculated using the annualised solar insolation and overall photosynthetic efficiency as described in Section 4.1. The algal biomass was assumed to consist of lipids, carbohydrate, proteins and inorganic material. The value of each could be set from 0 to 100 % with the total of all these components always being equal to 100 %. Default values were assumed of 20 % lipids, 30 % carbohydrate, 50 % proteins and 0 % inorganic material, equivalent to an overall empirical formula of $C_1H_{1.8}O_{0.5}N_{0.1}$ typical of micro-algae (Chisti, 2007, Grobbelaar, 2009a).

The higher heating value of the algal biomass was calculated from the typical empirical formulae for micro-algal lipid, carbohydrate and protein (Heaven et al., 2011) using a version of the Du Long equation (IFRF. International Flame Research Foundation, 2004)

Equation 13

$$\text{HHV(MJ kg}^{-1}\text{ dry fuel)} = \frac{34.1\text{C} + 102\text{H} + 6.3\text{N} + 19.1\text{S} - 9.85\text{O}}{100}$$

where C, H, N, S and O are the carbon, hydrogen, nitrogen, sulphur and oxygen in the biomass expressed as % VS; in the absence of values for sulphur this component is taken as zero. The biomass yield was calculated from the calorific yield and the estimated HHV of the micro-algae as in Section 4.1.4.1.

The potential methane and biogas yields were estimated from the typical empirical formulae for micro-algal lipid, carbohydrate and protein (Heaven et al., 2011) using the Buswell equation (Buswell and Mueller, 1952, Symons and Buswell, 1933). The HHV of methane was taken as 55.662 KJ g^{-1} at 0° C and 101.325 kPa (BSI, 2005).

5.1.2 Growth system

Open raceways were selected as the growth system because the overwhelming balance of the published information indicated that the energy inputs and costs of producing micro-algal biomass in open systems are significantly lower than PBRs (Section 2.1). The energy inputs in raceways are for ‘mixing’ and gas transfer.

5.1.2.1 Paddlewheel 'mixing' energy

Mixing in the raceway was assumed to be achieved by a paddlewheel, and the power and energy requirements were calculated using the fundamental equations for open channel flow previously used to estimate head losses and energy required to move a micro-algal suspension around a raceway (Sections 2.2.2.3 & 4.2).

5.1.3 Gaseous transfer energy

The carbon source for micro-algal growth was assumed to be gaseous CO_2 supplied from a waste source such as flue gas or biogas upgrading, and the embodied energy was assumed to be negligible. The concentration of CO_2 in the gas stream could be varied between 0 to 100 %. Gas was assumed to be bubbled into the cultivation medium via a sparger in a sump, with the total pressure drop being the sum of the hydrostatic head above the sparger and the pressure drop across the sparger from the friction losses in the supply pipework. It was assumed that CO_2 would only be supplied for an average of 12 hours per day during daylight hours (see Section 5.3.2.2).

The model allows both the sump depth and the sparger pressure drop to be set at any value. A typical pressure drop across a sparger of 6.89 kPa (1 psi) (Weissman and Goebel, 1987) was assumed. A sump depth of 2m was selected, as recent work has shown that a simple 1.8 m deep sump can transfer 90 % of the carbon dioxide from flue gas (University of Southampton, unpublished work).

The energy requirements for a blower to compress a gas can be calculated from the power for isothermal or adiabatic (where heat does not enter or leave the system concerned) compression. The model calculates both isothermal and adiabatic power requirements, but adiabatic power requirements are always greater and are typical of commercial blowers and compressors (Perry and Chilton, 1973, Rogers and Mayhew, 1992, Sinnott, 2005). The adiabatic compression power was therefore used to calculate the energy required for gaseous exchange in the raceway based on equations 14 and 15:

Equation 14

$$T_{out} = T_{in}\left(\frac{p_{out}}{p_{in}}\right)^{((z-1)/z)}$$

Where z is the ratio of specific heat at constant pressure to specific heat at constant volume and was assumed to be 1.4 as for air (Rogers and Mayhew, 1992). T_{in} and T_{out} are temperatures (degree K) in and out of the compressor and p_{in} and p_{out} are pressures (Pa) in out of the compressor

Equation 15

$$\text{Adiabatic Power (ideal gas)} = M^{*}C_p * [T_{out} - T_{in}]$$

Where M (kg s^{-1}) is mass flow and C_p is specific heat capacity at constant pressure taken as 1.005 kJ kg^{-1} K^{-1} for air (Rogers and Mayhew, 1992).

The mass flow requirement was determined from the CO_2 requirement for algal growth, calculated from the percentage of carbon in the micro-algal biomass based on the content and empirical formulae of lipid, carbohydrate and protein. An 80% transfer of carbon dioxide in the bubble to the growth media was assumed. Weissman et al. (1989) found 78 to 90% of CO_2 gas in bubbles was transferred in large scale open raceway sumps. Transfer declines exponentially with declining CO_2 concentration in the gas and higher transfer rates may not be economically attractive (Putt, 2007)

5.1.4 Outgassing of CO_2

In addition to that used for growth, CO_2 will be ‘lost’ to atmosphere as the growth medium is circulated around the raceway.

In raceways, gaseous exchange will not only take place at the site of the addition of CO_2 or flue gas (the sump), but also at the paddlewheel and around the entire raceway. Although high gas transfer coefficients occur at the paddlewheel, the low volume and the short residence time in the paddlewheel section result in lower gas transfer than the raceway and the sump (Mendoza et al., 2013b). The gas transfer at the paddlewheel was therefore excluded in order to simplify the model.

The rate of gas transfer to and from a liquid can be expressed as:

Equation 16

$$\frac{dC}{dt} = k_{gt} \left(C_s - C_o \right)$$

where $\frac{dC}{dt}$ is the rate of gas transfer (kg m^{-3} s^{-1}), k_{gt} is the gas transfer constant (s^{-1}), C_s is the saturation concentration of the gas and C_0 the concentration of the gas in the fluid (kg m^{-3}).

Initially gas transfer coefficients taken from results for ocean surfaces (Cole and Caraco, 1998, Wanninkhof, 1992) were considered for use in the model, but although this is a rich source of data it does not appear to be readily applicable to raceways with their shallow depth and fluid velocity. The re-aeration of polluted streams has produced a number of equations for the calculation of re-aeration coefficients or oxygen mass transfer coefficients (Chin, 2006, Cole and Wells, 1995, Jolhnkai, 1997, Melching and Flores, 1999), and are probably more applicable to the depths and velocities typically found in raceways. The equation originally developed by Owens (Chin, 2006, Cole and Wells, 1995) for depths of 0.1 to 3 m and fluid velocities of 0.03 to 1.5 m s^{-1} was chosen as being the most appropriate.

Equation 17

$$k_{gt} = 5.32 \, \frac{v^{0.67}}{d^{1.85}}$$

Where d is depth of the stream (m) and v is the fluid velocity (m s^{-1}).

The re-aeration coefficient for a 0.3 m deep raceway flowing at 0.3 m s^{-1} was calculated using Equation 17 as 22 day^{-1} (2.5×10^{-4} s^{-1}) which is in the range of values from 1 - 4×10^{-4} s^{-1} found for a large and small experimental raceway (University of Southampton, unpublished work).

The mechanism of gas transfer is similar for CO_2 and O_2 with the gas transfer coefficient of CO_2 being 0.923 times that of O_2 (Cole and Wells, 1995). The higher solubility of CO_2 relative to O_2 results in the mass transfer from a gas to a liquid being much greater for CO_2 than O_2 for the same gas partial pressure; there is much less CO_2 than O_2 in atmospheric air, however, so there is a bigger driving force for removal of CO_2 from the channels of a raceway. The mass transfer for outgassing of CO_2 from a raceway can therefore be expressed as:

Equation 18

$$k_{gt\,CO2} = 0.923 * 5.32 \frac{v^{0.67}}{d^{1.85}}$$

Raceways typically exhibit plug flow behaviour, with little or no longitudinal mixing (James and Boriah, 2010, Mendoza et al., 2013a, Molina Grima, 2011, Sompech et al., 2012). The Streeter Phelps equation is normally used to model how dissolved oxygen (DO) changes with distance in rivers or streams behaving as plug flow reactors (Chin, 2006). A modified Streeter Phelps equation was used to estimate the amount of CO_2 outgassed by using the mass transfer coefficient calculated from Equation 18 to replace the O_2 mass transfer coefficient.

5.1.5 Accumulation of O_2

O_2 produced during photosynthesis will accumulate in the fluid in the raceway until the concentration is such that the rate of outgassing is equal to that of net rate of production.

The equilibrium concentration reached in a raceway can then be determined using the equation below.

Equation 19

$$q_{O2} = k_{gt\,O2}\,(C_o - C_s)$$

where q_{O2} is the net rate of oxygen production from algal growth in the raceway (kg m^{-3} s^{-1}).

5.1.6 Pumping

The pumping energy required for each of the pumping stages was calculated for the frictional and static heads. Frictional head losses are due to shear stress on the pipe walls and are a complex function of the system geometry, the fluid properties and the flow rate in the system. The shear stress varies with velocity of flow and hence with Reynolds number.

The general Darcy-Weisbach equation for head loss due to friction was used to calculate head loss, h_p (m):

Equation 20

$$h_p = f_D \frac{L}{D}\frac{v^2}{2g}$$

Where D is the pipe diameter (m) and head loss is a function of the friction factor f_D which is not a constant, but depends on the parameters of the pipe and the velocity of the fluid flow.

For the laminar flow regime (Re < 2000) roughness has no discernible effect and friction factor was calculated using the following

Equation 21

$$f_D = \frac{64}{R_e}$$

For the turbulent flow regime the friction factor was calculated from the simplified equation developed by (Moody, 1944) based on the Colebrook-White equation:

Equation 22

$$f_D = 1.14 + 2\log_{10}\left(\frac{D}{e}\right)^{-2}$$

where D pipe diameter (m) and e is average roughness (m). Both values could be set in the model to any value. A default value of 0.046 mm was assumed for average roughness (e) (Coulson and Richardson, 1999). Typical commercial pipe diameters of 100 mm and 50 mm were also assumed. The diameter for the outflow pipe (50 or 100mm) for each pump being selected to give a flow velocity nearest to the suggested optimum velocity 3.0 m s^{-1} (Coulson and Richardson, 1999). Pipework length between units was initially assumed to be a nominal 10 m.

Static head losses could be set to any value, but were assumed to be minimal with default values of 0 m for suction heads and 0.3 – 3 m for outlet heads depending on the equipment type (raceway feed, 0.3m; centrifuge and lamellar harvester feed, 2m; conical settler feed, 3m; harvest return, 1m; and digestate return, 2m). The head for the supply pump to the digester was calculated from the required volume of the digester (see Section 5.1.8.1).

Fluid viscosity and density of fluids entering or exiting any one of the 3 areas of growth, harvesting or energy extraction via pumps could be set to any value. A data table was provided in the model giving the values for fresh (Kestin et al., 1978) or salt water (El-Dessouky and Ettouney, 2002) for a range of ambient temperatures between 10 and 70 °C (see data sheet within the spreadsheet titled "maxmin simple flow 1 fix vol ad", in the CD of accompanying material). Default values for all sections of the model were set at the values for fresh water as the viscosity and density of low concentration algal suspensions (<5%) are similar to water and the vast majority of the materials pumped and mixed were low concentration algal suspensions (Section 2.3.6).

5.1.7 **Harvesting**

The model was originally developed with one harvesting unit in which concentration factor, percentage recovery and energy input (kWh m^{-3}) could be varied. The flow volumes and concentration of micro-algae exiting the raceway and entering the harvesting system were calculated from the raceway volume and HRT and the biomass yields. The flow (m^3 hr^{-1}) and micro-algal concentration (% dry weight) leaving the harvesting unit were calculated based on the concentration factor and percentage recovery.

The model was later modified to include multiple harvesting units and to allow varying the flocculant dose and embodied and mixing energy (Section 6.9).

5.1.8 **Anaerobic digestion**

Completely Stirred Tank Reactor (CSTR) digesters are widely used to for treating liquid wastes with up to 10 % solids (Wilkie, 2005). They often operate mesophilically, with hydraulic retention times of about 20 days (Persson et al., 1979, Rittmann and McCarty, 2001). The CSTR digester design was therefore selected as suitable for handling micro-algal biomass suspensions.

5.1.8.1 **Digester volume and dimensions**

There are many shapes of digester, but a vertical cylindrical tank design is the most common in the UK and USA (Christodoulides, 2001). The digester was therefore assumed to be a vertical cylinder. The tank depth was taken to be equal to its diameter as recommended as an 'engineering rule of thumb' (Couper et al., 2005), and thus the area of the different sections of digester (top, bottom and sides) could be calculated for heat loss estimation.

The digester volume (m^3) was calculated from the daily flow rate from the harvesting system (m^3 day^{-1}) and the digester HRT (days).

5.1.8.2 **Heating**

Although anaerobic digestion can occur naturally at atmospheric temperature it is slow and digesters are typically heated to enhance biogas production (Rittmann and McCarty, 2001, Salter and Banks, 2008). The digester temperature could be set to any value between 10 – 70 °C. Default temperatures of 35 and 55 °C were assumed for mesophilic and thermophilic digestion.

The heat energy required by a digester is not only that to raise the temperature of the feedstock. Heat is also needed to replace that lost through the walls, roof and base of the digester. The heat required can be calculated using the following formulae:

Equation 23 Heat loss (kJ s^{-1})

$$H_l = UA\Delta T$$

Equation 24 Initial Heating of Feedstock (kJ s^{-1})

$$H = C_p Q \Delta T$$

Equation 25 Total Heat Requirement (kJ s^{-1})

$$H_t = H_l + H$$

where T is temperature (degree K), C_p is specific heat kJ kg^{-3} K^{-1}, Q is volumetric flow rate m^3 s^{-1} and U is heat transfer coefficient W m^{-2} K^{-1}.

The heat transfer coefficients U for digesters range from 0.3 to 5.2 W m^{-2} K^{-1} (US Enviromental Protection Agency, 1979) with typical values of 2 W m^{-2} K^{-1} (Energy Systems Research Unit, 1998). A digester with insulation 100 mm thick can have a heat transfer coefficient of 0.35 W m^{-2} K^{-1} (Banks, 2012). The model allows selection of any value of heat transfer coefficient for the top, sides and base, with a default value of 0.35 W m^{-2} K^{-1}, typical of a well-insulated digester.

5.1.8.3 **Mixing**

It is widely accepted that digesters need to be mixed to distribute enzymes and microorganisms and prevent the settling of solid particles (US Enviromental Protection Agency, 1979, Wu, 2012). It was assumed that the digester would be mixed continuously and a method of estimating mixing energy was needed and a number of approaches were considered.

The turbulent flow in impeller mixed systems is "inherently complex and not amenable to rigorous theoretical treatment" (Coulson and Richardson, 1999) and "there is no single design parameter that can be applied to all systems" (Gilbert, 1987). The root-mean square velocity gradient (G), which is function of energy per unit volume and viscosity, has been suggested as a measure for adequate reactor mixing with a value of between 50 to 80 s^{-1} being proposed as an adequate level of mixing in digesters (Meroney and Colorado, 2009). For low viscosity Newtonian liquid feedstocks that differ little in viscosity the simple measure of energy per unit volume is probably an adequate description of digester mixing, and the degree of mixing in anaerobic digesters has also been related to power delivered per unit volume of the digester (Christodoulides, 2001, Gilbert, 1987, Wu, 2012). The model therefore uses a power-per-unit-volume approach that can be set to any value.

Typically mixing power inputs are 5 - 8 W m^{-3} (Christodoulides, 2001, US Enviromental Protection Agency, 1979), although a more recent study has found that lower figures of 0.5 – 4 W m^{-3} may be sufficient (Wu, 2012). These recommendations are below the 'engineering rule of thumb' for blending of 40 - 100 W m^{-3} (Couper et al., 2005) and considerably below the average power consumption for mixed biochemical vessels of 1 - 2 kW m^{-3} (Uzir and Mat Don, 2007), but match those recently reported by the AD industry (Methanogen via Heaven personal communication, 2013). A default value of 5 W m^{-3} for AD mixing input was therefore used.

A Rushton turbine was assumed to be used. They are frequently used for industrial fermentation with low to medium viscosities. A power number of 5.5 (Uzir and Mat Don, 2007) and impeller diameter of 0.3 of the tank diameter (Couper et al., 2005) were taken as default values; other power numbers and diameters can be used. The speed of the impeller and mixing Reynolds number were calculated using standard mixing power equations (Perry and Chilton, 1973, Uzir and Mat Don, 2007). Root-mean square velocity gradient was also calculated. This additional data can be used to check that the flow is turbulent and for comparison, if required, with alternative mixing measurements

5.1.9 **Spreadsheet**

An example of the Excel spreadsheet, titled "maxmin simple flow 1 fix vol ad", is provided in the CD of accompanying materials. Some cells have a small red triangle in the top right corner. By placing the mouse pointer over the cell a comment box will open giving additional information or references relating to that cell. Data input cells are shaded grey.

5.2 Validation and calibration

The anaerobic digester section of the model was validated against an existing AD model, and checks on both loading rate and mass balance were built into the model.

5.2.1 Comparison with an existing AD model

In order to verify energy inputs and outputs of the AD section of the model an existing AD model was required. The AD4RD model developed by Salter and Banks (2008) was selected. It is a spreadsheet based tool, which calculates energy balances using a crop based AD system (http://www.ad4rd.soton.ac.uk). The AD4RD model produced energy inputs and biogas outputs for a range of terrestrial agriculture products and wastes, but not micro-algae.

5.2.1.1 Method and conditions of comparison

One feedstock in AD4RD is whey and the feedstock for comparison was assumed to be whey in both models. The micro-algal biogas energy balance was simplified to consist only of the anaerobic digester. The composition of the feed was adjusted to 13.2 % protein, 0.8 % fat, 76 % carbohydrate and 10 % ash with a total solids content TS of 6.1 % typical of that found in whey (de Wit, 2001). The average ambient temperature in the model was assumed to 10 ˚C and the Southampton location in AD4RD (average ambient temperature 10.6 ˚C)

The output of the modified model, with a simulated whey feedstock, was compared to the output of the AD4RD with whey as a feedstock. An Excel file, titled "whey ad energy inputs", is in the CD of accompanying materials.

5.2.1.2 Results and discussion

The results of comparison between the two anaerobic digestion models are shown in Table 11. There was fairly close agreement for the heating requirement; however energy for mixing was less than a quarter of the input calculated in the AD4RD model. The mixer efficiency in the model was set at 100 %. Adjusting for a typical small motor mixer efficiency of 50 % (Couper et al., 2005) increased the estimate of energy input, but it was still approximately half AD4RD model's estimate.

Table 11 Comparison with AD4RD AD model

			AD4RD		Model
Inputs					
Digestate			Whey		'Whey'
TS%			6.1		6.1
VS % of TS			90		90
Lipid % of TS					76
Carb' % of TS					13.2
Protein % of TS					0.8
% C conversion			95		95
HRT	days		18		18
Calculated Outputs					
CH_4 Yield	l CH_4 g^{-1}VS		0.45		0.43
% CH_4 in biogas			51		51
Biogas produced	m^3 day^{-1}		1162		1011
CH_4 produced	m^3 day^{-1}		593		532
Energy input					
Heat	kWh day^{-1}		800		763
Electric	kWh day^{-1}		220		51

The AD4RD model's energy input was based on the figures of Berglund and Borjesson (2006), which assumed that electrical energy for mixing was generated from natural gas with a conversion efficiency of <50%; "With 1 MJ of electricity corresponding to 2.2 MJ of primary energy, including distribution losses in the electricity grid and energy requirements in the production and distribution of natural gas" (Berglund and Borjesson, 2006). Further adjusting the models mixing energy for electricity production from natural gas gives a revised mixing energy input of 224 kWh day^{-1} that is in close agreement with 220 kWh day^{-1} estimated by AD4RD.

The data for gas output and input of the micro-algal biogas model and AD4RD are in reasonable agreement, and therefore this provides confidence that the model can be used to estimate biogas yields and energy inputs and outputs of micro-algal digestion.

5.2.2 Loading rate

A check was built into the system which compared digester loading rate to a maximum loading rate that could be set to any value. If the maximum loading rate was exceeded a warning message appeared and the digester volume was automatically increased to reduce loading rate. A default value of 6 kg VS m^{-3} day^{-1} (Banks, 2012) was assumed. All the variations of parameters in this research were within the assumed maximum loading rate.

5.2.3 Mass balance

A mass balance was carried out on the system and on the inflows and outflows of the digester. If the system is out of balance an error message appears. In all the runs discussed subsequently the system balanced.

5.3 Associated studies during model development

During the development of the model, equations and methods, that were considered and used, were used to assess areas which potentially had implications for the assumptions made in the model and for the operation of micro-algal biofuel plants. Three areas were considered:

a. The relative outgassing of oxygen in areas of the raceway
b. The de-oxygenation of raceway and deeper growth ponds due to micro-algal respiration.
c. The potential effect of bacterial growth on wastewater nutrients on the availability of CO_2 and nitrogen

5.3.1 Estimation of relative outgassing of oxygen in a raceway

Photosynthesis generates oxygen, but dissolved oxygen levels much greater than air saturation values inhibit photosynthesis (Chisti, 2007). The dissolved oxygen levels inhibiting micro-algal growth can be a problem in PBRs (Acién Fernández et al., 2013, Molina Grima et al., 2001). It has been 'generally assumed' that O_2 will be released to the atmosphere along the channels in a raceway (Mendoza et al., 2013b), but there have been reports of dissolved oxygen concentrations causing inhibition of micro-algal growth in raceways (Marquez et al., 1995, Mendoza et al., 2013b). Is the gaseous exchange sump or the raceway channel the main area controlling the level of dissolved oxygen?

5.3.1.1 Method of estimation

Stoichiometric analysis using a typical empirical micro-algal formula $C_1H_{1.83}O_{0.48}N_{0.11}$ (Chisti, 2007) estimates that photosynthesis will require 1.91 g of CO_2 for the growth of one gram of typical dry micro-algal biomass and will produce 1.6 g of O_2 giving a ratio of CO_2 consumed to O_2 produced of 0.83.

The following equilibrium relation between O_2 and CO_2 in a closed system was used

Equation 26

$$-0.83 = \frac{\cancel{k_{gt}}\,(C_{s\,O2} - C_{0\,O2})}{0.932\cancel{k_{gt}}(\,C_{s\,CO2} - C_{0\,CO2})}$$

Using Equation 26 the outgassing of O_2 relative to the transfer of CO_2 in the open channel sections of a raceway and the gaseous transfer sump were estimated, assuming typical flue gas concentrations of 12 % CO_2 and 9 % O_2. An Excel file, titled "oxygen relative outgassing", is in the CD of accompanying materials

5.3.1.2 Results and discussion

A plot of the outgassing of O_2 relative to the transfer of CO_2 in the open channel sections of a raceway and the gaseous transfer sump supplied with flue gas is shown in Figure 21.

The 0.83 rate of gas transfer for O_2 relative to CO_2 is achieved at lower concentrations in the raceway channels than in the sump indicating that the raceway may be more important in outgassing of O_2 than the sump. It is generally assumed in the literature that there is no major accumulation of DO in a raceway as it is released to the atmosphere along the channels (Mendoza et al., 2013b). These initial results confirm that the channels of the raceway are important in lowering O_2 concentration, but there may be still a significant build-up of O_2 which is further examined in Section 6.5. The results of these calculations also confirm the view of the literature that one of the advantages of raceways over PBRs is their potentially lower build-up of O_2 (Mendoza et al., 2013b).

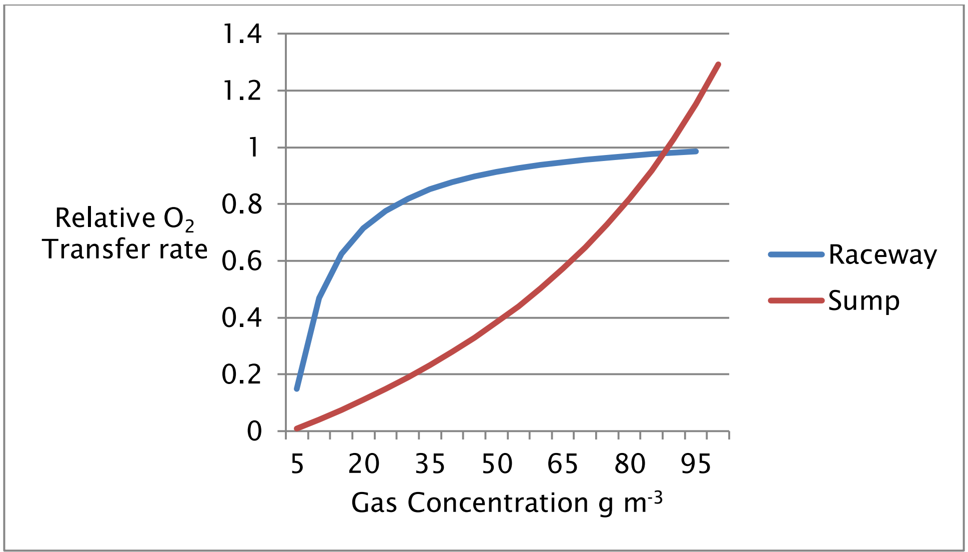


Figure 21 Outgassing of O_2 relative to CO_2 transfer in a flue gas sump and open raceway

5.3.2 Night-time de-oxygenation by micro-algal respiration

At night no O_2 is produced by micro-algal photosynthesis and O_2 is consumed by respiration. There has been a report of night-time crashes of deep micro-algal ponds (0.7 to 1.2 m) that were not mixed or aerated (Kroon private communication, 2012). Could this be due to de-oxygenation by micro-algal respiration? If de-oxygenation occurs in deep algal ponds could it occur in shallower micro-algal raceways?

5.3.2.1 Method of calculation

The QUAL-W2 water quality model uses a default value for algal respiration of 0.04 day^{-1} (Cole and Wells, 1995). Using the typical empirical formula for algae of $C_1H_{1.83}O_{0.48}N_{0.11}$ (Chisti, 2007, Grobbelaar, 2009a) gives a COD of 1.6 g g^{-1} VS. Assuming an average algal concentration of 0.035 % (typical algal suspensions 0.02 % - 0.05 % dry solids (Zamalloa et al., 2011)), therefore, the de-oxygenation rate by algal respiration is 22.4 g O_2 m^{-3} day^{-1} (0.04*350*1.6).

The re-aeration constant at 20°C for small still ponds is normally very low at between 0.1 and 0.2 day^{-1} (Chin, 2006). The saturation concentration of O_2 is 9 g m^{-3}. The maximum rate of oxygen transfer through the surface of the pond for an initial maximum DO concentration 9 g m^{-3} is, therefore, 1.8 g O_2 m^{-2} day^{-1} (0.2*9-0).

The Streeter Phelps equation (Chin, 2006) was modified, replacing the BOD removal constant by the algal respiration rate, to estimate de-oxygenation flux.

Equation 17 was used to calculate re-aeration coefficient k_{gt} for a range of raceway depths and velocities

5.3.2.2 **Results and discussion**

In an unmixed micro-algal pond, 1 m deep pond, the maximum de-oxygenation flux was estimated at 20.6 g m^{-3} day^{-1} and the oxygen in the system could be completely consumed in 10 hours. Night-time crashes in deep unmixed algal ponds, therefore, could be the result of de-oxygenation by micro-algal respiration. Supersaturation of oxygen from micro-algal photosynthesis during daylight, however, may considerably extend the time required for de-oxygenation.

A plot of re-aeration coefficient k_{gt} for depths of 0.1 to 1 m and fluid velocities 0.1 to 1 m s^{-1} is using Equation 17 is shown in Figure 22.

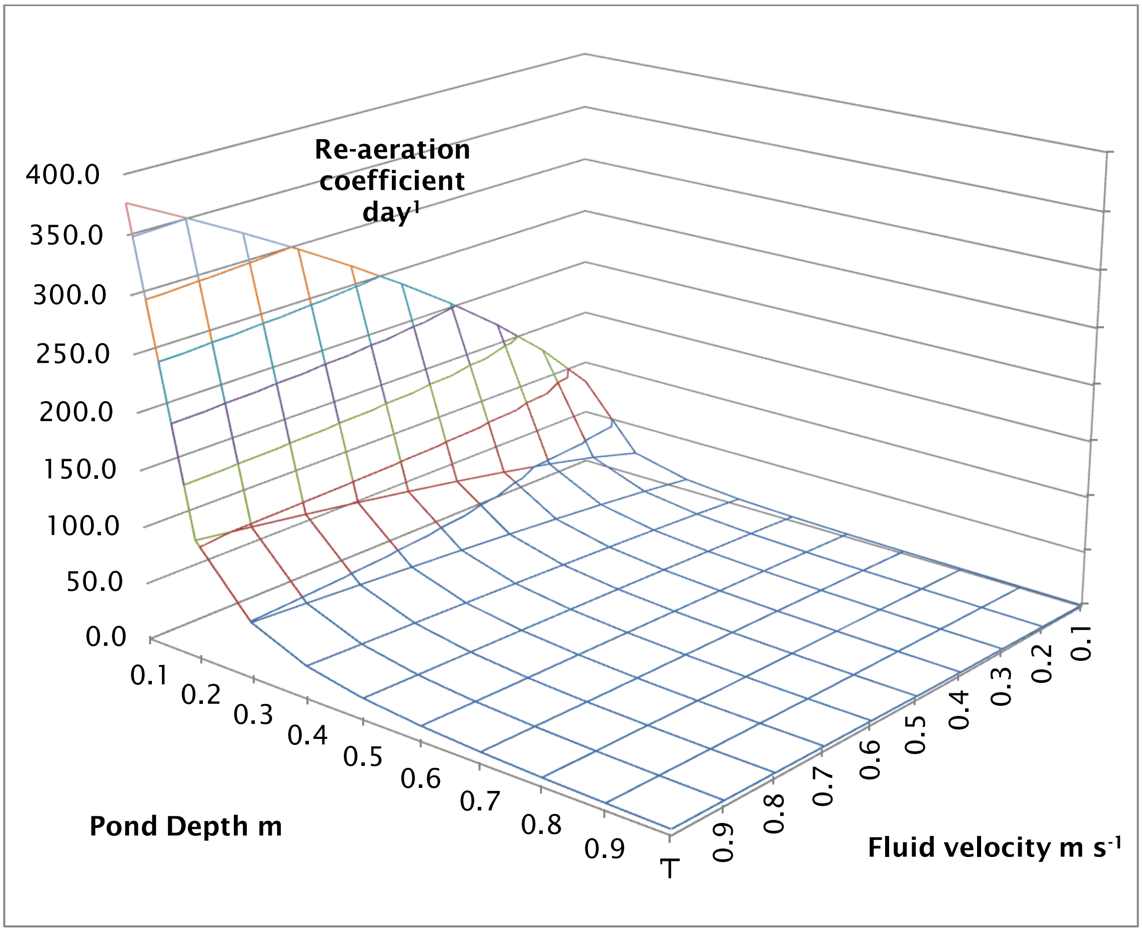


Figure 22 Influence of depth and velocity on re-aeration coefficient

Although re-aeration coefficient increases with increasing velocity the change is much less than the dramatic increase in re-aeration coefficient for depths below 0.5m. Increasing the re-aeration rate by reducing depth and keeping the fluid moving eliminates the problem of night-time de-oxygenation. There are no reports of night-time crashes in typical algal raceways with a depth of <0.3 m and a fluid velocity 0.15 to 0.3 m s^{-1} and this work confirms that night-time de-oxygenation will not be a major problem in a shallow flowing raceway. Night time aeration of open algal raceways to prevent oxygen depletion, that could double the energy cost of gas pumping, is not required for raceways. The assumption that gas pumping was only required for 12 hours used in Section 7.1.3 on micro-algal biodiesel and in the energy balance model developed in this work for micro-algal biogas production (Section 5.1.3) appears correct.

5.3.3 Bacterial CO_2 production and nitrogen utilisation

The growth of bacteria, particularly in wastewater, could have a significant influence on the requirement to supply both CO_2 and nutrients such as nitrogen. Could bacteria growing in wastewater supply significant CO_2 for algal growth or could their more rapid growth lead to nutrient depletion?

5.3.3.1 Method of estimation

Using an empirical formula of $C_5H_7O_2N$ for municipal sludge and bacteria grown on sludge, (Henze et al., 2008), together with an estimated empirical formula for 20 % lipid content algae $C_{4.37}H_{8.02}O_{1.99}N_{0.54}$ it was possible to calculate, using stoichiometric analysis, the CO_2 released and nitrogen available for micro-algal growth (not incorporated in bacterial biomass) for a range of bacterial respiration rates. Carbon dioxide released from wastewater nutrients by bacterial respiration and nitrogen not incorporated in bacterial biomass was assumed to available for micro-algal growth. An Excel spreadsheet, titled "bacteria n and CO_2 .xlsx", is in the CD of accompanying materials.

5.3.3.2 Results and discussion

As can be seen in Figure 23, for typical bacterial respiration rates of 30 to 50%, growth is restricted more by CO_2 released by respiration rather than nitrogen not incorporated in bacterial biomass. As bacteria respire over a 24-hour period and autotrophic micro-algae only grow in light the restriction of micro-algal growth due to supply of CO_2 will be even greater than this simple estimate.

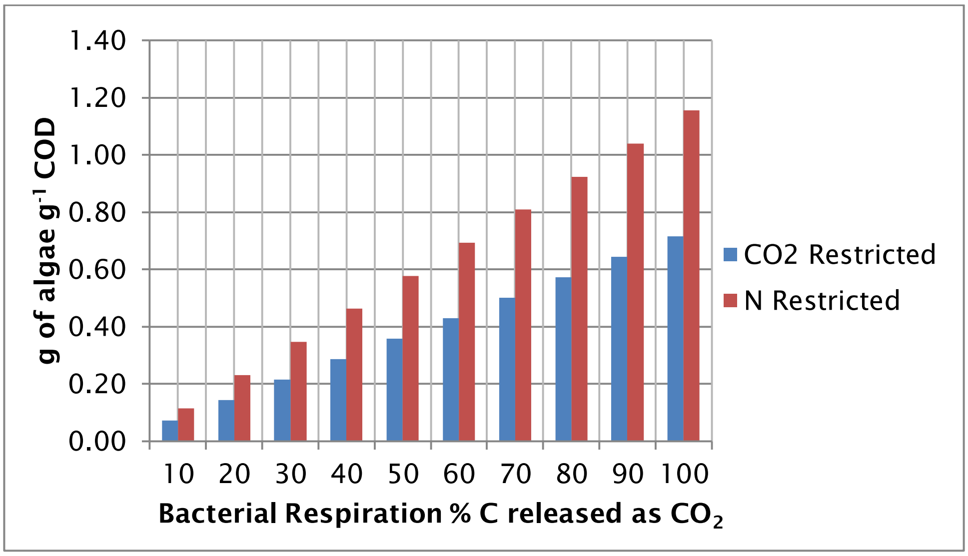


Figure 23 Potential micro-algal growth on typical municipal wastewater relative to bacterial respiration

Although CO_2 for micro-algal growth is provided by bacteria in facultative wastewater treatment ponds (Section 2.1.1.1), additional CO_2 is required to utilise all the nitrogen for micro-algal biomass. Bacteria also utilise some nitrogen and make it unavailable for micro-algal biomass growth. This may be problematic if micro-algae are the only required target product. However overall biomass yield, for biogas production by anaerobic digestion, may be very similar for the same quantity of available nitrogen as the typical composition of micro-algae and bacteria is similar (McKinney, 2004).

6. Modelling of scenarios

In this part of the work, the model developed in the previous sections was used to investigate the viability of a number of options and scenarios for micro-algal biofuel production in terms of energy output, and to identify the most critical parameters affecting net energy production. The model was first used to examine the effect of raceway dimensions on head losses, to establish whether a raceway of 1 hectare was feasible; and then to study energy input relative to biomass energy production and outgassing in a raceway system. A number of options were then examined involving operation at different digester temperatures, CO_2 concentration in the gas supply and hydraulic retention times in both the digester and raceway, in order to establish the relative importance of these parameters and to develop an overall ‘pragmatic' case. The pragmatic case was then used to examine scenarios based on a combination of different harvesting options and multiple raceway arrangements to establish net energy balances and assess the energetic viability of micro-algal biogas.

6.1 Head loss in raceways

The model was first used to look at the effect of channel width and length on the head losses in raceways with areas ranging from 103 m^2, typical of pilot scale, to 1 hectare, representing a likely size for a commercial scale plant (Section 2.2.6).

Output head loss results from the model are shown in Table 12. The head loss for a 1 hectare raceway of 0.052 m is agreement with that previously estimated in Table 9. The maximum head difference recommended by a paddlewheel manufacturer for commercial algal raceways is 0.076 m (Section 2.2.6) and thus a 1 hectare lined raceway appears to be possible.

Halving the width of a 219 m long raceway has little effect on head loss, but halving the length of a 20 m wide raceway has a significant effect, with head losses reduced from 0.052 to 0.041 m. A 20 m metre wide raceway 109 m long has a head loss similar to a raceway 50 m long and only 1 m wide, thus raceways should be as wide as possible. The effect of raceway width is further analysed in Section 6.3.1.

Table 12 Head loss in raceways of different dimensions

Raceway channel length	m	50	219	219	109
Raceway channel width	m	1	20	10	20
Area	m^2	103	10017	4694	5617
Head	m	0.040	0.052	0.052	0.041

6.2 Energy input relative to biomass energy production and outgassing in a raceway

The model was used to study the energy inputs and potential energy output, in the form of micro-algal biomass, or EROOI, for a raceway consisting of two 50 m long and 1 m wide channels, similar to the dimensions of the University of Southampton's experimental raceway in Spain (Mendoza et al., 2013a). A photosynthetic yield of 1.5 % was assumed, equivalent to a biomass yield of 13 g m^2 day^{-1}.

6.2.1 Effect of depth and velocity on energy ratio

The model was used to look at the effect of average fluid velocities from 0.15 to 0.45 m s^{-1} and raceway depths from 0.15 to 0.45 m on the ratio of energy input to potential biomass energy output. The results are shown in Figure 24. The biomass energy output of the raceway was estimated at 8.48 kWh day^{-1}. The energy input increases with increasing depth and fluid velocity: a 0.45 m deep raceway flowing at 0.45 m s^{-1} was estimated to use 4 kWh day^{-1}, or 48 % of the energy potentially available in the micro-algal biomass. Depth and fluid velocities should therefore be minimised. At depths below 0.15 m, however, it is difficult to achieve sufficiently even grading of the raceway bottom to ensure consistent flow around the entire raceway (Weissman et al., 1989); mixing problems and temperature variation may also occur at depths below 0.25 m (Lundquist et al., 2010). There appears to be no energy balance advantages to raceways being deeper than 0.3 m or flowing faster than 0.3 m s^{-1}.The typical raceway depths of 0.2 - 0.3 m (Aquafuels, 2011, Jimenez et al., 2003, Johnson et al., 1988, Lundquist et al., 2010) and velocities 0.15 -0.3 m s^{-1} (Borowitzka, 2005, Chiaramonti et al., 2013) therefore appear to be appropriate.

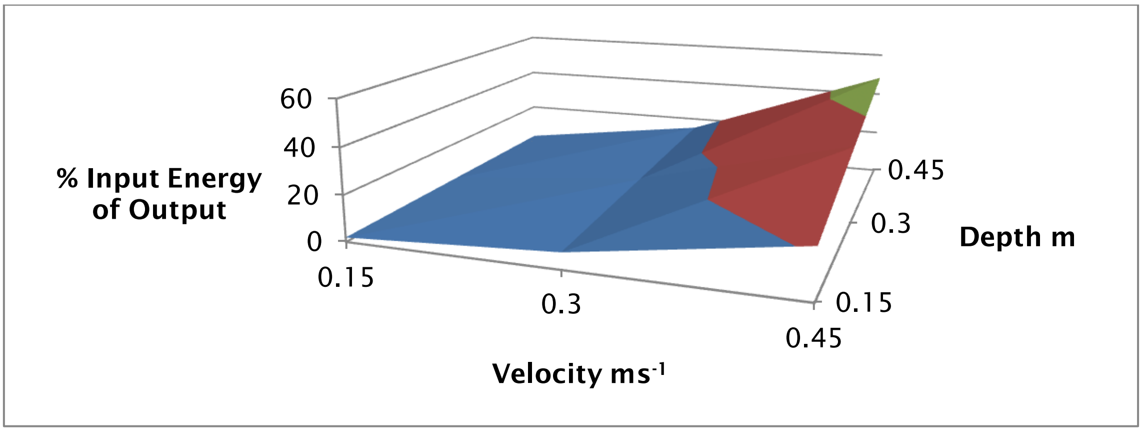


Figure 24 Effect of fluid velocity and raceway depth on the ratio of energy input to potential biomass energy output in a raceway

The ratio of blower to paddlewheel input energy is shown in Figure 25. Blower energy of 0.1 kWh day^{-1} is the main energy input into raceway at a depth of 0.15 m flowing at 0.15 m s^{-1}. The ratio of blower to paddlewheel input energy increases with reducing depth and fluid velocity. This change is the result of reducing paddlewheel energy rather than increases in blower energy.

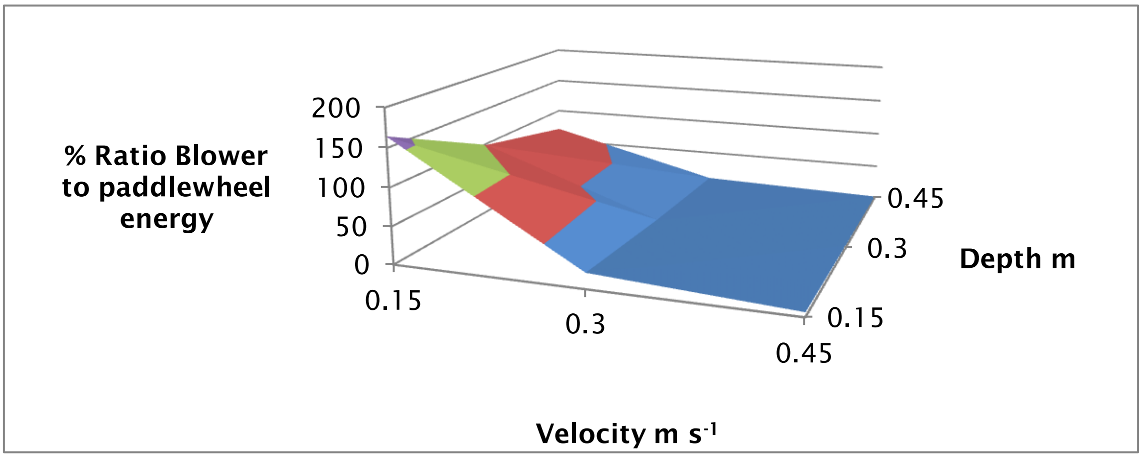


Figure 25 Effect of fluid velocity and raceway depth on the ratio of blower to paddlewheel energy

6.2.2 Effect of depth and velocity on outgassing of CO_2 in raceways

The model was used to look at the effect on the outgassing of CO_2 for average fluid velocities from 0.15 to 0.45 m s^{-1} and raceway depths from 0.15 to 0.45 m.

The results in Table 13 show that increasing fluid velocity from 0.15 to 0.45 m s^{-1} at the same depth (0.3 m) reduces relative outgassing, despite the increased energy input. This appears to be due to the shorter residence time in the raceway channels before returning to the gaseous exchange sump.

Reducing the depth from 0.45 m to 0.15 m at the same fluid velocity (0.3 m s^{-1}) increases the amount of CO_2 outgassed relative to the amount required for algal growth from 4 % to 34 %, confirming the suggestion of Lundquist et al. (2010) that high rates of carbon dioxide outgassing may also occur at depths below 0.25 m. The ratio of energy input to output is lower in the shallower raceway, however, and thus if the CO_2 is from a waste emission source, with little embodied energy or cost, raceways shallower than 0.25 m could be energetically and economically advantageous.

Table 13 Effect of velocity and depth on outgassing and energy ratio in raceways

Depth	m	0.3	0.3	0.3	0.15	0.45
Velocity	m s^{-1}	0.15	0.3	0.45	0.3	0.3
Ratio Input energy to output	%	2.4	11.1	34.5	7.1	15.2
Ratio CO_2 outgassed to growth requirement	%	11	8	7	34	4

6.2.3 Effect of width and length on outgassing and energy ratio

The model was used to look at the effect of channel width (1, 5 or 10 m) and length (50 or 100 m) on outgassing and energy ratio in raceways. The results are shown in Table 14. The energy return improves with both increasing raceway width and length. Table 14 also shows that the outgassing of CO_2 relative to the amount of CO_2 required for algal growth increases with increasing channel width and length: this is due to the longer residence time in the raceway channels before the flow returns to the gaseous exchange sump.

Table 14 Effect of width and length on outgassing and energy ratio in raceways

Width	m	1	5	10	1	5	10
Length	m	50	50	50	100	100	100
Ratio input energy to output	%	11.1	9.3	8.3	7.3	6.2	5.8
Ratio CO_2 outgassed to growth requirement	%	8	9	10	17	18	19

The outgassing of CO_2 will be more significant in large raceways. In a raceway of ~1 hectare (219 m by 20 m), the model predicts that flue gas losses due to outgassing are equivalent to 45 % of the amount required to meet algal growth needs.

Figure 26 shows the effect of raceway width on the ratio of blower energy to mixing energy, and on the ratio of raceway energy inputs to biomass energy output. The ratio of blower energy to mixing energy in the raceway increases with increasing width, but the ratio of input to output energy decreases with increasing width. It would thus appear that raceways should be as wide as practicable. The results therefore support the recommendation from the US Energy Department study of a length to width ratio of 11 to 1 (Weissman et al., 1989) for a raceway of 1 hectare (219 m by 20 m).

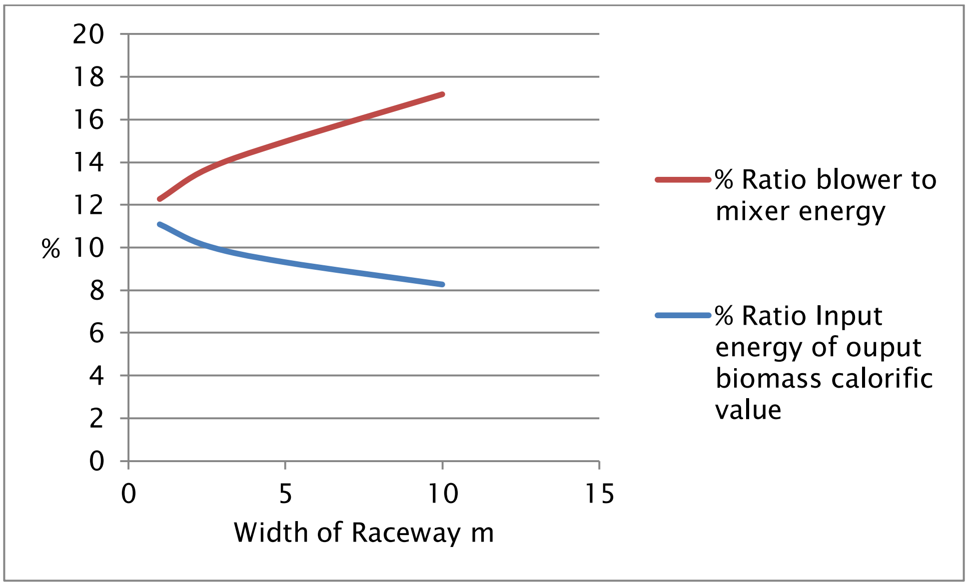


Figure 26 Effect of width on energy inputs in raceways

6.2.4 Effect of micro-algal photosynthetic efficiency on outgassing and energy ratio in raceways

The effects of variations in micro-algal photosynthetic efficiency on outgassing and energy ratio in a raceway are shown in Table 15. Photosynthetic efficiency has negligible effect on the amount of CO_2 outgassed relative to the amount of CO_2 required for algal growth.

Table 15 Effect of micro-algal photosynthetic efficiency on outgassing and energy ratio in raceways

Photosynthetic Efficiency	%	1.5	3	4.5
Ratio input energy to output	%	11.1	6.1	4.5
Ratio CO_2 outgassed to growth requirement	%	8	8	8

6.3 Effect of selected process parameters on the concentration factor required to achieve an EROOI of 1

The model was used to estimate the concentration factor required to achieve an energy return on operational energy invested of 1 for a variety of process options. For this purpose, equipment efficiencies and carbon conversion were taken as 100 % and the energy input for harvesting was assumed to be zero. In general the higher the harvesting concentration factor, the greater the energy input needed to achieve it (Axelsson, 2000, Bolhouse, 2010, Purchas, 1981), and process options with the lowest concentration factors will thus have the highest probability of achieving a positive energy balance. The concentration factor to achieve an EROOI of 1 was, therefore used as a measure of the potential energy efficiency for a range of process options.

6.3.1 Effect of raceway dimensions

The model was used to look at the effect of raceway width and depth on the concentration factor required to achieve an EROOI of 1, with the same range of dimensions as previously used in Section 6.1.

Table 16 shows the concentration factor to achieve an EROOI equal to 1 for various raceway dimensions. The small raceway (103 m^2) required a higher concentration factor to achieve a positive energy balance than the larger raceways. Halving the length or width of the largest raceway had little effect on the concentration factor needed to achieve a positive energy balance. Variation of raceway dimensions to give areas of 0.5 to 1 ha with widths of 10 -20 m made a negligible differences to the concentration factor, and this implies that dimensions of raceways of this size can be varied to suit land and operational constraints with minimal effect on the overall energy balance ratio. A raceway area of ~1 hectare (20 m channels 219 m long) was assumed in all subsequent scenarios used in this work.

Table 16 Concentration factor to achieve an EROORI equal to 1 for various raceway dimensions

Raceway channel length	m	50	219	219	109
Raceway channel width	m	1	20	10	20
Estimated Area	m^2	103	10017	4694	5617
Concentration Factor		30	22	22	22

6.3.2 Concentration factor to achieve an EROORI equal to 1 for various process options

The model was used to determine the concentration factor associated with an EROOI of 1 for a 'benchmark' case (scenario H) and a range of alternative process conditions. The parameters for the benchmark case (scenario H) are shown in Table 17 and are discussed in the subsequent Section 6.3.2.1. The various alternative scenarios, with a brief description of their variation from the benchmark (scenario H) and their results are shown in Table 18, with the results ranked by order of concentration factor with the lowest at the top. The specific assumptions are further detailed in the appropriate results and discussions sections below. The lower the concentration factor the lower the potential harvesting energy and overall energy input, and the higher the potential energy return.

Table 17 Assumptions for 3% PE, 25 g m^{-2} day^{-1} 'benchmark'

Data inputs		
Environmental		
Solar insolation	kWh m^{-2} $year^{-1}$	2000
Photosynthetic efficiency (PE)	%	3
Ambient temperature	°C	20
Raceway		
Area	m^2	10017
Depth	m	0.3
Average fluid velocity	ms^{-1}	0.3
Hydraulic retention time (HRT)	days	2
Gaseous exchange		
CO_2 concentration in supply	%	12
Harvesting		
Algal harvesting recovery	%	100
Concentration factor		22
Anaerobic Digestion		
% of "Buswell" estimated CH_4 yield	%	100
Hydraulic retention time (HRT)	days	20
Mesophilic digester temperature	°C	35
Equipment efficiencies		
Paddlewheel efficiency	%	100
Gas transfer efficiency	%	80
Blower efficiency	%	100
Pump efficiency	%	100
Percentage heat recovery	%	0
Heater efficiency	%	100
Mixer efficiency	%	100

Table 18 Concentration factor to achieve an EROORI equal to 1 for various process scenarios

Scenario	Input Assumptions	Concentration Factor
A	Maximum theoretical PE (11%)	6
B	4 day raceway HRT 3% PE	11
C	Reduced depth 0.15 m	11
D	10 day HRT in anaerobic digester	20
E	Pure CO_2 used for carbonation sump gas supply	21
F	Reduced raceway velocity 0.15 ms^{-1}	21
G	Reduced raceway mixing time 12 hours	21
H	**'BENCHMARK' 3% PE 25 g $m^{-2}day^{-1}$**	**22**
I	6% CO_2 used for carbonation sump gas supply	22
J	4 Day Raceway HRT 1.5% PE	22
K	60% harvest efficiency	23
L	30 day HRT in anaerobic digester	23
M	Typical Equipment efficiencies (Table 20)	27
N	Reduced Insulation 2W m^{-2} K^{-1}	28
O	60% of predicted maximum CH_4 yield by Buswell equation	38
P	Lower Yield PE 1.5%	45
Q	Thermophilic anaerobic digestion 55° C	48
R	Raceway located in Southampton with 3% PE Yield	73
S	Raceway located in Southampton with 1.5% PE Yield	158
T	Air used for carbonation sump gas supply (0.038% CO_2)	905>

6.3.2.1 'Benchmark' case

Scenario H (underlined) with a 3% PE with solar insolation of 2000 kWh m^{-2} day^{-1} and average temperature of 20°C was used as the 'benchmark'. The parameters of the 'benchmark' are shown in Table 17 while the key outputs are shown in Table 19.

Table 19 Model output for 'benchmark'

Outputs		
Concentration factor		22
Biomass yield	g m^{-2} day^{-1}	25
Algal concentration in raceway		0.017%
Biomass calorific yield	kWh day^{-1}	1646.57
% Algae DW concentration in feed to AD		0.36%
Calorific value of CH_4 production	kWh day^{-1}	1559.27
Energy Inputs		
Paddlewheel mixing of raceway	kWh day^{-1}	21.83
Supply pump energy	kWh day^{-1}	3.04
Harvest supply pump energy	kWh day^{-1}	10.15
AD supply pump energy	kWh day^{-1}	2.36
Harvest 'return' pump energy	kWh day^{-1}	5.54
Digestate 'return' pump energy	kWh day^{-1}	0.39
Total pumping energy	kWh day^{-1}	21.48
Blower energy	kWh day^{-1}	22.79
AD reactor volume	m^3	1420.7
AD heating energy	kWh day^{-1}	1322.76
AD mixing	kWh day^{-1}	170.44
Total AD input energy	kWh day^{-1}	1493.20
Total operational energy input	kWh day^{-1}	1559.30
Energy return on operational energy invested		1.0

The energy inputs in the 'benchmark' case for paddlewheel mixing, pumping and the blower to supply CO_2 in flue gas are of a similar order at 21.83, 21.48 and 22.79 kWh day^{-1} respectively. Each is equivalent to 1.4 to 1.5 % of the potential energy available in the methane produced. The major energy inputs are in the heating and mixing in AD digester, using 84.8 % and 10.9 % of the energy of methane produced. The benchmark case assumes no heat recovery, which would significantly reduce the required heating energy. The feed concentration of micro-algal VS to the digester is 0.36 % is lower than typically found in CSTR reactors. Increasing the VS feed concentration to 1 % reduces the mixing and heating energy by 64 % and 63 %, or a total energy reduction in AD of 551 kWh day^{-1}. This is equivalent to 0.13 kWh m^{-3} of harvesting energy, and if the additional energy input to the harvesting system to achieve the higher output concentration were less than this there would be an improvement in the net energy output. At low concentration micro-algal suspension (<5 %) the viscosity is similar to water with no significant increase with increased concentration up to 5 % (Section 2.3.6).

6.3.2.2 **CO_2 Concentration**

The highest concentration factor required and the biggest deviation from the 'benchmark' case was with the use of air (scenario T) rather than flue gas containing 12 % CO_2 as a source of carbon. At the required concentration factor of 900 the suspension would be 15 % DW micro-algae and may no longer be fluid. The energy to provide air to the medium was far greater than the energy in the algal biomass, and the net energy return was massively negative with an EROOI of 0.002. It is clearly not energetically viable to bubble air into a raceway as a source of CO_2.

Halving the CO_2 concentration of flue gas to 6 % (scenario I) had little effect on the required concentration factor compared to that for the 'benchmark' value of 12% CO_2. Flue gas typically has a CO_2 content of 6 to 13 %, and variation in CO_2 between these values has little effect on the concentration factor.

Using pure CO_2 (scenario E) marginally improved the required concentration factor from 22 to 21. The energy to produce and transport pure CO_2, however, is likely to be greater than the reduction in pumping cost of pure CO_2 compared to flue gas. As previously discussed (Section 2.2.7.2) the use of pure CO_2 is unlikely to be energetically and economically viable.

6.3.2.3 **Effect of factors increasing micro-algal concentration**

Scenarios A to C considered the effect of factors that could increase the concentration of the micro-algal culture, by increasing the maximum theoretical PE yield to 11% (scenario A), implementing a 4 day HRT in the raceway (scenario B), or reducing the depth to 0.15m (scenario C)
All these factors increased the concentration of the algal suspension leaving the raceway and entering the harvester, thus reducing the concentration factor required to achieve an EROOI of 1. Increasing yield reduces the required concentration factor, but as previously discussed (Section 4.1.4.4) yields above 3 % PE may not be achievable in practice.

Increasing the raceway HRT to 4 days may also not be practicable. Raceways appear to produce stable cultures at 2 to 3 days HRT (Weissman et al., 1989), but 'crashes' regularly occur at 4 days (Benemann et al., 1980). Zamalloa et al. (2011) recommended a raceway HRT of 2 days.

Reducing the depth to 0.15 m may also not be practicable due to problems in grading the raceway bed (Section 2.2.2.4). In this scenario, the head loss around the raceway was estimated at 0.082 m, greater than the maximum differential head recommended by a paddlewheel manufacturer and half the mean depth suggested as a maximum by Oswald (1988).

6.3.2.4 **Effect of paddlewheel operation factors**

In scenario F the fluid velocity in the raceway was reduced to 0.15 m s^{-1} and in scenario G paddlewheel operation was reduced to 12 hours per day during daylight. Both scenarios had a similar effect on the required concentration factor with both causing a reduction from 22 to 21.

The suspension of paddlewheel mixing at night (scenario G) may not be practicable as without mixing the micro-algae will settle to the bottom of the raceway. Reducing the fluid velocity in the raceway (scenario F) to 0.15 m s^{-1} reduced the paddlewheel input energy from 21.83 to 2.73 kWh day^{-1}, but increased the required blower energy from 22.79 to 23.37 kWh day^{-1} due to the greater CO_2 outgassing as a result of the longer circuit time. This ~10-fold reduction in paddlewheel energy was previously shown in Section 4.2, but was only 1% of the total micro-algal biogas process operation energy. The reduction of fluid velocity from 0.3 to 0.15 m s^{-1}, therefore may be less significant than it was considered to be in Section 4.2; however as previously concluded the fluid velocity should be the minimum to prevent micro-algal sedimentation and provide regular exposure of the cells to light.

6.3.2.5 Anaerobic digestion factors

Scenarios D, L, N, O and Q involved changes to AD parameters. Thermophilic anaerobic digestion processes can offer accelerated biochemical reactions and micro-algal growth rates with more rapid methane production and lower hydraulic retention times (Gavala et al., 2003). Operation of the digester at 55 °C (scenario Q) gave a required concentration factor of 48, reflecting the increased heat input. There was little difference, however, between required concentration factors for HRTs of 10 (scenario D), 20 (scenario H) and 30 days (scenario L) at 20, 22 and 23 respectively. It thus appears probable that the increased energy input of thermophilic temperatures will not be offset by shorter HRTs and that mesophilic operation is the preferred option energetically. In all scenarios considered the main energy input for AD, was heat. In thermophilic conditions 95 % of the energy requirement was for heat, in good agreement with 94 % found in a practical study of a thermophilic pilot digester (Espinosa-Solares et al., 2006).

Reducing digester insulation (scenario N) by increasing the heat transfer coefficient from 0.35 to 2 W m^{-2} K^{-1} increased the concentration factor from 22 to 28, a greater effect than found for changes in HRT. A heat transfer coefficient of 2 W m^{-2} K^{-1} is typical of 25 mm of insulation on a steel vessel (Spirax Sarco, 2012) and of many commercial AD plants (Basrawi et al., 2010, Wang et al., 2009). The heat loss from the AD reactor, with a heat transfer coefficient of 2 W m^{-2} K^{-1}, was estimated at 0.35 °C, well below the maximum loss of 1 °C day^{-1} recommended for the design and operation of agricultural digesters (Persson et al., 1979). Lower heat transfer coefficients have been reported for typical digesters (Banks, 2012, Banks et al., 2011b), however, and the heat loss from a well-insulated digester (0.35 W m^{-2} K^{-1}) is estimated at 0.05 ˚C day^{-1} with a corresponding reduction in the energy requirement for heating of over 23 %. Using the model, the reduction in heat loss for a digester with a volume of 1000 m^3 for a change in heat transfer coefficient from 2 to

0.35 W m^{-2} K^{-1} was calculated at 30.1 kWh day^{-1}. This reduction in heat transfer coefficient could be achieved by increasing the depth of fibreglass insulation from 0.025 to 0.1m (Banks, 2012, Spirax Sarco, 2012). This was calculated to require an additional 3.8 m^3 of insulation for a 1000 m^3 vertical-cylinder digester. An embodied energy for fibreglass 269 Kwh m^{-3} was assumed and it was calculated that the additional embodied energy in the fibreglass could be recovered in reduced heat loss in less than 34 days. These calculations strongly confirm the importance of controlling heat loss for an energy-efficient process.

Practical yields of methane from the anaerobic digestion of micro-algae are considerably below the theoretical. If the methane yield is reduced to 60 % (scenario O) of the theoretical maximum the concentration factor to achieve an EROOI increases from 22 to 38. This is a smaller effect than that associated with increasing the temperature from 35 to 55 °C, but greater than increasing the HRT or reducing the insulation. Maximizing methane yield from algae will be vital for an energy efficient process, and slower growing strains with a higher operational methane yield may have an energy balance advantage in terms of energy output over faster growing strains which are more recalcitrant.

6.3.2.6 **Climate conditions**

Scenarios R and S looked at the effect of siting the system in a location with an average temperature of 10 °C and insolation 1000 kWh m^{-2}, similar to Southampton, UK. At 3% PE (scenario R) the required concentration factor was increased to 73 compared to the 22 for the benchmark case (equivalent to the climatic conditions of typical target areas such as the south-west USA and south-west Africa). Micro-algal growth rates are often reduced in lower temperatures (Mata et al., 2010, Moheimani and Borowitzka, 2006, Olguín, 2003, Verma et al., 2010). A lower PE efficiency of 1.5 % (scenario S) was therefore assumed in addition to the average temperature of 10 °C and the estimated required concentration factor increased to 158.

6.3.2.7 **Harvesting and equipment efficiencies**

Actual equipment energy efficiencies and harvesting recovery rates for micro-algal biomass are less than 100%. Scenarios K and M therefore looked at the effects of introducing more realistic typical values for process efficiency.

6.3.2.7.1 **Harvesting efficiency**

For harvesting by sedimentation, typical micro-algal biomass recovery rates are 60% (Shen et al., 2009). Adopting a reduced harvesting recovery rate of 60 % (scenario K) marginally increased the required concentration factor, from 22 to 23. The presence of micro-algal biomass in the dilute stream exiting the harvesting system may not be a problem if it can be recycled to the growth system, but could represent an additional expense, a loss of resources and a potential waste management problem.

6.3.2.7.2 **Other equipment efficiencies**

In scenario M the typical paddlewheel (Section 2.2.3), pump, mixer, heater and blower efficiencies (Couper et al., 2005) were all assumed to be as shown in Table 20. The efficiencies assumed are at the upper end of typical values (Couper et al., 2005).

In this scenario the concentration factor increased from 22 to 27. This effect was less than that caused by reducing digester insulation in scenario N, but as in the case of improving insulation the increased cost and embodied energy of providing more energy efficient systems may be recovered in reduced operational energy.

Table 20 Typical equipment efficiencies

Paddlewheel efficiency	%	50
Blower efficiency	%	80
Pump efficiency	%	80
Heater efficiency	%	80
Mixer efficiency	%	80

6.3.2.8 **Summary of scenario results**

Table 18 shows the concentration factors for all of the scenarios considered ranked in order with the lowest at the top. The lower the concentration factor the lower the potential harvesting energy and overall energy input and higher potential energy return.

The highest concentration factor required and the biggest deviation from the 'benchmark' case was with the use of air (scenario T) rather than flue gas containing 12 % CO_2 as a source of carbon. The lowest concentration factors required were for the maximum theoretical PE yield 11% (scenario A), a 4 day raceway HRT (scenario B) or reducing the depth to 0.15m (scenario C).
In the scenarios involving changes to AD parameters (scenarios D, L,N, O and Q) the highest concentration factor was associated with using a thermophilic (55 ˚C) (scenario Q) rather than a mesophilic operating temperature (35 ˚C) (scenario H). The scenarios involving climatic conditions gave the second and third highest required concentration factors and thus confirmed the production of micro-algal biofuel in UK would be energetically challenging at best.

The model clearly provides a useful tool for evaluating the effect of different parameters.

6.4 Pragmatic case

The choice in Section 6.3 of equipment, harvesting and methane yield efficiencies of 100 %, although useful for assessing the effect of selected process variables on concentration factor, leads to an underestimate of the energy inputs of 'real systems'. In this part of the work, a pragmatic case was therefore defined to allow a more realistic and detailed analysis of energy balances of the entire micro-algal biogas production process. The equipment efficiencies used for this purpose were taken from Table 20. The pragmatic case assumed a methane yield of 60 % of the theoretical, equivalent to an estimated methane yield of 0.33 g CH_4 g^{-1} VS for 20 % lipid content algae corresponding with the highest quoted yields (Section 2.4.9). Recent studies found a 60 % yield for *Dunaliella salina* (Roberts private communication, 2012) and 59 -79 % yield for 5 commercially exploited micro algae (Zhao et al., 2012).

All other assumptions were as the benchmark case (Scenario H)

6.4.1 **Heat Recovery**

In scenario U it was assumed that there was no heat recovery and in scenario V heat recovery was assumed to be 50 %. Heat recovery in AD systems has been reported at 33 – 66 % (FEC Services, 2003) and 40 % (Puchajda and Oleszkiewicz, 2008). Boissevain (2012) found that year round operation of mesophilic AD system with average external temp of 10 °C was possible with a waste heat recovery of 40 %. As can be seen in Table 21 the use of heat recovery reduced the estimated concentration factor required from 52 to 30. Heat recovery of 50 % was thus assumed in all further analysis in this work.

Table 21 Effect of heat recovery and raceway evaporation on the concentration factor for the pragmatic case

Scenario	Input Assumptions	Conc' Factor
U	Pragmatic case - No heat recovery	52
V	Pragmatic case + Heat recovery (New Pragmatic Case)	30
W	Pragmatic case + Heat recovery + evaporation (10mm day^{-1})	30

6.4.2 **Evaporation**

Evaporation from open raceways can be considerable. The NREL study found average evaporation rates in the south-west USA of 5.7 to 6.2 mm day^{-1} (Sazdanoff, 2006). However rates of 10 mm day^{-1} or greater can occur (Ferguson, 1952, Ministry of Environment and Tourism Namibia, 2002) and evaporation could thus be 400 kg for each kg of dry algal biomass produced. Assuming an evaporation rate of 0 (scenario V) or 10 mm day^{-1} (scenario W) had little effect on the required concentration factor, which was 30 in both cases. The total pumping energy increased by 2 % due to the additional fluid flow to replace water lost by evaporation. The total operational energy increased by < 0.05%. Evaporation could be a significant factor in the process operation and cost of micro-algal biofuel, but had little effect on concentration factor, input energy and EROOI, and thus was assumed to be zero for the further use of the new pragmatic case in this work.

The new pragmatic case with zero evaporation and 50 % heat recovery (Scenario V) was then used as the baseline condition to study the effect of a number of combinations of processes for producing micro-algal biogas on the resulting energy balance. Options studied were centrifugation, settlement, flocculation and combinations of these harvesting methods, together with multiple pond and pumping and harvesting sequences. The specific assumptions are further detailed in the appropriate results and discussions sections below.

Based on the new pragmatic case (Scenario V) the model was also used to estimate the maximum dissolved oxygen concentration in the raceway.

6.5 Dissolved oxygen

The maximum dissolved oxygen concentration for the new pragmatic case (Scenario V) was calculated to be 22 g m^{-3}, in good agreement with reported DO concentrations of 20–25 mg l^{-1} reached at around noon in an experimental reactor using flue gas for carbon supplementation (Mendoza et al., 2013b). Although this is over twice the saturation level of O_2 in air, it is below the limit of 3 times considered to inhibit micro-algal growth (Camacho Rubio et al., 1999, Fernandez et al., 2001).

6.6 Micro-algal biogas production with centrifugal harvesting

In this scenario the model was used to estimate EROOI for the production of micro-algal biomass with harvesting by disc-stack centrifuge assuming the new pragmatic case (Scenario V). The harvesting recovery efficiency and concentration factor for disc-stack centrifuges, based on literature values, were assumed to be 90 % (Porteous, 1983, Shen et al., 2009) and 120 (Molina-Grima et al., 2003). Two values for energy input per unit volume were assumed of 1.4 (Goh, 1984) and 1.0 kWh m^{-3} (Molina-Grima et al., 2003). Both EROOIs were substantially below 1 (Table 22), with twice to three times more energy required for operation than is produced as biogas.

As discussed in Section 2.3, disc-stack centrifuges, can achieve micro-algal concentrations of more than 2 % DW. Even at a concentration of 10 %, however, the EROOI remained ≤ 0.5 with more energy being used than is produced (Table 22).

Table 22 EROOI for micro-algal biogas production with centrifugal harvesting

		Typical Concentration Factor		10% Algal Output	
Harvesting	%	90	90	90	90
Concentration factor		120	120	604	604
Algae DW concentration in feed to AD	%	2	2	10	10
Harvesting equipment energy input	kWh m^{-3}	1.4	1	1.4	1
Energy Return on Operational Energy Invested EROOI		0.3	0.5	0.4	0.5

The use of a disc-stack centrifuge as the sole harvesting method in the production of micro-algal biogas, as previously demonstrated for micro-algal biodiesel in Section 2.3.5.1.1, is not energetically viable.

Based on the model it was estimated that an energy input of less than 0.37 kWh m^{-3} for harvesting would be required to achieve an EROOI of ≥1.

6.6.1 Effect of viscosity

In all the previous scenarios the viscosity of the fluids in all sections of the model had been set to that of fresh water, as the viscosity of low concentration algal suspensions is similar to water (Section 2.3.6). High concentration algal suspension (>5 %) with a higher viscosity might occur after harvesting. The condition were assumed to be as above with a harvesting recovery rate of 90 %, a concentration factor of 120, and a harvesting energy input of 1 kWh m^{-3}. Two levels of post-harvest viscosity were used, 0.001 and 0.035 Pa-s, representing fresh water and an 8 % algal suspension (Bolhouse, 2010), in order to see the effect on process energy input. The results are shown in Table 23.

Table 23 Effect of postharvest viscosity on energy inputs

		Fluid viscosity postharvest	
	Pa-s	0.001	0.035
<u>Energy inputs</u>			
Raceway mixing	kWh day^{-1}	43.67	43.67
Pumping energies			
Supply pump energy	kWh day^{-1}	3.80	3.80
Harvest supply pump energy	kWh day^{-1}	12.68	12.68
AD supply pump energy	kWh day^{-1}	0.2604	0.262
Harvest "return" pump energy	kWh day^{-1}	7.37	7.37
Digestate "return" pump energy	kWh day^{-1}	0.08	0.08
Total pumping energy	kWh day^{-1}	24.20	24.20
Blower energy for raceway	kWh day^{-1}	28.48	28.48
Harvesting energy	kWh day^{-1}	1536.50	1536.50
AD energy			
Heating	kWh day^{-1} [1]	146.19	146.19
Mixing	kWh day^{-1}	34.57	34.57
Total AD input energy	kWh day^{-1}	180.76	180.76
Total operational energy input	kWh day^{-1}	1813.60	1813.60

There is only one small change in energy inputs due to the increase in post-harvest viscosity, for the post-harvest AD supply pump which increases 0.05% from 0.260 to 0.262 kWh day^{-1}. The overall change in total energy input was negligible, and therefore the assumption that the post-harvest viscosity was equivalent to that of fresh water was maintained for all further scenarios.

6.7 Maximum harvesting energy to achieve a net energy return

Assuming the new pragmatic case the model was used to estimate the maximum harvesting energy input to achieve an EROOI = 1 for a range of concentration factors. A 90 % harvesting recovery rate was assumed. Two values of raceway HRTs were used, 2 and 4 days. Results are displayed in Figure 27.

The harvesting energy input to achieve a net energy output is small at low concentration factors, and increases rapidly with increasing concentration factor. Harvesting energy reaches a maximum value of around 0.4 kWh m^{-3} for a 2-day HRT. At a 4-day HRT the concentration of micro-algal biomass leaving the raceway was estimated to be about 0.033 %, and unless higher concentrations can be achieved the maximum harvesting energy for a net energy return will be ~0.9 kWh m^{-3}.

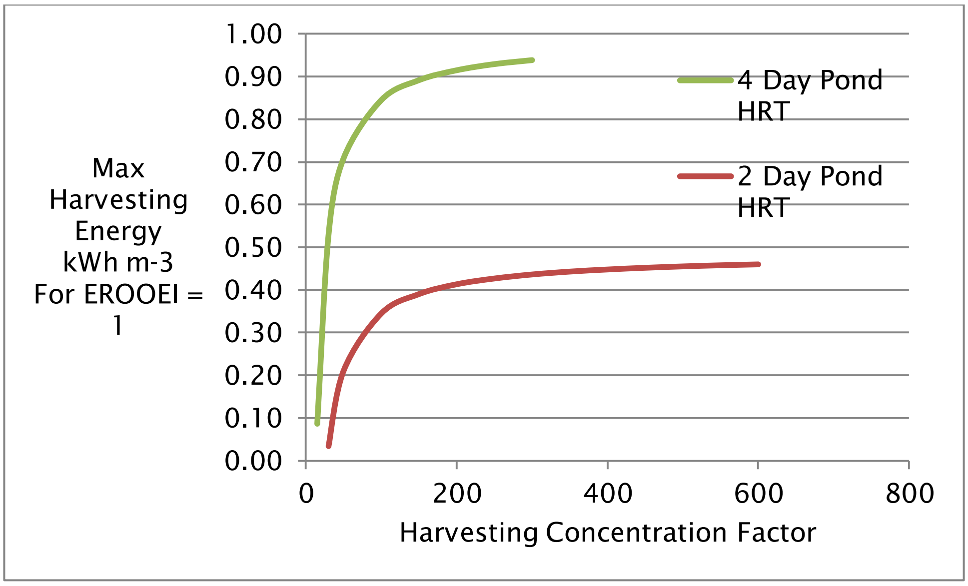


Figure 27 Maximum harvesting energy to achieve an EROOI = 1

6.8 Micro-algal biogas production with sedimentation harvesting

As previously discussed in Section 2.3.1, sedimentation or settlement is a low cost and low energy method of harvesting, but recovery rates and concentration factors are low. A recovery rate of 60 % with a concentration factor of 20 was assumed for three harvesting energy inputs:

a. 0.1 kWh m^{-3}. This value was chosen as it is typical of lamella settlers, a well-known and widely used technology for liquid-solid separation with limited space requirements (Uduman et al., 2010, Van den Hende et al., 2011)..
b. 0.05 kWh m^{-3}. This value was based on data from various manufacturers of lamella settlers which suggested that the energy requirement may be less than 0.1 kWh m^{-3} (various private communications with equipment manufacturers, 2012).
c. 0.05 kWh m^{-3}. This value is typical of scraped conical settlement tanks (Irish Environmental Protection Agency, 1997). For this case the head loss in the harvest supply pump was increased to 3 m to achieve an up-flow in the settlement tank ≤ 1.5 m $hour^{-1}$ (Forster, 2003).

Results from these three scenarios are displayed in Table 24. For all three harvesting energy inputs the EROOIs are below 1 indicating no net energy production. Concentration factors would need to be increased to 46, 36 and 31 and harvester output algal concentrations to 0.76, 0.60 and 0.51 % to achieve an EROOI of 1 for the harvesting energy inputs of 0.1, 0.05 and 0.005 kWh m^{-3} respectively.

Table 24 EROOI for micro-algal biogas production with sedimentation harvesting

		Typical recovery rate & concentration factor			EROOI of 1			Maximum DW concentration of 1.5 %		
Harvesting energy input	kWh m^{-3}	0.1	0.05	0.005	0.1	0.05	0.005	0.1	0.05	0.005
Algal harvesting	%	60	60	60	60	60	60	60	60	60
Concentration factor		20	20	20	46	36	31	91	91	91
Algae DW concentration exiting settler	%	0.33	0.33	0.33	0.76	0.60	0.51	1.50	1.50	1.50
EROOI		0.6	0.6	0.7	1.0	1.0	1.0	1.4	1.7	2.1

Micro-algal concentrations of up to 1.5 % have been achieved by sedimentation, and assuming this value EROOIs of 1.4 - 2.1 are achieved. A micro-algal biogas process with sedimentation harvesting can give a positive net energy balance; unfortunately, however the majority of micro-algae do not settle readily (Section 2.3.1.). Sedimentation could be a viable technique for harvesting of some micro-algae, or could potentially be combined with other harvesting techniques to increase the concentration of VS entering the digester.

6.9 Micro-algal biogas production with flocculation harvesting

The addition of flocculant can improve the rate of sedimentation, and has been suggested as a superior method to separate algae as it is suitable for large volumes and a wide range of micro-algae (Section 2.3.2).

Where flocculants are used to improve sedimentation harvesting, the embodied energy of the flocculant needs to be included in the harvesting and operational energy inputs. The embodied energy of a typical organic flocculant is 5.56 kWh kg^{-1} (Beal et al., 2012b). Alum (aluminium chloride), commonly used in water and wastewater treatment to remove algae, and perhaps the most effective inorganic flocculant for micro-algae, (Section 2.3.2.1), has an embodied energy of 4.04 kWh kg^{-1} (Maas, 2009). A major drawback of using mineral salts is that higher flocculant doses are required compared with organic flocculants, ranging from 120 - 1000 g m^{-3} compared to 1 - 10 g m^{-3} (Section 2.3.2). The harvesting recovery efficiency of flocculation-assisted sedimentation ranges from 50 to 90 % (Granados et al., 2012, Pushparaj et al., 1993, Shen et al., 2009) and the concentration factor from 35 to 800, although concentration factors of 800 are not typical for micro-algae (Knuckey et al., 2006, Molina Grima et al., 2003).

Flocculants need to be mixed into the micro-algal suspension prior to settlement. A Root Mean Square Velocity Gradient (G) for mixing of 300 s^{-1} was assumed, a value recommended by the Environmental Protection Agency and IWA for flocculation of municipal wastewater (Chen et al., 1998, International Water Association, 2010). This produced a flocculant mixing energy input of 90 W m^{-3}, in reasonable agreement with the 'engineering rule of thumb' to mix flocculants of 100 W m^{-3} (Couper et al., 2005, Sinnott, 2005, Stephenson, 2009).

The EROOI was estimated for alum at a dosage of 120 g m^{-3}, at concentration factors of 35 and 800 and an algal recovery rate of 90%; and for an organic flocculant at a dosage of 1 and 10 g m^{-3} and harvesting recovery efficiencies of 70 and 90 %. The results are shown in Table 25.

Table 25 EROOI for micro-algal biogas production with flocculation

		Alum			Organic Flocculant			
		0.005 kWh m^{-3} Settlement input 3m harvest pump head						
Algal harvesting	%	90	90		90	90	70	70
Concentration factor		35	800		35	35	35	35
Flocculant dose	mg l^{-1}	120	120		10	1	10	1
Algae DW concentration exiting settler		0.58%	13.26%		0.58%	0.58%	0.58%	0.58%
EROOI		0.6	1.0		1.1	1.2	1.0	1.1
Embodied energy as % of energy produced		65	65		10	1	13	1

Alum at a dosage of 120 g m^{-3}, the lowest found in the literature, achieves an EROOI of 1 only at the highest concentration factor for flocculation found in the literature of 800. The embodied energy in the alum was 65 % of the total energy produced. Flocculation by alum is therefore not a viable option for the production of micro-algal biogas.

The lower dosages of organic flocculant, despite their higher embodied energy per unit of mass, resulted in a lower input energy for flocculation than alum, with the embodied energy in the flocculant being 1 to 13 % of the energy output. The EROOI for organic flocculants were estimated at between 1.0 and 1.2 for the lowest suggested concentration factor of 35, and thus organic flocculants may be a viable harvesting method, especially if higher concentration factors are achieved. EROOI could be improved by combining flocculation with other harvesting techniques.

6.10 Micro-algal biogas production with a combination of harvesting methods

The next scenarios considered a combination of sedimentation with centrifugation and flocculation with centrifugation to achieve an estimated output concentration of 10 % DW micro-algae from the harvesting operation. The feedstock concentration of 10 % was selected as being at the upper end of that reported as typically encountered with CSTR digesters (Rittmann and McCarty, 2001, Wilkie, 2005); micro-algal suspensions above 10 % will also behave as non-Newtonian fluids with a high viscosity and may be problematic to pump (Section 2.3.6.).

The parameters were as the new pragmatic case (scenario V) with the harvesting concentration factors, recovery rates and harvesting energy inputs as previously used in Sections 6.6, 6.8 and 6.9.

The results are displayed in Table 26. All the combinations of harvesting assessed produced a positive net energy output with EROOIs ranging from 2.5 to 3.8. Settlement and flocculation greatly reduces the flow rate of material entering the centrifuge, and thus the energy input for centrifugation.

Table 26 EROOI for micro-algal biogas production with a combination of harvesting methods

		Sedimentation			Flocculation			
		Centrifugation			Centrifugation			
Harvesting					Organic 1 mg l^{-1}		Organic 10 mg l^{-1}	
Recovery rate sedimentation/floc'	%	60	60	60	70	90	70	90
Concentration factor sediment'/floc'		20	20	20	30	30	30	30
Recovery rate centrifugation	%	90	90	90	90	90	90	90
Concentration factor centrifugation		30	30	30	20	20	20	20
Harvesting equipment sedimentation	kWh d^{-1}	0.005	0.005	0.005	0.005	0.005	0.005	0.005
Harvesting equipment centrifugation	kWh m^{-3}	1.4	1	0.35	1	1	1	1
Energy output								
Calorific Value of CH_4 production	kWh d^{-1}	505.2	505.2	505.2	589.4	757.8	589.4	757.8
Energy inputs								
Raceway mixing	kWh d^{-1}	43.7	43.7	43.7	43.7	43.7	43.7	43.7
Total pumping energy	kWh d^{-1}	29.5	29.5	29.5	29.4	29.5	29.4	29.5
Raceway blower energy	kWh d^{-1}	28.5	28.5	28.5	28.5	28.5	28.5	28.5
Harvesting energy	kWh d^{-1}	72.2	53.8	23.8	52.4	62.6	129.2	139.4
Total AD input energy	kWh d^{-1}	24.3	24.3	24.3	28.0	35.5	28.0	35.5
Total operational energy input	kWh d^{-1}	198.1	179.7	149.7	182.0	199.7	258.8	276.5
Net energy	kWh d^{-1}	307.1	325.5	355.5	407.5	558.1	330.6	481.3
EROOI		2.5	2.8	3.4	3.2	3.8	2.3	2.7

The concentration factor required for centrifugation following sedimentation is only 30, 25 % of the typical centrifugation factor of 120 (Molina-Grima et al., 2003). The concentration factor achieved by a disc-stack centrifuge is proportional to the flow rate (Axelsson, 2000, Porteous, 1983, Purchas, 1981), and this lower concentration factor could therefore allow a higher flow rate through the centrifuge. The energy per unit of flow for each unit of concentration factor, known as Dewatering energy D_w, can be expressed as (Bolhouse, 2010, Mohn, 1988):

Equation 27

$$D_W = \frac{E}{C_f}$$

Where E is the energy of centrifugation per unit volume and C_f is the concentration factor. For a centrifugal energy consumption of 1.4 kWh m^{-3} and a concentration factor of 120, D is 11.7 W m^{-3}, and for a concentration factor of 30 the centrifugal energy consumption would be reduced to 0.35 kWh m^{-3}. Assuming a value of 0.35 kWh m^{-3} in the estimate of energy inputs improves the EROOI to 3.4 for sedimentation with centrifugation. This reduced energy input for disc-stack centrifugation needs to be verified experimentally for large-scale equipment as it not possible to 'precisely predict' the energy requirement (Porteous, 1983), and the linearity of the relationship between concentration factor and flow rate drops off dramatically above a critical flow rate specific to each centrifuge (Axelsson, 2000). The reduction in required concentration factor following initial concentration by sedimentation or flocculation may, however, reduce the centrifuge energy input compared to typical literature values of 1 to 1.4 kWh m^{-3}.

Flocculation by an organic flocculant may be a more reliable and widely applicable means of pre-concentration than sedimentation, but the concentration of flocculant is critical to the energy balance. At a flocculant dosage of 1 mg l^{-1}, for harvesting recovery rates of 70 and 90 %, the EROOI is 3.2 and 3.8 i.e. higher than that for sedimentation and centrifugation at the same centrifuge energy input (Table 26). At a flocculant dose of 10 mg l^{-1}, however, the EROOI is lower than for settlement. Flocculation at low doses followed by centrifugation for typical concentration factors and harvest recovery rates can produce EROOI greater than 3 for the production of micro-algal biogas. An EROI of 3 has been suggested as the minimum that is viable to 'support continued economic activity' (Clarens et al., 2011a, Hall et al., 2009). Micro-algal biogas production using flocculation and sedimentation could therefore be viable, but a low-cost low-dose (~1.0 mg l^{-1}) organic flocculant that is broken down in the digester is required, together with low or no cost and embodied energy nutrients and CO_2.

6.11 **Multiple raceways**

The model assumed a single raceway supplying biomass to a single digester, but commercial digesters can have a considerably greater volume (Basrawi et al., 2010) than that estimated in previous scenarios. The required reactor volume assuming a single raceway in the sedimentation centrifugation case was estimated at 28 m^3. As the reactor volume increases its surface area relative to volume decreases and thus the heat loss per unit volume will also decrease.

The digester volume in the model was therefore set to 3000 m^3, typical of large-scale commercial digesters. It was estimated that the digester would need 108 x 1 hectare growth ponds. The assumptions used were as those in the sedimentation centrifugation scenario, but with 108 raceways supplying a single digester. An average minimum pumping distance from the pond to the digester of 425 m was estimated based on geometrical considerations. The EROOI was calculated for no additional pumping distance (not possible, but used for initial comparison with the single raceway system) and with a 425 m pumping distance, for 3 scenarios:

a. Pumping (distance 425 m) from each raceway to a single central harvesting 'zone' (sedimentation centrifugation) adjacent to the digester.
b. Sedimentation adjacent to each raceway and then pumped (distance 425 m) to a single central centrifugation harvesting 'zone' adjacent to the digester.
c. Harvesting (sedimentation centrifugation) adjacent to the raceway and then pumped (distance 425 m) to the digester.

A further scenario of 4 raceways supplying a single digester was also considered. The pumping distance from the raceway through the harvesting system was assumed to be the same for each raceway in the 4 raceway scenario as in the single raceway sedimentation centrifugation scenario.

The outputs for the various process configurations described above are shown in Table 27.

Table 27 Effect of number of raceways on EROOI

		Single raceway & AD	Multiple hectare raceway supplying 1 AD unit (~3000 m^3)				4 raceways & 1 AD
			No additional piping	425 m pipe run	Settle locally	Harvest locally	
Number of hectare raceways		1	108	108	108	108	4
AD volume	m^3	28	2,987	2,987	2,987	2,987	111
EROOI		2.8	2.9	1.4	2.9	2.9	2.8
Total pumping energy per hectare raceway	kWh day^{-1} ha^{-1}	29.5	29.6	214.9	29.7	29.6	29.5
Total AD heat energy per hectare raceway	kWh day^{-1} ha^{-1}	20.1	16.1	16.1	16.1	16.1	18.2

With no additional piping, compared to a single raceway, the EROOI improves from 2.8 to 2.9. This is due to the reduction in the heat energy requirement of the digester from 20.1to 16.1 kWh day^{-1} ha^{-1}, as a result of the lower surface area per unit volume. However, it is not possible to have 108 raceways supplying a single digester without additional pipework. When the additional average pipe run of 425 m to transport the micro-algal suspension to the harvesting operation adjacent to the digester is included, the EROOI reduces to 1.4 as a result of a more than 7-fold increase in the total pumping energy.

Local sedimentation reduced the amount of liquid to be pumped 20-fold, and increased the EROOI to 2.9, but local harvesting with settlement and centrifugation brings no additional improvement. Although concentration during local harvesting reduces pumping energy it does not produce a reduction in the number of items of harvesting equipment with the consequent economies of scale and reductions in capital cost.

Groups of 4 raceways could be arranged around a central harvesting and digester unit with little or no additional piping. Although there was no improvement in EROOI this may allow lower equipment costs due to economies of scale.

Pumping energy should be minimised and must be taken into account when considering multiple raceway layouts.

6.12 Energy supply

The EROOI does not take into account the generation and transmission costs of input energy. These, as shown in Section 5.2.1.2, can more than double the energy requirement. The energy transmission costs can be reduced by producing power locally and 'parasitically' from the biogas produced. Combined heat and power (CHP) is the simultaneous production of electricity and heat from a fuel, and is more efficient than conventional separate electrical and heat generation, as shown in Figure 28. CHP units have been widely used to exploit biogas from AD. Biogas burned in a CHP unit requires minimal or no gas scrubbing to remove hydrogen sulphide (H_2S) and other impurities (Salter and Banks, 2008, Wellinger and Lindberg, 2001). However upgrading of the biogas is normally required if the gas is used as a vehicle fuel or added to the natural gas grid. The upgrading of biogas typically uses ~11 % of the energy content in the biogas (Berglund and Borjesson, 2006).

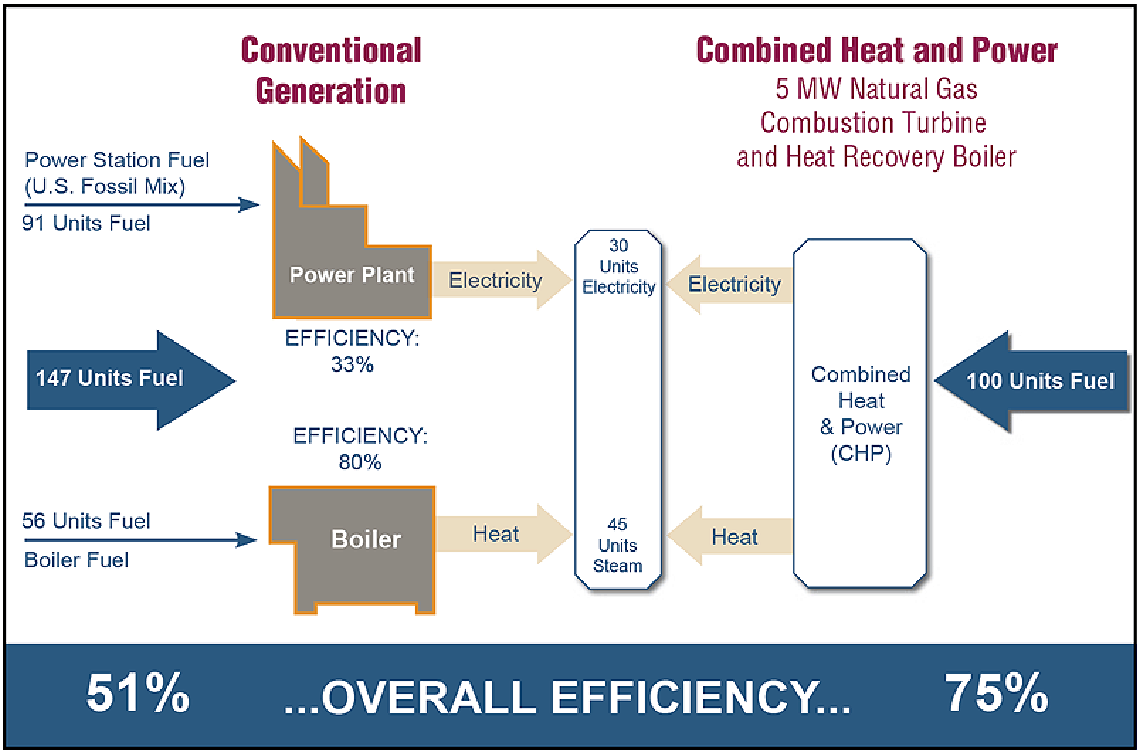


Figure 28 CHP (US Environmental Protection Agency, 2013)

If the biogas produced from micro-algae can be exploited directly in a CHP unit, can the heat and power produced be used efficiently for the process electrical and heating energy in micro-algal biogas production? The ratio of electrical to heat energy produced from a CHP unit is ~0.67.

Using the scenarios for sedimentation centrifugation and flocculation centrifugation (Section 6.10) the ratio of electrical to heat energy required by the micro-algal production process was estimated using the model.

Table 28 displays the ratio of electrical to heating energy needs in micro-algal biogas production. The ratio is very different to that generated by CHP, with more electrical than heat energy required. If heat from the CHP unit is not fully exploited or if separate electrical and heat generators are used the energy input into the micro-algal biogas process more than doubles and EROOI is reduced. The sedimentation centrifugation scenario requires 178 and 20.1 kWh day^{-1} of electrical and heat energy inputs, and produces 505 kWh day^{-1} as methane (HHV) from the micro-algal biogas. If all the biogas is burnt in a CHP unit, at the output ratio in Figure 28, then a 151 kWh day^{-1} of electricity and 227 kWh day^{-1} of heat are produced with 126 kWh day^{-1} of operational energy losses in the CHP unit. If only the 20 kWh day^{-1} of heat required by the process is used the total loss of energy from CHP unit in operational losses and 'waste heat' totals 333 kWh day^{-1}. The total input and waste energy is thus 531 kWh day^{-1} giving an EROOI of less than 1.

In order for the micro-algal biogas to be energy efficient a local source for exploitation of the excess heat generated needs to be found. Finding local uses for excess heat is one of the major operational problems in the current exploitation of CHP, and not just for micro-algal fuel production.

Table 28 Ratio of electrical to heating energy in micro-algal AD

		Settlement			Flocculation			
		Centrifugation			Centrifugation			
Harvesting					Organic 1 mg l^{-1}		Organic 10 mg l^{-1}	
Harvesting settlement/floc'	%	60	60	60	70	90	70	90
Concentration factor settlement/floc'		20	20	20	30	30	30	30
Harvesting centrifugation	%	90	90	90	90	90	90	90
Concentration factor centrifugation		30	30	30	20	20	20	20
Harvesting equipment settlement	kWh day^{-1}	0.005	0.005	0.005	0.005	0.005	0.005	0.005
Harvesting equipment centrifugation	kWh day^{-1}	1.4	1	0.35	1	1	1	1
Electrical energy	kWh day^{-1}	178.0	159.6	129.6	150.2	161.9	150.2	161.9
Heating energy	kWh day^{-1}	20.1	20.1	20.1	23.2	29.2	23.2	29.2
Ratio Electrical to Heat Energy		8.8	7.9	6.4	6.5	5.5	6.5	5.5

6.13 Conclusions from modelling

The production of micro-algal biogas is energetically possible with potential EROOIs over 3 using the technologies proposed, but requires:

a. Favourable climatic conditions. The production of micro-algal biofuel in UK would be energetically challenging at best.
b. Achievement of 'reasonable yields' equivalent to ~3 % photosynthetic efficiency (25 g m^{-2} day^{-1})

c. Low or no cost and embodied energy sources of CO_2 and nutrients from flue gas and wastewater
d. Mesophilic rather than thermophilic digestion
e. Adequate conversion of the organic carbon to biogas (≥ 60 %)
f. A low dose and low embodied energy organic flocculant that is readily digested, or micro-algal communities that settle readily
g. Additional concentration after flocculation or sedimentation
h. Exploitation of the heat produced from parasitic combustion of micro-algal biogas in CHP units
i. Minimisation of pumping of dilute micro-algal suspension

The model provides a powerful assessment tool both for comparison of alternative options and potentially for benchmarking real schemes.

7. Case studies

In order to test the approaches and methods previously discussed and to determine their usefulness in assessment of an algal biomass cultivation scheme, a case study was carried out in which values calculated in accordance with the above were then used in conjunction with equipment manufacturers' data for outline sizing of a potential plant and to establish an initial energy balance. It was originally intended to use the model on the FP7 All-Gas project, which is based on raceways cultivation plus anaerobic digestion for biogas production; but this project was insufficiently advanced to allow the acquisition of good cost data for the economic part of the assessment. It was therefore decided to look at a scheme initially intended for biodiesel production in conjunction with Pure Energy Fuels Ltd.

Pure Energy Fuels Ltd was a UK-based biodiesel supplier interested in producing biodiesel from micro-algae. No commercial systems are available for this purpose, and an outline micro-algal biodiesel production process design and initial energy input assessment was therefore developed. The extraction of oil from biomass and the production of the biodiesel were to be achieved by extraction systems used for terrestrial plant oils and trans-esterification with methanol in the presence of an alkaline catalyst. Both processes were familiar to the company and the previous cost data were available.

Information for the Pure Energy Fuels Ltd case study was initially gathered during 2008 and 2009, with a follow-up visit for further data collection in 2013.The technical scheme considered was applicable to a range of locations with similar climatic conditions, but site-specific data was gathered for Namibia.

Pure Energy Fuels Ltd did the economic assessment of the biodiesel scheme, but as part of the current research another economic assessment was prepared for co-production of *Dunaliella* and salt, using data obtained directly from manufacturers of relevant equipment on the performance and costs of their equipment.

7.1 Micro-algal biodiesel production

7.1.1 System design

An open raceway system was selected as the most cost effective system for growing large quantities of algal biomass for biofuel (Section 2.1). Growth of a single micro-algal species for biodiesel feedstock in an open raceway was considered not to be feasible, and it was anticipated that a mixed culture could be established that was adapted to the local environment, stable and produced reasonable yields. The culture might differ between locations and perhaps also vary over the year as climatic conditions varied. This approach has recently been endorsed by Kazamia et al. (2012), who suggested the development of a 'synthetic ecology' where a robust community of multiple algal species growing in a consortium offers the potential both of increased yields, and the 'crop protection' necessary for wide-scale commercialisation. 'Crop protection' from invasion by contaminants, predators and competitors is likely to be particularly difficult in open culture. "A synthetic community, by its nature of having many of the available niches already occupied, can serve as a mechanism for crop-protection" (Kazamia et al., 2012). A yield based on 3 % photosynthetic efficiency was selected as a realistic and achievable target, equivalent to approximately 25 g m^{-2} day^{-1} (Section 4.1.). A target of 20 % useable oil content for biodiesel was also considered as achievable.

Nutrient costs have been shown to be an important factor in the cost and energy requirement of micro-algae production (Section 2.2.7.1). It was assumed that nutrients could be provided by wastewater, reducing fertiliser costs and embodied energy inputs. Carbon dioxide is another important requirement and a potential cost for micro-algal growth, and it was anticipated that the plant could be sited adjacent to a large producer of carbon dioxide, such as a power station or cement works. The carbon dioxide cost and embodied energy would, therefore, be negligible (Sections 2.2.4 & 2.2.7.2).

As noted earlier, the most common method of mixing and circulation in open raceways is by means of a paddlewheel (Section 2.2.3); while for gaseous transfer of carbon dioxide a sump is the most common method. Putt (2007) at Auburn University, Alabama proposes a design combining a gaseous transfer sump with a new low-cost cantilever sludge pump (Figure 29). This design was initially considered worthy of further attention because it held the possibility of additional refinement through combining separation by flotation with mixing and gaseous transfer. Discussions with Auburn University and with gas blower manufacturers suggested, however, that a modified cantilever pump gaseous sump designed to achieve flotation separation of micro-algae would require excessive amounts of operating energy to obtain the small bubble size required. The cantilever pump was found to be substantially more expensive than a paddlewheel, with the cost of pumps to power a one hectare raceway being 2 - 3 times higher than a paddlewheel at an installed power 2.5 - 3 times higher (various private communications with equipment manufacturers, 2008). It was therefore decided to base the proposed system on a paddlewheel and a separate gaseous exchange sump, as in the model developed for the current work (Section 5).

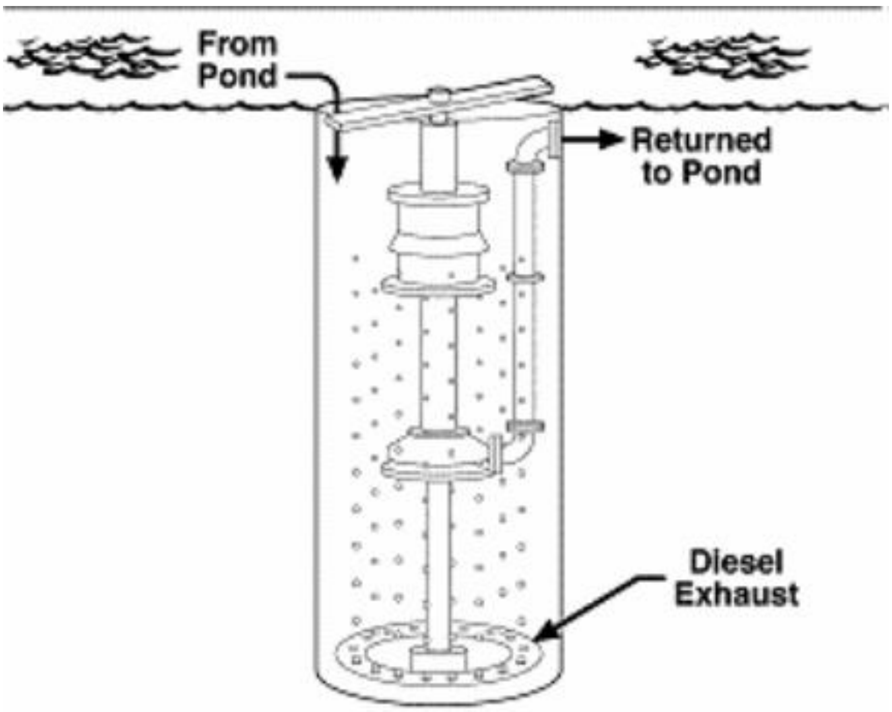


Figure 29 Gaseous transfer sump with cantilever pump (Putt, 2007)

A raceway area of one hectare (Sections 2.2.6) was used for all costing and energy calculations, with a channel width of 25 m and length 200 m (Sections 6.1 & 6.2.3). It was considered preferable to use a lined system (Section 2.2.1). It has been suggested that lined ponds are twice the cost of unlined ponds

(Lundquist et al., 2010), but discussions with UK excavation and liner suppliers suggested that the cost of a liner could be 2.7 times the cost of raceway pond excavation and over 5 times the cost if reinforced liners were required (various private communications with equipment manufacturers, 2008).

7.1.2 Paddlewheel power

The head loss and hydraulic power required for mixing was calculated at 0.05 m and 1.1 kW for a flow velocity of 0.3 m s^{-1} for a smooth lined raceway having a Manning Friction Factor of 0.01 (see Sections 2.1.1.3 & 4.2 for relevant equations). The power requirement for a 40 % efficient paddlewheel was calculated as 2.8 kW, but a paddlewheel manufacturer recommended a total installed power of 3.4 kW (Waterwheel Factory Inc. private communication, 2008).

7.1.3 Gaseous transfer power

Discussions were held with a number of blower manufactures to establish type, size, cost and power requirements for blowers to deliver flue gas to a gaseous sump in the raceway. The sizing of the blower was based on a flue gas concentration of 12 % v/v CO_2. The CO_2 concentration in the surrounding atmosphere should be the minimum possible for the health and safety of those in the area of the raceways and to comply with regulations concerning flue gas emissions at ground level (EC 80, 2001). In the current case a depth of 3m was selected for the gaseous transfer sump to ensure 90% removal of CO_2 from flue gas, based on the more conservative calculation of Putt (2007) and Stephenson et al. (2010) (Section 2.2.5).

A single-stage side channel blower was recommended by a number of UK suppliers for a total required pressure below 0.6 bar. If the pressure required is greater than 0.6 bar, a more expensive blower with higher energy consumption would be required. The static pressure in a 3 m deep sump would be 0.3 bar, allowing for a pressure drop in the remainder of the gas flow system of 0.3 bar. Using a shallower sump would mean that sufficient pressure was available for longer transfer distances and/or smaller bubbles. To meet algal growth needs, assuming a CO_2 requirement of 1.8 g g^{-1} of micro-algae, a yield of 25 g m^{-1} day^{-1} and a 90 % CO_2 transfer rate, the demand for flue gas at

12 % v/v CO_2 is 3410 m^3 ha^{-1} day^{-1}. It was assumed that CO_2 would only be supplied for an average of 12 hours per day during daylight hours. Based on a flow of up to 300 m^{-3} $hour^{-1}$, a single-side channel blower with an installed power of 5.5 kW was therefore selected (Northey private communications, 2009 and 2012).

7.1.4 Harvesting power

The wastewater treatment industry processes large quantities of water to remove organic material and, although treated water is the target product rather than organic matter, it was believed that existing technology in the wastewater treatment industry could offer a potentially viable harvesting system. Activated sludge treatment of wastewater and harvesting was considered to be analogously the closest process to the potential process of growth and harvesting of micro-algae for biofuel. The Thames Water activated sludge treatment plant at Longreach, Dartford, UK was visited and the harvesting of sludge observed. The harvesting process consisted of a settlement tank followed by a belt thickener with flocculant addition, and finally a belt filter press. Volatile solids concentrations in the sludge at various parts of the wastewater treatment were approximately 0.5 % from the settler, 5 % from the belt thickener and 25 % from the belt press (Thames Water and Ashbrook Simon-Hartley private communications, 2008 and 2012). Although the process of settlement, thickening and belt filtration does not appear to have been used for the large-scale commercial harvesting of micro-algae it has been successful in removing microorganisms from wastewater and was therefore selected as a part of the proposed design.

7.1.5 Process energy demand and EROOI

The installed motor power for the major items of equipment is given in Table 29, together with estimates, based on discussions with equipment manufacturers, of the percentage usage of this installed power for the growth and harvesting of micro-algae to produce a wet micro-algal sludge suitable for further processing into biodiesel.

Table 29 Energy requirements of growth and harvesting equipment

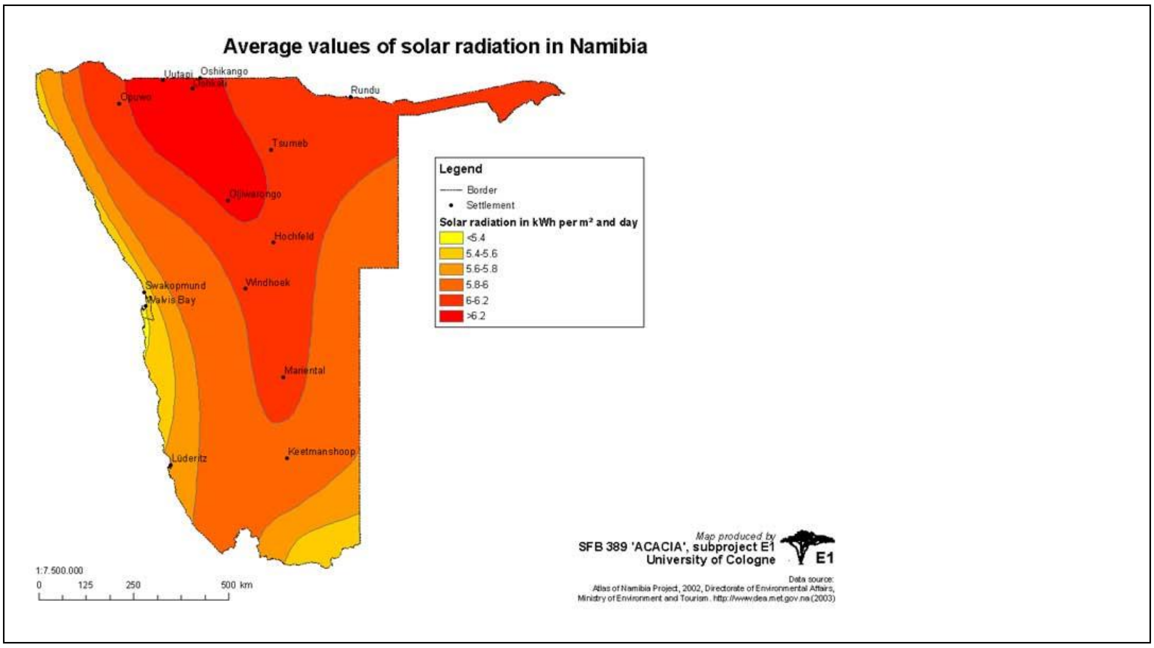
	Motor Rating	Raceway area covered	Power per unit area	Running time	Utilisation	Energy use
	kW	ha	kW ha^{-1}	hours	%	kW ha^{-1} day^{-1}
Raceway						
Paddlewheel	3.4	1	3.4	24	80%	65.3
Blower& sparger	5.5	1	5.5	12	80%	52.8
Total raceway			8.9			118.1
Settlement & harvesting						
Transfer pumps 2 per raceway @ 3kw	6	1	6.0	24	60%	86.4
Settlement tank	2	28	0.1	24	80%	1.4
Flocculation equipment & belt press	29	48	0.6	24	60%	8.7
Total harvesting			6.7			96.5
Total per hectare			**15.6**			**214.6**

The yield of micro-algae was assumed to be 250 kg ha^{-1} day^{-1} with a 20 % lipid content (Section 4.1.4.4), equivalent to a total calorific value yield of 1512 kWh day^{-1} in the total biomass, of which 547 kWh day^{-1} is in the lipid fraction. The operational EROOI for the growth and harvesting of wet total biomass is 7.0, and 2.5 for lipid only, but the biomass must be processed further before a usable fuel can be produced. The majority of the energy in the micro-algal biomass is not in the lipid fraction, but in the non-lipid residue, and the exploitation of the entire micro-algal biomass to produce biogas (Sections 2.4.9 & 6.13) could potentially give a significant improvement in the energy balance of a micro-algal biofuel system.

The operational energy required for the raceway cultivation system expressed as a percentage of the calorific value in the micro-algal biomass is 7.8 %, with a further 6.8 % for harvesting, fluid transfer and recycling. The energy used by the blower for gaseous exchange is 0.2 kWh kg^{-1} of dried micro-algal biomass, in good agreement with a previous model (Brune et al., 2009). Kadam (2002) estimated an energy usage of 22 kWh $tonne^{-1}$ of flue gas sparged into a micro-algal pond in a study of co-firing of micro-algae and coal, in reasonable agreement with this study's estimate of 19 kWh $tonne^{-1}$ of flue gas. The harvesting energy (excluding fluid transfer) of 0.13 kWh m^{-3} of dilute micro-algal suspension compares very favourably to that for centrifugation at 1.4 kWh m^{-3} (Milledge and Heaven, 2011), but is dependent on settlement and the use of flocculants, and the whole process needs to be demonstrated for micro-algae on a large scale. The energy inputs do not include any embodied energy for flocculant use.

7.2 Potential production sites

Selection of sites that have the appropriate climate together with a source of flue gas, wastewater and additional supplies of water is challenging. Namibia is a relatively politically stable country with a warm, sunny and dry climate, and was identified in conjunction with Pure Energy Fuels as possible location of sites for potential micro-algal biomass production. Maps of Namibia displaying average solar radiation and rainfall together with major towns and cities are shown in Figure 30.



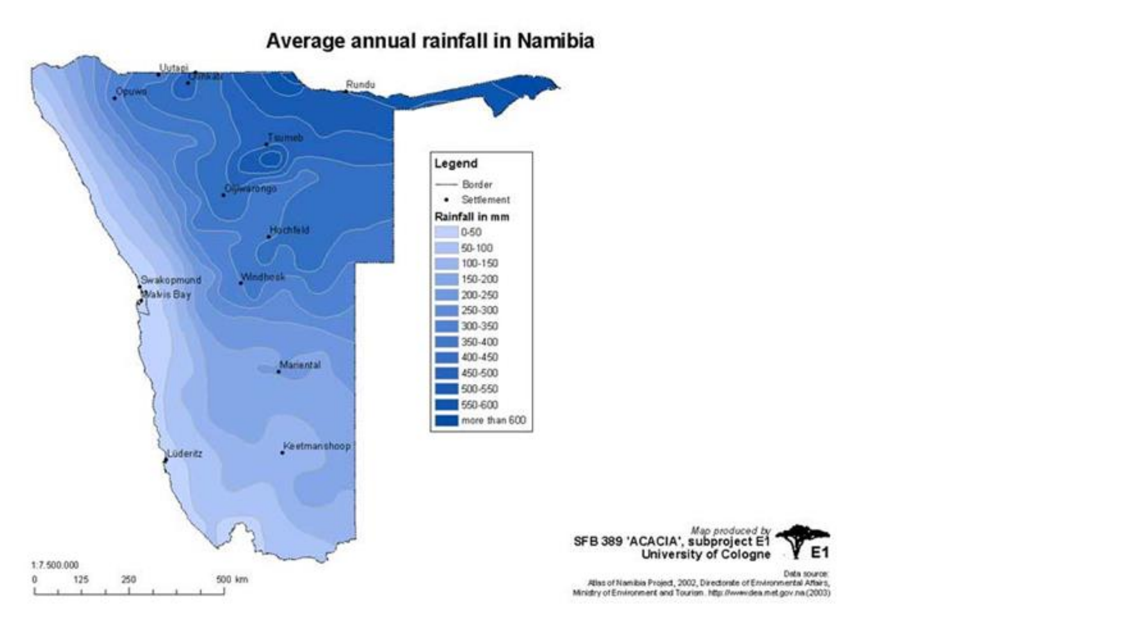


Figure 30 Average solar radiation and rainfall in Namibia (Ministry of Environment and Tourism Namibia, 2002)

7.2.1 **Van Eck power station, Windhoek**

Much of Namibia's electricity needs are met from power produced outside the country, but NamPower, the main electricity generating company within Namibia, has four major generating facilities: Ruacana hydro-electric on the Kunene River on the Angolan border in the north, producing up to 240 MW; Van Eck coal-powered station in northern Windhoek, producing up to 120 MW; Paratus diesel-powered standby station at Walvis Bay, producing up to 18 MW; and ANIXAS, opened in July 2011,situated adjacent to the Paratus Power Station at Walvis Bay, providing an emergency standby diesel-generated electrical capacity of 22.5 MW.

A capacity of 100,000 tonnes biomass year^{-1} was considered the preferred size for a commercial scale micro-algal biodiesel plant by Pure Energy Fuels, and the only plant capable of producing sufficient CO_2 was Van Eck in Windhoek (Figure 31). This station was constructed between 1972 and 1979 and operates on imported coal. Calculations suggested that at full output the plant could produce over 700,000 tonnes of CO_2 for 12 hours day^{-1} to supply algal growth during daylight. The estimated area required for micro-algal raceways and ancillary plant was 1400 ha and initial investigations suggested that sufficient land was available at the site. There was also a source of brackish water and a wastewater treatment plant in the area for the supply of water and nutrients.

Figure 31 Van Eck power station viewed from the North

The Van Eck power station's emission figures for CO_2 and NO_x production, showed an increase for both gases from 2003 to 2007 with output in 2007 being 181,992 and 919 tonnes respectively (Langenhoven, NamPower private communication, 2008). The information provided by NamPower on the rising emissions and production from Van Eck, together with the availability of a water source and excellent climatic conditions for micro-algal growth, led to the conclusion that a visit to NamPower in Windhoek was justified to further investigate the potential for micro-algal growth.

Discussions with NamPower in late 2008 indicated that the 'life' of Van Eck was well below the minimum 10-year span considered as the basis for any proposed investment by Pure Energy Fuels. An email from Rinus Castens of NamPower (Castens, NamPower private communication, 2008) stated, "You need a reliable supply of CO_2 which none of our existing power stations can offer. The new trend in NamPower according to our "Integrated Resource Plan" is to develop our Hydro potential." Investigation of biodiesel production from micro-algae at Van Eck power station therefore ceased at that time.

The Van Eck power station continues to operate, and with Namibia facing a power shortage NamPower has implemented several short-term measures including ‘ramping up’ the station (Duddy, 2012). NamPower has recently announced a potential project for a new coal fired power station in the Erongo district of Namibia (Duddy, 2012) and there have been announcements for the past ten years about a potential gas-powered station in the southwest of Namibia using gas from the Kudu gas field (Anon, 2013, Maletsky, 2004). Power stations at Erongo and Kudu could have reasonable access to sea water and large area of land, but neither has a large centre of population, and the supply of wastewater nutrients is thus likely to be lower than at Van Eck. The climatic conditions at both Erongo and Kudu are dry and sunny, and if power plants are established, they would be worthy of further investigation for micro-algal fuel production.

The proposed process of micro-algal biomass cultivation followed by solar drying then extraction of the lipid and trans-esterification for biodiesel production with residual biomass sold for fertiliser was subjected to financial analysis by Pure Energy Fuels, and found to be uneconomic. The focus of this part of the research therefore shifted to alternative approaches to improve the economic aspects of the scheme.

7.3 Alternative options

Although none of NamPower’s existing power stations currently offer a reliable supply of CO_2, the original reasons for selection of Namibia could, however, make it a potential area for commercialisation of micro-algae for non-fuel products or the production of biofuel as co-product in a biorefinery.

Fact finding studies were undertaken in November 2008 and April 2013 to the Namibian coastal towns of Walvis Bay and Swakopmund to assess their suitability for other forms of commercial-scale micro-algal production.

Discussions with national and local government organisations and other interested parties indicated that:

a. wastewater could be available to provide nutrients. Figure 32 shows the new activated sludge wastewater treatment plant at Swakopmund. The management at the Swakopmund wastewater plant were particularly interested in the development of micro-algal raceways integrated with the existing plant to remove nutrients and produce biomass for biogas production
b. sea water could be extracted for the growth of marine algae
c. there are large areas of level ground that could be suitable for raceways

Walvis Bay and Swakopmund were, therefore, both considered to be potential sites for the cultivation of *Dunaliella salina* for the production of β-carotene or as feedstock for a biorefinery, as this type of culture is suited to areas where land costs are low, there is supply of salt water and that have favourable climatic conditions with low rain fall and high solar insolation. Biomass residues from the production of β-carotene, such as glycerol and post-extraction biomass, could then be a potential source of biofuel.

Dunaliella is often found in open pan salt production ponds. Evidence of the presence of *Dunaliella* can be seen in orange colouration of water in the salt pans at Walvis Bay (Figure 33) and in the intense orange colour of the foam in the channel containing the water from the washing of salt crystals at Swakopmund (Figure 34), due to the release of β-carotene from fractured cells. Could the growth of *Dunaliella* for β-carotene be combined with natural evaporative salt production?

Figure 32 Aeration of wastewater at Swakopmund wastewater treatment plant

Figure 33 Open salt evaporation pans at Walvis Bay

Figure 34 Salt wash water channel at Swakopmund salt works

7.3.1 Techno-economic assessment of co-production of *Dunaliella* and salt

The costs of growing *Dunaliella* could be reduced by combining growth of the algae with natural evaporative salt production. An economic assessment using commercial equipment suppliers' data (2009) was carried out for production of 300 tonnes year^{-1} of spray-dried *Dunaliella salina* as a β-carotene supplement (based on discussions with equipment manufacturers and Pure Energy Fuels) produced from unmixed ponds, mixed raceways with gaseous transfer and co-production in unmixed evaporative salt ponds.

7.3.2 Basis of assessment

Productivities for *Dunaliella* grown in conjunction with salt production are unknown, and were therefore assumed to be the same as those found in commercial unmixed ponds.

The cost of a one hectare lined raceway with paddlewheel mixing was estimated at ~US$ 100,000, whilst a less productive unmixed and unlined pond could be excavated for US$ 25,000 (private communications with equipment manufacturers, 2008). *Dunaliella* is commonly harvested using disc-stack centrifuges. Although energy intensive these are effective in harvesting micro-algae (Section 2.3.5.1). A total yield of dried algae of 300 tonnes year^{-1} would require processing of 80 m^{-3} hour^{-1} of 0.05 % algal suspension, requiring two Westfalia-type separators costing US$ 929,529. These separators produce slurry that can be processed by a spray dryer with a budget cost of US$ 3,310,651 giving a total capital cost for harvesting and drying of US$ 4,240,180. In addition transfer pumps, process control, packing and handling equipment and buildings to house the plant will be required, giving an estimated total of US$ 5,250,000 of capital costs downstream of the growth ponds (private communications with equipment manufacturers, 2009). Energy costs for centrifugation and drying are high, at an annual estimated cost of US$ 400,000.

The assumed sale price was US$ 20 kg^{-1}, approximately one third of the price for wholesale product obtained from China (private communications with suppliers, 2009). Labour costs were taken as 12 % of capital expenditure excluding land costs, but no labour or capital costs were included for the ponds used in salt production as these are assumed to be covered by the original investment for the production of salt.

7.3.3 **Results and discussion**

Table 30 shows the estimated capital, income and expenditure for the three types of plant to produce 300 tonnes year^{-1} of spray-dried *Dunaliella salina* for sale as a β-carotene supplement. All three pond types show a profit and a return on investment, with the best return from co-production of *Dunaliella salina* with salt, if this is practicable. The results indicate that the growth of *Dunaliella salina* in the currently unexploited route of co-production in salt ponds is worthy of further investigation; however no practical studies appear to have been published on this to date. The estimates of productivity used need to be verified in a practical trial, together with tests on extraction and drying, before the viability of a co-production process could be fully established.

Table 30 Potential capital expenditure, income & profit from production of *Dunaliella salina*

	Mixed Raceway Ponds	Unmixed Ponds	Salt Ponds	Calculation notes
Pond costs construction & energy				
Target production, tonnes	300	300	300	
Yield, tonnes ha^{-1} $year^{-1}$	6	1.5	1.5	
Yield, g m^{-2} per day	1.6	0.4	0.4	
Total pond area, ha	50	200	200	
Pond construction cost, US$ ha^{-1}	100,000	25,000	0	
Total pond construction cost	5,000,000	5,000,000	0	
Power for mixing and aeration, kW	5.5	0	0	
Energy cost 300 t algal ponds, US$ $year^{-1}$	410,494	0	0	US$ 0.19 kWh
Harvesting & dryer costs				
Capital cost, US$	5,250,000	5,250,000	5,250,000	
Cost of energy for harvesting & drying, US$ $year^{-1}$	400,000	400,000	400,000	
Total capital cost excluding land	**10,250,000**	**10,250,000**	**5,250,000**	
Land Cost 300 t Algal Ponds, US$	500,000	2,000,000	0	US$10000 ha^{-1}
Land Cost Access etc., US$	100,000	100,000	0	10 ha
Land for plant, US$	20,000	20,000	20,000	2 ha
Total land cost, US$	**620,000**	**2,120,000**	**20,000**	
Total capital cost, US$	**10,870,000**	**12,370,000**	**5,270,000**	
Projected Income and Expenditure				
Target production, tonnes	300	300	300	
Sale price, US$ kg^{-1}	20	20	20	
Total income, US$	**6,000,000**	**6,000,000**	**6,000,000**	
Depreciation (excluding land), US$	1,025,000	1,025,000	525,000	10 years
Interest, US$	543,500	618,500	263,500	5% Total Cap
Energy, US$	810,494	400,000	400,000	
Labour, US$	1,230,000	1,230,000	630,000	12 % capital excluding land
Sundry (Maintenance etc.), US$	410,000	410,000	210,000	4 % Cap excluding land
Total expenditure, US$	**4,018,994**	**3,683,500**	**2,028,500**	
Net profit, US$	**1,981,006**	**2,316,500**	**3,971,500**	
% Annual Return on Investment including land	18%	19%	75%	

Preliminary work based on the concept put forward in this research is now being undertaken by Swakopmund Salt and the University of Greenwich, and if found to be viable a more detailed process design could be completed and a discounted cash flow analysis carried out.

The addition of mixing and gaseous transfer produced no improvement in profitability, and the negative effect would be even greater if lower land costs are used. More complex mixed open growth systems have been used for *Dunaliella* in both Israel and the USA where land and site preparation costs are high (Borowitzka, 1999), although the latter are no longer in operation. The lower productivity of simple unmixed systems has caused them to be largely supplanted by open systems with some form of circulation (Terry and Raymond, 1985); however the growth of *Dunaliella* is an exception.

This analysis has shown the economic viability of production of spray-dried *Dunaliella salina* as a β-carotene supplement. None of the proposed processes would appear viable for fuel production, however, as the potential value of *Dunaliella* for fuel is considerably less than for food supplements. A price for *Dunaliella salina* dry biomass for combustion of £ 0.21 kg^{-1} (~US$ 0.34 kg^{-1}) can be estimated from its calorific value of 18.47 kJ g^{-1} (5.1 kWh kg^{-1}) (Yang et al., 2011a), and the cost of dry wood pellet biomass fuel for combustion at £ 0.042 kWh (Biomass Energy Centre, 2012).This analysis is based on the sale of the entire biomass as food supplement, but purified β-carotene is currently worth considerably more at US$ 520 kg^{-1} (Harvey et al., 2012). With *Dunaliella* containing up to 14 % β-carotene, income from the refined product would increase to US$ 21,840,000, but the energy and solvent costs are potentially considerable. The additional refining energy cost could potentially be offset by the use of biomass for energy. *Dunaliella salina* can contain between 33 and 85 % glycerol when grown in high salinity environments (Ben-Amotz, 2013, Harvey et al., 2012) . Glycerol has a HHV ~18.0 MJ kg^{-1} (Weast, 1985) and a value of ~US$ 325 (Harvey et al., 2012), equivalent to US$ 0.065 kWh^{-1} (£ 0.042 kWh^{-1}). It can be burned in CHP units, and therefore could supply 1782 to 4590 GJ of energy as glycerol biomass or 32175 to US$ 82875 of additional income.

7.3.4 Horizontal biorefinery

Salt production could also support the growth of other micro-algae, as the seawater is concentrated by evaporation in a series of open pans and each step of increased salinity will naturally select for a different micro-algal strain. This variety of micro-algae could be capable of producing a wider range of useful chemicals and products than from the single feedstock of *Dunaliella.* This novel concept could be termed a 'horizontal biorefinery', and is shown schematically in Figure 35. The growth of a variety of micro-algae may also allow the commercial exploitation of salt pans in winter months.

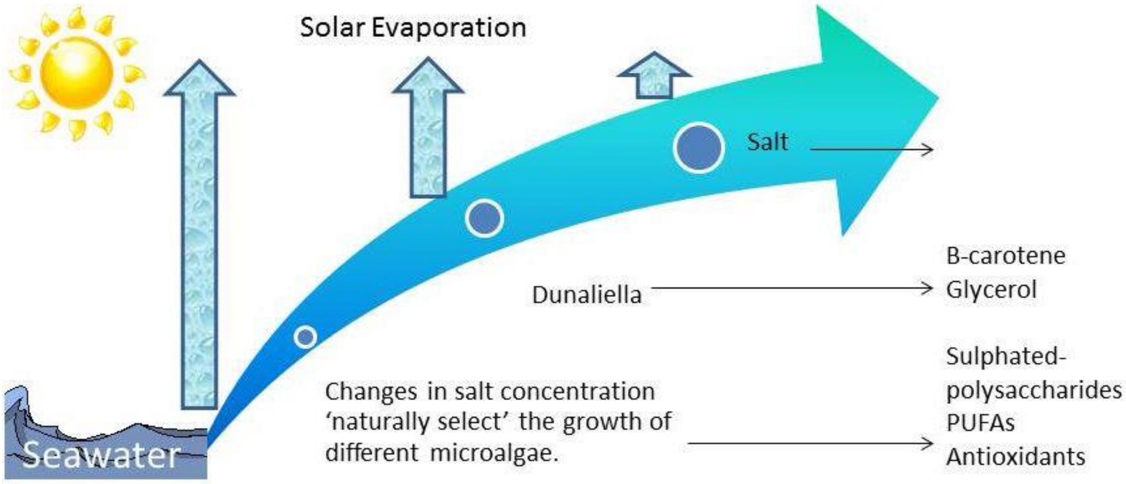


Figure 35 Horizontal Biorefinery

The use of any algal biomass residues for energy production via the anaerobic digestion would also contribute to meeting process energy requirements, in an integrated biorefinery concept. Salt is a known inhibitor of anaerobic digestion, however, and consideration will need to be given to the concentrations likely to occur at different points in the process.

7.4 Conclusions

The study of the proposed process for Pure Energy Fuels has indicated that the production of only biodiesel from micro-algae is not economically or energetically viable using current commercial technology. Biodiesel production from micro-algae may be more energetically favourable if new methods of oil extraction and trans-esterification are developed, but it appears essential that the full commercial and energetic value of the non-lipid micro-algae biomass is fully exploited. The economic and energetic balance of the system can be improved by the use of a process such as anaerobic digestion which is potentially capable of using the majority of the organic material in the biomass to produce biogas as a biofuel, and a comprehensive energy balance of the combined micro-algal biogas production process can be produced using the methods in Chapters 5 & 6.

The use of energy data from equipment manufacturers, although helpful for establishing a simple energy balance, requires the estimation of utilisation factors. Manufacturers are often unable to provide accurate values, and if process conditions such as flow rates change, further information on the effect of these variations needs to be obtained. The modelling approaches adopted in Sections 5 and 6 provide a more fundamental means of establishing the energy required for paddlewheel mixing and fluid and gas pumping.

Harvesting by settlement followed by a belt thickener, with addition of flocculant, and a belt filter press has a low operational energy requirement compared to other potential methods of harvesting such as centrifugation. This method is widely used in wastewater treatment, but does not appear to have been applied on a commercial scale to micro-algae. Practical studies are needed to confirm its applicability to micro-algae, but these were outside the scope of this research.

Namibia is climatically suited to the production of micro-algae, but there is currently a lack of a reliable low cost supply of CO_2 from flue gases for micro-algal fuel production. Namibia could, however, be a future site for growth of micro-algae non-fuel products and for biorefineries. The co-production of salt appears economically attractive and the novel idea of a horizontal biorefinery also appears to have some merit, but both need further research to establish if they are feasible and practicable. The production of halotrophic micro-algae, although potentially economically attractive for non-fuel products, does not currently appear to be economically or energetically viable for biomass or glycerol production solely for fuel. The energy recovered from glycerol or through AD of residual biomass, however could be used in the running of micro-algal processes to produce non-fuel products. The current research is focused on the production of energy from micro-algae, but the study of the commercialisation of micro-algae for non-fuel products in Namibia, in salt co-production, biorefineries and horizontal biorefineries could form the basis of further research projects.

8. Conclusions and further work

There are a wide range of combinations of growth, harvesting and energy extraction unit operations that can form a micro-algal biofuel production system, but as yet there is no successful economically viable system producing biofuel.

Currently the commercial exploitation of micro-algae is for non-fuel products, and there appears to be increasing research interest in the production of non-fuel products rather than fuel from micro-algae. In the short term, and possibly in the medium term, higher value products appear to be needed for economic exploitation of micro-algae. Biorefineries could allow the exploitation of the micro-algal biomass for a range of products, and produce fuel either for 'export' or for parasitic use to improve sustainability. The lessons learned from non-fuel products, together with their potential for co-production with fuel, may lead to the more rapid commercial realisation of micro-algal biofuel. Although high value algal products may allow the commercialisation of algae in the short term, the immense scale that is required to replace fossil fuels will result in the creation of such large quantities of algal non-fuel materials that the market price is likely to be dramatically reduced. Methods that allow exploitation of the entire organic biomass for energy production and that can produce energy profitably independent of co-product pricing are therefore needed.

A methodology has been developed that allows calculation of maximum theoretical dry algal biomass and oil yields, and this can be used to counter some of the extreme yield values suggested in the 'grey literature'. It can also produce realistic, practical or pragmatic yields, and can be adapted for a wide range of locations. There was no ready-made platform to rapidly produce an energy balance model for micro-algal biofuel. An Excel spreadsheet model was successfully developed which allowed the production of energy balances for a number of process options and scenarios.

The majority of the literature indicated that the open systems are the most energetically viable method of producing micro-algae for biofuel. This study has shown that open raceway system can be part of an energetically viable micro-algal biogas production process. It was also estimated that the build-up of oxygen in the raceway should remain below the level predicted to inhibit growth, and night time ‘crashes’, due de-oxygenation by micro-algal respiration, are unlikely to occur in raceways of typical depths.

Effective and energy efficient harvesting are vital for viable micro-algal biofuel. Disc-stack centrifuges, although suited to the separation of the particle sizes and concentrations found in micro-algal suspensions, have too high an energy consumption to be the sole means of harvesting micro-algae for the production of algal biofuel. Flocculation and/or sedimentation may not produce sufficiently high micro-algal concentrations. The combination of flocculation and/or sedimentation with centrifugation was shown to be energetically viable as part of micro-algal biogas production process, but a low dose and low embodied energy organic flocculant that is readily digested or micro-algal communities that settle readily is required.

The challenge for achieving energy-positive micro-algal biofuel is converting low energy density materials to energy-dense fuels; a process that requires energy input. The production of only biodiesel from micro-algae is not economically or energetically viable using current commercial technology. The energy balance assessment, however, has shown that the production of micro-algal biogas is energetically viable, but will be dependent on the exploitation of the heat generated by CHP.

Although this research has shown the production of micro-algal biogas being energetically viable, the production of biofuel from micro-algae is still in its embryonic stage, and thus considerable future research is required.

The specific contributions of this work are:

a. Development of a calculation method to estimate the maximum, realistic or achievable theoretical dry algal biomass and oil yields, which can be adapted for a wide range of locations by varying the value for solar insolation.
b. Appraisal and evaluation of current process integration software which showed that much of the data and many of the unit operations required

for micro-algal biofuel production are not available within the current commercially available software packages.

c. Development of a spreadsheet-based calculation tool to estimate the mixing energy requirement and biomass calorific yields for raceways, and the use of this to show that the required energy inputs relative to the biomass calorific value decrease with the addition of a liner, reduction in fluid velocity and increase in raceway size.
d. Development of a mechanistic energy balance model for the production of biogas from the anaerobic digestion of micro-algal biomass grown in raceways.
e. Demonstration that night-time crashes are unlikely to occur in raceway ponds due to oxygen depletion by micro-algal respiration.
f. Although CO_2 for micro-algal growth is provided by bacteria in facultative wastewater treatment ponds, the work showed additional CO_2 is required to utilise all the nitrogen in wastewater for micro-algal biomass production.
g. Demonstration that 1 hectare lined raceways appear to be possible and that raceways should be as wide as practicable. These results support the recommendation from the US Energy Department of a length to width ratio of 11 to 1.
h. Results from the model showing that increasing fluid velocity from 0.15 to 0.45 m s^{-1} at the same depth (0.3 m) reduces relative CO_2 outgassing, but increases energy input.
i. Results showing that there appears to no energy balance advantage to raceways being deeper than 0.3 m or flowing faster than 0.3 m s^{-1}.
j. A clear demonstration that it is not energetically viable to bubble air into a raceway as a source of CO_2.
k. Results from the model showing thermophilic digestion of micro-algal biomass is unlikely to be energetically viable,
l. A clear demonstration that flocculation by alum is not a viable option for the production of micro-algal biogas.
m. Results confirming that production of micro-algal biogas in the UK would be energetically challenging at best.
n. A clear demonstration that the use of a disc-stack centrifuge as the sole means of harvesting micro-algae for biodiesel and biogas production is too energy intensive to be energetically viable.

o. Proposal of the use of disc stack centrifugation for joint cell separation and disruption, and identification of required performance for process viability.
p. Quantitative evidence that the production of micro-algal biogas was energetically viable, but is dependent on the exploitation of the heat generated by the combustion of biogas in combined heat and power units to show a positive balance.
q. Proposal and evaluation of the co-production of *Dunaliella* and salt, and demonstration that it is potentially economically viable.
r. Proposal of a novel concept for a novel horizontal biorefinery.

Specific further research following on from this work:

a. Further development, refinement and extension of the methods reported in this dissertation to make them more user-friendly and capable of assessing more process combinations and variables.
b. Comparison of the output of the energy model with data from micro-algal pilot and demonstration plants, such as those of the FP7 All-Gas project, as they are developed. This will allow the accuracy of the model to be assessed and adjustments to be made.
c. The further examination of the co-production of micro-algae with salt.
d. The assessment of the feasibility of the novel concept of a horizontal biorefinery.

References

ACIÉN FERNÁNDEZ, F. G., FERNÁNDEZ SEVILLA, J. M. & MOLINA GRIMA, E. 2013. Photobioreactors for the Production of Microalgae. *Reviews in Environmental Science and Bio/Technology,* 12,(2), 131-151.

ADESANYA, V. O., VADILLO, D. C. & MACKLEY, M. R. 2012. The Rheological Characterization of Algae Suspensions for the Production of Biofuels. *Journal of Rheology,* 56,(4), 925-939.

AHRENS, T. & SANDER, H. 2010. Microalgae in Waste Water Treatment; Green Gold from Sludge? *Bioforum Europe,* 14,(3), 16-18.

ALBA, L. G., TORRI, C., SAMORI, C., VAN DER SPEK, J., FABBRI, D., KERSTEN, S. R. A. & BRILMAN, D. W. F. 2012. Hydrothermal Treatment (HIT) of Microalgae: Evaluation of the Process as Conversion Method in an Algae Biorefinery Concept. *Energy & Fuels,* 26,(1), 642-657.

AMER, L., ADHIKARI, B. & PELLEGRINO, J. 2011. Technoeconomic Analysis of Five Microalgae-to-Biofuels Processes of Varying Complexity. *Bioresource Technology,* 102,(20), 9350-9359.

ANANTHASWAMY, A. 2012. How Clean Is Green? *New Scientist,* 21,(2849), 35-38.

ANON. 2013. Namibia Seeks Bids for Delayed Kudu Power Plant. *The Namibian*, 15.04.

AQUAFUELS 2011. Algae and Aquatic Biomass for a Sustainable Production of 2nd Generation Biofuels. *Aquafuel FP7 – 241301-2.*

ARAD, S. & LEVY-ONTMAN, O. 2010. Red Microalgal Cell-Wall Polysaccharides: Biotechnological Aspects. *Current Opinion in Biotechnology,* 21,(3), 358-364.

ARAVANIS, A. 2012. Research & Development - Algae. *California Initiative for Large Molecule Sustainable Fuels - Roadmap Meeting.* San Diego.

ARENOVSKI, A. L. 1994. *The Distribution, Abundance and Ecology of Mixotrophic Algae in Marine and Freshwater Plankton Communities.* PhD, MIT.

ASPEN TECHNOLOGY INC. 2012. Bioprocess Innovator Relies on Process Modeling to Optimize Algae-to-Biofuel Conversion.

ATKINS, M. J., MORRISON, A. S. & WALMSLEY, M. R. W. 2010. Carbon Emissions Pinch Analysis (Cepa) for Emissions Reduction in the New Zealand Electricity Sector. *Applied Energy,* 87,(3), 982-987.

AXELSSON, H. 2000. Recent Trends in Disc Bowl Centrifuge Development. *Filtration & Separation,* 37,(4), 20-23.

AYLOTT, M. 2010. Forget Palm Oil and Soya, Microalgae Is the Next Big Biofuel Source. *The Ecologist.*

BABICH, I. V., VAN DER HULST, M., LEFFERTS, L., MOULIJN, J. A., O'CONNOR, P. & SESHAN, K. 2011. Catalytic Pyrolysis of Microalgae to High-Quality Liquid Bio-Fuels. *Biomass & Bioenergy,* 35,(7), 3199-3207.

BAILEY, J. E. & OLLIS, D. F. 1977. *Biochemical Engineering Fundamentals,* New York, McGraw Hill.

BALAT, M., BALAT, H. & OZ, C. 2008. Progress in Bioethanol Processing. *Progress in Energy and Combustion Science,* 34,(5), 551-573.

BANKS, C. 2011. Integration of Algal Production with Wastewater Treatment Plants: Advantages and Barriers. *Biodiesel production from microalgae: science, engineering and economic evaluation.* Almeria.

BANKS, C. 2012. Digester Mass and Energy Balances. *MSc Lecture part of CENV6141 Bioenergy.* University of Southampton.

BANKS, C., HEAVEN, S., LARA CORONA, E. S. & ROGALLA, F. 2011a. Energy Production from Integration of High Rate Anaerobic and Algal Wastewater Treatment: The EU FP7 All-Gas Project. *Water Stabilisation Ponds.* Adelaide: IWA.
BANKS, C. J., SALTER, A. M., HEAVEN, S. & RILEY, K. 2011b. Energetic and Environmental Benefits of Co-Digestion of Food Waste and Cattle Slurry: A Preliminary Assessment. *Resources Conservation and Recycling,* 56,(1), 71-79.
BASRAWI, F., YAMADA, T. & NAKANISHI, K. 2010. Effect of Ambient Temperature on the Energy Balance of Anaerobic Digestion Plants. *Journal of Environment and Engineering,* 5,(3), 526-538.
BATAN, L., QUINN, J., WILLSON, B. & BRADLEY, T. 2010. Net Energy and Greenhouse Gas Emission Evaluation of Biodiesel Derived from Microalgae. *Environmental Science & Technology,* 44,(20), 7975-7980.
BEAL, C., HEBNER, R., WEBBER, M., RUOFF, R. & SEIBERT, A. 2012a. The Energy Return on Investment for Algal Biocrude: Results for a Research Production Facility. *BioEnergy Research,* 5,(2), 341-362
BEAL, C. M. 2011. Algal Biofuels: Energy and Water. *WEG Symposium.* Austin Texas University of Texas
BEAL, C. M., HEBNER, R. E., WEBBER, M. E., RUOFF, R. S., SEIBERT, A. F. & KING, C. W. 2012b. Comprehensive Evaluation of Algal Biofuel Production: Experimental and Target Results. *Energies,* 5,(6), 1943-1981.
BECKER, E. W. 1994. *Microalgae. Biotechnology & Microbiology.,* Cambridge, Cambridge University Press.
BEER, T., GRANT, T. & CAMPBELL, P. K. 2007. The Greenhouse and Air Quality Emissions of Biodiesel Blends in Australia. CSIRO.
BELOSEVIC, S. 2010. Modeling Approaches to Predict Biomass Co-Firing with Pulverized Coal. *The Open Thermodynamics Journal,* 4, 50-70.
BEN-AMOTZ, A. 2008a. *Bio-Fuel and CO2 Capture by Algae* [Online]. NASA. Available: http://newbusiness.grc.nasa.gov/wp-content/uploads/2008/12/ben-amotz-nasa-nov-2008.pdf [Accessed 17/10/2009].
BEN-AMOTZ, A. 2008b. Large Scale Open Algal Ponds. *11th International Conference on Applied Phycology.* Galway Ireland: International Society for Applied Phycology ISAP.
BEN-AMOTZ, A. 2013. Marine Algal Systems. *Towards Establishing Value Chains for Bioenergy.* Swakopmund, Namibia: The African, Caribbean and Pacific Group of States (ACP) Science and Technology Programme.
BENEMANN, J., KOOPMAN, B., WEISSMAN, J., EISENBERG, D. & GOEBEL, R. 1980. Development of Microalgae Harvesting and High-Rate Pond Technologies in California. *In:* SHELEF, G. & SOEDER, C. J. (eds.) *Algae Biomass.* Amsterdam: Elsevier.
BENEMANN, J. & OSWALD, W. J. 1996. Systems and Economic Analysis of Microalgae Ponds for Conversion of CO2 to Biomass. Pittsburgh: Pittsburgh Energy Technology Centre.
BENEMANN, J., WEISSMAN, J. C., KOOPMAN, B. L. & OSWALD, W. J. 1977. Energy-Production by Microbial Photosynthesis. *Nature,* 268,(5615), 19-23.
BENEMANN, J. R. 2000. Hydrogen Production by Microalgae. *Journal of Applied Phycology,* 12,(3), 291-300.
BENEMANN, J. R. 2008. *Opportunities & Challenges in Algae Biofuels Production* [Online]. FAO. Available: http://www.fao.org/uploads/media/algae_positionpaper.pdf [Accessed 10/12/2009].
BENNETT, O. 2011. Biofuels. London: House Of Commons Library.

BERG-NILSEN, J. 2006. Production of Micro-Algae Based Products. Oslo: Nordic Innovation Centre.
BERGLUND, M. & BORJESSON, P. 2006. Assessment of Energy Performance in the Life-Cycle of Biogas Production. *Biomass & Bioenergy,* 30**,**(3), 254-266.
BHOLA, V., DESIKAN, R., SANTOSH, S. K., SUBBURAMU, K., SANNIYASI, E. & BUX, F. 2011. Effects of Parameters Affecting Biomass Yield and Thermal Behaviour of Chlorella Vulgaris. *Journal of Bioscience and Bioengineering,* 111**,**(3), 377-382.
BIENFANG, P. K. 1981. Sinking Rates of Heterogeneous Temperate Phytoplankton Populations. *Journal of Plankton Research,* 3**,**(2), 235-254.
BIOMASS ENERGY CENTRE. 2012. *Fuel Cost Per Kwhr* [Online]. Available: http://www.biomassenergycentre.org.uk/portal/page?_pageid=75,59188&_dad=portal [Accessed 05/03/2012].
BOISSEVAIN, B. 2012. *Waste Heat Utilization in an Anaerobic Digestion System.* MSc, Utah State University.
BOLHOUSE, A. M. 2010. *Rheology of Algae Slurries.* MSc, University of Texas.
BOROWITZKA, M. A. 1995. Microalgae as Sources of Pharmaceuticals and Other Biologically Active Compounds. *Journal of Applied Phycology,* 7, 3-15.
BOROWITZKA, M. A. 1999. Commercial Production of Microalgae: Ponds, Tanks, Tubes and Fermenters. *Journal of Biotechnology,* 70**,**(1-3), 313-321.
BOROWITZKA, M. A. 2005. Culturing of Microalgae in Outdoor Ponds. *In:* ANDERSEN, R. A. (ed.) *Algal Culturing Techniques.* London: Elsevier.
BOROWITZKA, M. A. 2006. *Biotechnological & Environmental Applications of Microalgae* [Online]. Murdoch University. Available: http://www.bsb.murdoch.edu.au/groups/beam/BEAM-Appl0.html [Accessed 26 11 2008].
BOSMA, R., VAN SPRONSEN, W. A., TRAMPER, J. & WIJFFELS, R. H. 2003. Ultrasound, a New Separation Technique to Harvest Microalgae. *Journal of Applied Phycology,* 15**,**(2-3), 143-153.
BOSMA, R., VAN ZESSEN, E., REITH, J. H., TRAMPER, J. & WIJFFELS, R. H. 2007. Prediction of Volumetric Productivity of an Outdoor Photobioreactor. *Biotechnology and Bioengineering,* 97**,**(5), 1108-1120.
BOYCHYN, M., YIM, S. S. S., BULMER, M., MORE, J., BRACEWELL, D. G. & HOARE, M. 2004. Performance Prediction of Industrial Centrifuges Using Scale-Down Models. *Bioprocess and Biosystems Engineering,* 26**,**(6), 385-391.
BP. 2011. *Bp Statistical Review of World Energy June 2011* [Online]. London: BP. Available: http://www.bp.com/statisticalreview [Accessed 04/07/2011].
BRENNAN, J. G., BUTTERS, J. R., COWELL, N. D. & LILLY, A. E. V. 1969. *Food Engineering Operation,* London, Elsevier.
BRENNAN, L. & OWENDE, P. 2010. Biofuels from Microalgae--a Review of Technologies for Production, Processing, and Extractions of Biofuels and Co-Products. *Renewable and Sustainable Energy Reviews,* 14**,**(2), 557-577.
BRENTNER, L. B., ECKELMAN, M. J. & ZIMMERMAN, J. B. 2011. Combinatorial Life Cycle Assessment to Inform Process Design of Industrial Production of Algal Biodiesel. *Environmental Science & Technology,* 45**,**(16), 7060-7067.
BROERE, W. 2008. *Harvesting Energy from Algae* [Online]. Shell. Available: http://www.shell.com/home/content/innovation/about_us/news_publications/shell_world_stories/2008/algae/ [Accessed 20/05/2009].

BROWN, M. R. & JEFFREY, S. W. 1995. The Amino Acid and Gross Composition of Marine Diatoms Potentially Useful for Mariculture. *Journal of Applied Phycology,* 7,(6), 521-527.
BROWN, P. 2009. *Algal Biofuels Research, Development, and Commercialization Priorities: A Commercial Economics Perspective* [Online]. ep Overviews Publishing, Inc. Available: http://www.epoverviews.com/oca/Algae%20Biofuel%20Development%20Priorities%20.pdf [Accessed 11/12/2008].
BROWN, T. M., DUAN, P. & SAVAGE, P. E. 2010. Hydrothermal Liquefaction and Gasification of Nannochloropsis Sp. *Energy & Fuels,* 24,(6), 3639-3646.
BRUNE, D. E., LUNDQUIST, T. J. & BENEMANN, J. R. 2009. Microalgal Biomass for Greenhouse Gas Reductions: Potential for Replacement of Fossil Fuels and Animal Feeds. *Journal of Environmental Engineering-Asce,* 135,(11), 1136-1144.
BRUTON, T., LYONS, H., LERAT , Y., STANLEY, M. & RASMUSSEN, M. B. 2009. A Review of the Potential of Marine Algae as a Source of Biofuel in Ireland. Dublin: Sustainable Energy Ireland.
BSI 2005. Natural Gas - Calculation of Calorific Values, Density, Relative Density & Wobbe Index from Composition. *BS EN ISO 6976:2005.*
BUHR, H. O. & MILLER, S. B. 1983. A Dynamic Model of the High Rate Algal Bacterial Waste Water Treatment Pond. *Water Research,* 17,(1), 29-38.
BUSWELL, A. M. & MUELLER, H. F. 1952. Mechanism of Methane Fermentation. *Industrial & Engineering Chemistry,* 44,(3), 550-552.
CAMACHO RUBIO, F., ACIEN FERNANDEZ, F. G., SANCHEZ PEREZ, J. A., GARCIA CAMACHO, F. & MOLINA GRIMA, E. 1999. Prediction of Dissolved Oxygen and Carbon Dioxide Concentration Profiles in Tubular Photobioreactors for Microalgal Culture. *Biotechnology and Bioengineering,* 62,(1), 71-86.
CAMPBELL, P. K., BEER, T. & BATTEN, D. 2009. Greenhouse Gas Sequestration by Algae- Energy & Greenhouse Gas Life Cycle Studies. *In:* CSIRO (ed.) *6th Australian Conference on Life Cycle Assessment.* Melbourne: CSIRO.
CANNELL, M. G. R. & HOOPER, M. D. (eds.) 1990. *The Greenhouse Effect and Terrestrial Ecosystems of the UK,* London: HMSO.
CARLSON, L. S., LEE, M. Y., OJE, C. A. E. & XU, A. 2010. Algae to Alkanes. *Senior Design Reports.* University of Pennsylvania.
CHANSON, H. 1999. *Hydraulics of Open Channel Flow,* London, Arnold.
CHEN, L. A., SERAD, G. A. & CARBONELL, R. G. 1998. Effect of Mixing Conditions on Flocculation Kinetics of Wastewaters Containing Proteins and Other Biological Compounds Using Fibrous Materials and Polyelectrolytes. *Brazilian Journal of Chemical Engineering,* 15, 358-368.
CHEN, W. H., HAN, S. K. & SUNG, S. 2003. Sodium Inhibition of Thermophilic Methanogens. *Journal of Environmental Engineering-Asce,* 129,(6), 506-512.
CHEN, Y., CHENG, J. J. & CREAMER, K. S. 2008. Inhibition of Anaerobic Digestion Process: A Review. *Bioresource Technology,* 99,(10), 4044-4064.
CHENG, N. S. 2007. Power-Law Index for Velocity Profiles in Open Channel Flows. *Advances in Water Resources,* 30,(8), 1775-1784.
CHERUBINI, F. 2010. The Biorefinery Concept: Using Biomass Instead of Oil for Producing Energy and Chemicals. *Energy Conversion and Management,* 51,(7), 1412-1421.
CHIARAMONTI, D., PRUSSI, M., CASINI, D., TREDICI, M. R., RODOLFI, L., BASSI, N., ZITTELLI, G. C. & BONDIOLI, P. 2013. Review of Energy Balance in

Raceway Ponds for Microalgae Cultivation: Re-Thinking a Traditional System Is Possible. *Applied Energy,* 102,(0), 101-111.
CHIN, D. A. 2006. *Water-Quality Engineering in Natural Systems,* Hoboken, Wiley.
CHISTI, Y. 2000. Marine Biotechnology - a Neglected Resource. Editorial. *Biotechnology Advances,* 18, 547-548.
CHISTI, Y. 2001. Hydrodynamic Damage to Animal Cells. *Critical Reviews in Biotechnology,* 21,(2), 67-110.
CHISTI, Y. 2006. Microalgae as Sustainable Cell Factories. *Enviromental Engineering Management Journal,* 5,(3), 261-274.
CHISTI, Y. 2007. Biodiesel from Microalgae. *Biotechnology Advances,* 25, 294 - 306.
CHISTI, Y. 2010. Fuels from Microalgae. *Biofuels,* 1,(2), 233-235.
CHISTI, Y. & MOO YOUNG, M. 1986. Disruption of Microbial-Cells for Intracellular Products. *Enzyme and Microbial Technology,* 8,(4), 194-204.
CHLUP, P. H., BERNARD, D. & STEWART, G. G. 2008. Disc Stack Centrifuge Operating Parameters and Their Impact on Yeast Physiology. *Journal of the Institute of Brewing,* 114,(1), 45-61.
CHOI, S. K., LEE, J. Y., KWON, D. Y. & CHO, K. J. 2006. Settling Characteristics of Problem Algae in the Water Treatment Process. *Water Science and Technology,* 53,(7), 113-119.
CHOI, S. P., NGUYEN, M. T. & SIM, S. J. 2010. Enzymatic Pretreatment of Chlamydomonas Reinhardtii Biomass for Ethanol Production. *Bioresource Technology,* 101,(14), 5330-5336.
CHOW, V. T. 1959. *Open-Channel Hydraulics,* New York, McGraw-Hill.
CHOWDHURY, R., VIAMAJALA, S. & GERLACH, R. 2012. Reduction of Environmental and Energy Footprint of Microalgal Biodiesel Production through Material and Energy Integration. *Bioresource Technology,* 108, 102-111.
CHRISTENSON, L. & SIMS, R. 2011. Production and Harvesting of Microalgae for Wastewater Treatment, Biofuels, and Bioproducts. *Biotechnology Advances,* 29,(6), 686-702.
CHRISTODOULIDES, J. S. 2001. *Mixing in Anaerobic Digesters.* PhD, University of Southampton.
CHUNG, T. S., PATINO-ECHEVERRI, D. & JOHNSON, T. L. 2011. Expert Assessments of Retrofitting Coal-Fired Power Plants with Carbon Dioxide Capture Technologies. *Energy Policy,* 39,(9), 5609-5620.
CLARENS, A. F., NASSAU, H., RESURRECCION, E. P., WHITE, M. A. & COLOSI, L. M. 2011a. Environmental Impacts of Algae-Derived Biodiesel and Bioelectricity for Transportation. *Environmental Science & Technology*, null-null.
CLARENS, A. F., RESURRECCION, E. P., WHITE, M. A. & COLOSI, L. M. 2011b. Response to Comment on "Environmental Life Cycle Comparison of Algae to Other Bioenergy Feedstocks". *Environmental Science & Technology,* 45,(2), 834-834.
CLARKE, A., PRESCOTT, T., KHAN, A. & OLABI, A. G. 2010. Causes of Breakage and Disruption in a Homogeniser. *Applied Energy,* 87,(12), 3680-3690.
COLE, J. J. & CARACO, N. F. 1998. Atmospheric Exchange of Carbon Dioxide in a Low-Wind Oligotrophic Lake Measured by the Addition of Sf6. *Limnology and Oceanography,* 43,(4), 647-656.
COLE, T. M. & WELLS, S. A. 1995. Ce-Qual-W2. A Two Dimensional, Laterally Averaged, Hydrodynamic & Water Quality Model, Version 3.5: User Manual. Vicksburg MS: US Army Corps Engineers.

COLLET, P., HÉLIAS, A., LARDON, L., RAS, M., GOY, R.-A. & STEYER, J.-P. 2011. Life-Cycle Assessment of Microalgae Culture Coupled to Biogas Production. *Bioresource Technology,* 102,(1), 207-214.
COMMITTEE ON CLIMATE CHANGE 2011. The Renewable Energy Review.
COULSON, J. M. & RICHARDSON, J. F. 1999. *Coulson & Richardson's Chemical Engineering . Fluid Flow, Heat Transfer and Mass Transfer.,* Oxford, Elsevier.
COUPER, J. R., PENNEY, W. R., FAIR, J. R. & WALAS, S. M. 2005. *Chemical Process Equipment :Selection and Design,* Oxford, Elsevier.
CRAGGS, R. 2003. Advanced Integrated Wastewater Ponds. *In:* SHILTON, A. (ed.) *Pond Treatment Technology.* London: IWA.
DAINTITH, J. & MARTIN, E. (eds.) 2010. *A Dictionary of Science* Oxford Oxford University Press.
DANQUAH, M. K., ANG, L., UDUMAN, N., MOHEIMANI, N. & FORDEA, G. M. 2009. Dewatering of Microalgal Culture for Biodiesel Production: Exploring Polymer Flocculation and Tangential Flow Filtration. *Journal of Chemical Technology and Biotechnology,* 84,(7), 1078-1083.
DAVIDSON, P. A. 2004. *Turbulence. An Introduction for Scientists and Engineers,* Oxford, Oxford University Press.
DAVIS, R., ADEN, A. & PIENKOS, P. T. 2011. Techno-Economic Analysis of Autotrophic Microalgae for Fuel Production. *Applied Energy,* 88,(10), 3524-3531.
DE BOER, K., MOHEIMANI, N., BOROWITZKA, M. & BAHRI, P. 2012. Extraction and Conversion Pathways for Microalgae to Biodiesel: A Review Focused on Energy Consumption. *Journal of Applied Phycology,* 24,(6), 1681-1698
DE SCHAMPHELAIRE, L. & VERSTRAETE, W. 2009. Revival of the Biological Sunlight-to-Biogas Energy Conversion System. *Biotechnology and Bioengineering,* 103,(2), 296-304.
DE WIT, J. N. 2001. Lecturer's Handbook on Whey and Whey Products. Brussels: European Whey Products Association.
DECC 2010. Third Overview of the Work of the UK Coal Forum: August 2008 to December 2009.
DECC 2011. Digest of UK Energy Statistics (Dukes). London.
DEFRA 2010. Guidelines to Defra / Decc's Ghg Conversion Factors for Company Reporting. LONDON: Produced by AEA for the Department of Energy and Climate Change (DECC)and the Department for Environment, Food and Rural Affairs (Defra).
DELRUE, F., SEITER, P. A., SAHUT, C., COURNAC, L., ROUBAUD, A., PELTIER, G. & FROMENT, A. K. 2012. An Economic, Sustainability, and Energetic Model of Biodiesel Production from Microalgae. *Bioresource Technology,* 111, 191-200.
DEMIRBAS, A. 2001. Biomass Resource Facilities and Biomass Conversion Processing for Fuels and Chemicals. *Energy Conversion and Management,* 42,(11), 1357-1378.
DEMIRBAS, A. 2010. Hydrogen from Mosses and Algae Via Pyrolysis and Steam Gasification. *Energy Sources Part a-Recovery Utilization and Environmental Effects,* 32,(2), 172-179.
DIAS, M. O. S., ENSINAS, A. V., NEBRA, S. A., MACIEL, R., ROSSELL, C. E. V. & MACIEL, M. R. W. 2009. Production of Bioethanol and Other Bio-Based Materials from Sugarcane Bagasse: Integration to Conventional Bioethanol Production Process. *Chemical Engineering Research & Design,* 87,(9A), 1206-1216.

DIC. 2008. *Natural Blue Colorant Derived from Dic Spirulina* [Online]. DIC Corporation Available: http://www.dlt-spl.co.jp/business/en/spirulina/linablue.html [Accessed].
DOUCHA, J. & LÍVANSKÝ, K. 2009. Outdoor Open Thin-Layer Microalgal Photobioreactor: Potential Productivity. *Journal of Applied Phycology,* 21,(1), 111-117.
DOULAH, M. S. 1977. Mechanism of Disintegration of Biological Cells in Ultrasonic Cavitation. *Biotechnology and Bioengineering,* 19,(5), 649-660.
DOUSKOVA, L., DOUCHA, J., LIVANSKY, K., MACHAT, J., NOVAK, P., MYSOVA, D., ZACHLEDR, V. & VITOVA, M. 2009. Simultaneous Flue Gas Biomerediation and Reduction of Microalgal Biomass Production Costs. *Appied Microbiology Biotechnology,* 82, 179-185.
DOWNEY, K. 2012. Research & Development - Algae. *California Initiative for Large Molecule Sustainable Fuels - Roadmap Meeting.* San Diego.
DU, Z. Y., LI, Y. C., WANG, X. Q., WAN, Y. Q., CHEN, Q., WANG, C. G., LIN, X. Y., LIU, Y. H., CHEN, P. & RUAN, R. 2011. Microwave-Assisted Pyrolysis of Microalgae for Biofuel Production. *Bioresource Technology,* 102,(7), 4890-4896.
DUDDY, J.-M. 2012. Nampower Gets Clean Bill of Credit Health. *The Namibian*, 16/04/2012.
DUNLOP, E. 2012. *Aspen Tech Webinar* [Online]. Available: http://www.panpacific.com.au/aspen.html [Accessed 16/05/2013].
EC 80 2001. Limitation of Emissions of Certain Pollutants into the Air from Large Combustion Plants. Official Journal of the European Communities.
ECOFYS 2008. Worldwide Potential of Aquatic Biomass. Utrecht: Ecofys.
ECOFYS 2011. International Biodiesel Markets.Developments in Production and Trade.
EDZWALD, J. K. 1993. Algae, Bubbles, Coagulants, and Dissolved Air Flotation. *Water Science and Technology,* 27,(10), 67-81.
EL-DESSOUKY, H. T. & ETTOUNEY, H. M. 2002. *Fundamentals of Salt Water Desalination* Elsevier.
ELLIOTT, A. 2006. Substitution of Ferric Chloride with Magnafloc 1597 in Autothermal Thermophyllic Aerated Digestion (Atad)Sludge De-Watering Processes. *69th Annual Water Industry Engineers and Operators' Conference.* Bendingo, Australia.
ENERGY SYSTEMS RESEARCH UNIT. 1998. *Case Study; Investigated Routes of Disposal* [Online]. University of Strathclyde Available: http://www.esru.strath.ac.uk/EandE/Web_sites/97-8/energy_from_waste/page6.htm [Accessed 10/12 2012].
ENVIROTECH. 2004. *Greenfuel Breathes Easier with Good Results from Mit Emissions Reduction Test* [Online]. Boston: Mass High Tech. [Accessed 2008].
ESPINOSA-SOLARES, T., BOMBARDIERE, J., CHATFIELD, M., DOMASCHKO, M., EASTER, M., STAFFORD, D. A., CASTILLO-ANGELES, S. & CASTELLANOS-HERNANDEZ, N. 2006. Macroscopic Mass and Energy Balance of a Pilot Plant Anaerobic Bioreactor Operated under Thermophilic Conditions. *Applied biochemistry and biotechnology*, 129-132, 959-68.
EUROPEAN COMMISSION. 2011. *Agriculture and Bioenergy* [Online]. Available: http://ec.europa.eu/agriculture/bioenergy/index_en.htm [Accessed 13/01/2012].
FAO. 1997. Fao. *FAO Agricultural Services Bulletin - 128 (Chapter 1 - Biological energy production)* [Online]. Available: http://www.fao.org/docrep/w7241e/w7241e05.htm.

FEC SERVICES 2003. Anaerobic Digestion, Storage, Oligolysis, Lime, Heat and Aerobic Treatment of Livestock Manures.
FERGUSON, J. 1952. The Rate of Natural Evaporation from Shallow Ponds. *Australian Journal of Scientific Research, Series A: Physical Sciences,* 5,(315-330).
FERNANDEZ, F. G. A., SEVILLA, J. M. F., PEREZ, J. A. S., GRIMA, E. M. & CHISTI, Y. 2001. Airlift-Driven External-Loop Tubular Photobioreactors for Outdoor Production of Microalgae: Assessment of Design and Performance. *Chemical Engineering Science,* 56,(8), 2721-2732.
FERRELL, J. & SARISKY-REED, V. 2010. National Algal Biofuels Technology Roadmap. A Technology Roadmap Resulting from the National Algal Biofuels Workshop Washington U.S. Department of Energy, Office of Energy Efficiency and Renewable Energy, Biomass Program. available online @ http://www1.eere.energy.gov/biomass/pdfs/algal_biofuels_roadmap.pdf.
FORSTER, C. F. (ed.) 2003. *Wastewater Treatment and Technology,* London: Thomas Telford
FRANCESCHIN, G., ZAMBONI, A., BEZZO, F. & BERTUCCO, A. 2008. Ethanol from Corn: A Technical and Economical Assessment Based on Different Scenarios. *Chemical Engineering Research & Design,* 86,(5A), 488-498.
FRANK, E. D., PALOU-RIVERA, I., ELGOWAINY, A. & WANG, M. 2011. Introduction to Algal Fuel LCA in Greet 1_2011. *GREET Workshop.* Argonne, IL.
GARCIA-GONZALEZ, M., MORENO, J., CANAVATE, J. P., ANGIUS, V., PRIETO, A., MANZANO, C., FLORENCIO, F. J. & GUERRERRO 2003. Conditions for Open-Air Outdoor Culture of Dunaliella Salina in Southern Spain. *Journal of Applied Phycology,* 15,(2/3), 177-184.
GARCIA CAMACHO, F., GALLARDO RODRIGUEZ, J. J., SANCHEZ MIRON, A., BELARBI, E. H., CHISTI, Y. & MOLINA GRIMA, E. 2011. Photobioreactor Scale-up for a Shear-Sensitive Dinoflagellate Microalga. *Process Biochemistry,* 46,(4), 936-944.
GAVALA, H. N., YENAL, U., SKIADAS, I. V., WESTERMANN, P. & AHRING, B. K. 2003. Mesophilic and Thermophilic Anaerobic Digestion of Primary and Secondary Sludge. Effect of Pre-Treatment at Elevated Temperature. *Water Research,* 37,(19), 4561-4572.
GAVRILESCU, M. & CHISTI, Y. 2005. Biotechnology - a Sustainable Alternative for Chemical Industry. *Biotechnology Advances,* 23, 471-499.
GEA PROCESS ENGINEERING. 2011. *Algal Cell Disruption* [Online]. Available: http://www.niroinc.com/gea_liquid_processing/algae_cells_disruption.asp [Accessed 20/09/2011].
GEUN GOO, B., BAEK, G., JIN CHOI, D., IL PARK, Y., SYNYTSYA, A., BLEHA, R., HO SEONG, D., LEE, C.-G. & KWEON PARK, J. 2013. Characterization of a Renewable Extracellular Polysaccharide from Defatted Microalgae Dunaliella Tertiolecta. *Bioresource technology,* 129, 343-50.
GHASEMI, Y., RASOUL-AMINI, S., NASERI, A. T., MONTAZERI-NAJAFABADY, N., MOBASHER, M. A. & DABBAGH, F. 2012. Microalgae Biofuel Potentials (Review). *Applied Biochemistry and Microbiology,* 48,(2), 126-144.
GILBERT, W. G. 1987. Anaerobic Digester Mixing Systems. *Journal Water Pollution Control Federation,* 59,(3), 162-170.
GOH, A. Year. Production of Microalgae Using Pig Waste as a Substrate. *In:* BARCLAY, W. R. & MCINTOSH, R. P., eds. Algal Biomass Technologies, 1984 Colorado. J Cramer.
GOLDMAN, J. C. 1979a. Outdoor Algal Mass Cultures--I. Applications. *Water Research,* 13,(1), 1-19.

GOLDMAN, J. C. 1979b. Outdoor Algal Mass Cultures--Ii. Photosynthetic Yield Limitations. *Water Research,* 13,(2), 119-136.
GOLUEKE, C. G. & OSWALD, W. J. 1959. Biological Conversion of Light Energy to the Chemical Energy of Methane. *Appl Microbiol,* 7,((4)), 219-227.
GOLUEKE, C. G., OSWALD, W. J. & GOTAAS, H. B. 1957. Anaerobic Digestion of Algae. *Appl Microbiol,* 5,((1)), 47-55.
GONZALEZ-DELGADO, A. D. & KAFAROV, V. 2011. Microalgae Based Biorefinery: Issues to Consider. *Ct&F-Ciencia Tecnologia Y Futuro,* 4,(4), 5-21.
GONZALEZ-FERNANDEZ, C., SIALVE, B., BERNET, N. & STEYER, J. P. 2012a. Impact of Microalgae Characteristics on Their Conversion to Biofuel. Part Ii: Focus on Biomethane Production. *Biofuels Bioproducts & Biorefining-Biofpr,* 6,(2), 205-218.
GONZALEZ-FERNANDEZ, C., SIALVE, B., BERNET, N. & STEYER, J. P. 2012b. Thermal Pretreatment to Improve Methane Production of Scenedesmus Biomass. *Biomass & Bioenergy,* 40, 105-111.
GOSLING, I. 2005. Process Simulation and Modeling for Industrial Bioprocessing: Tools and Techniques. *Industrial Biotechnology,* 1,(2), 106-109.
GOUVEIA, L. 2011. *Microalgae as a Feedstock for Biofuels,* Heidelberg, Springer.
GOUVEIA, L., MARQUES, A. E., SOUSA, J. M., MOURA, P. & BANDARRA, N. M. 2010. Microalgae – Source of Natural Bioactive Molecules as Functional Ingredients. *Food Science & Technology Bulletin: Functional Foods,* 7,(2), 21-37.
GRANADOS, M. R., ACIÉN, F. G., GÓMEZ, C., FERNÁNDEZ-SEVILLA, J. M. & MOLINA GRIMA, E. 2012. Evaluation of Flocculants for the Recovery of Freshwater Microalgae. *Bioresource Technology,* 118, 102-110
GREEN, F. B., LUNDQUIST, T. J. & OSWALD, W. J. 1995. Energetics of Advanced Integrated Waste-Water Pond Systems. *Water Science and Technology,* 31,(12), 9-20.
GREEN, M. A., EMERY, K., HISHIKAWA, Y. & WARTA, W. 2011. Solar Cell Efficiency Tables (Version 37). *Progress in Photovoltaics,* 19,(1), 84-92.
GREENWELL, H. C., LAURENS, L. M. L., SHIELDS, R. J., LOVITT, R. W. & FLYNN, K. J. 2010. Placing Microalgae on the Biofuels Priority List: A Review of the Technological Challenges. *Journal of the Royal Society Interface,* 7,(46), 703-726.
GRESSEL, J. 2008. Transgenics Are Imperative for Biofuel Crops. *Plant Science,* 174,(3), 246-263.
GROBBELAAR, J. 1994. Turbulence in Mass Algal Cultures and the Role of Light/Dark Fluctuations. *Journal of Applied Phycology,* 6,(3), 331-335.
GROBBELAAR, J. U. 2009a. Factors Governing Algal Growth in Photobioreactors: The "Open" Versus "Closed" Debate. *Journal of Applied Phycology,* 21,(5), 489-492.
GROBBELAAR, J. U. 2009b. Upper Limits of Photosynthetic Productivity and Problems of Scaling. *Journal of Applied Phycology,* 21,(5), 519-522.
GROBBELAAR, J. U. 2010. Microalgal Biomass Production: Challenges and Realities. *Photosynthesis Research,* 106,(1-2), 135-144.
GUAN, Q. Q., SAVAGE, P. E. & WEI, C. H. 2012a. Gasification of Alga Nannochloropsis Sp in Supercritical Water. *Journal of Supercritical Fluids,* 61, 139-145.
GUAN, Q. Q., WEI, C. H. & SAVAGE, P. E. 2012b. Kinetic Model for Supercritical Water Gasification of Algae. *Physical Chemistry Chemical Physics,* 14,(9), 3140-3147.

HALL, C., BALOGH, S. & MURPHY, D. 2009. What Is the Minimum EROI That a Sustainable Society Must Have? *Energies,* 2,(1), 25-47.
HALL, C. A. S. & KLITGAARD, K. A. 2012. Energy and the Wealth of Nations: Understanding the Biophysical Economy. New York: Springer.
HANNON, M., GIMPEL, J., TRAN, M., RASALA, B. & MAYFIELD, S. 2010. Biofuels from Algae: Challenges and Potential. *Biofuels,* 1,(5), 763–784.
HANOTU, J., BANDULASENA, H. C. H. & ZIMMERMAN, W. B. 2012. Microflotation Performance for Algal Separation. *Biotechnology and Bioengineering,* 109,(7), 1663-1673.
HARITH, Z. T., YUSOFF, F. M., MOHAMED, M. S., DIN, M. S. M. & ARIFF, A. B. 2009. Effect of Different Flocculants on the Flocculation Performance of Microalgae, Chaetoceros Calcitrans, Cells. *African Journal of Biotechnology,* 8,(21), 5971-5978.
HARUN, R. & DANQUAH, M. K. 2011. Enzymatic Hydrolysis of Microalgal Biomass for Bioethanol Production. *Chemical Engineering Journal,* 168,(3), 1079-1084.
HARUN, R., DANQUAH, M. K. & FORDE, G. M. 2010. Microalgal Biomass as a Fermentation Feedstock for Bioethanol Production. *Journal of Chemical Technology & Biotechnology,* 85,(2), 199-203.
HARUN, R., DAVIDSON, M., DOYLE, M., GOPIRAJ, R., DANQUAH, M. & FORDE, G. 2011a. Technoeconomic Analysis of an Integrated Microalgae Photobioreactor, Biodiesel and Biogas Production Facility. *Biomass & Bioenergy,* 35,(1), 741-747.
HARUN, R., JASON, W. S. Y., CHERRINGTON, T. & DANQUAH, M. K. 2011b. Exploring Alkaline Pre-Treatment of Microalgal Biomass for Bioethanol Production. *Applied Energy,* 88,(10), 3464-3467.
HARVEY, P. J., PSYCHA, M., KOKOSSIS, A., ABUBAKAR, A. L., TRIVEDI, V., SWAMY, R., COWAN, A. K., SCHROEDER, D., HIGHFIELD, A., REINHARDT, G., GARTNER, S., MCNEIL, J., DAY, P., BROCKEN, M., VARRIE, J. & BEN-AMOTZ, A. 2012. Glycerol Production by Halophytic Microalgae: Strategy for Producing Industrial Quantities in Saline Water. *20th European Biomass Conference and Exhibition.*
HEAVEN, S., MILLEDGE, J. & ZHANG, Y. 2011. Comments on 'Anaerobic Digestion of Microalgae as a Necessary Step to Make Microalgal Biodiesel Sustainable'. *Biotechnology Advances,* 29,(1), 164-7.
HEILMANN, S. M., DAVIS, H. T., JADER, L. R., LEFEBVRE, P. A., SADOWSKY, M. J., SCHENDEL, F. J., VON KEITZ, M. G. & VALENTAS, K. J. 2010. Hydrothermal Carbonization of Microalgae. *Biomass & Bioenergy,* 34,(6), 875-882.
HENDRICKS, D. 2010. *Fundamentals of Water Treatment Unit Processes: Physical, Chemical, and Biological,* Boca Raton, FL, CRC Press.
HENRIKSON, R. 2008. *In the Beginning Were Blue-Green Algae* [Online]. Ronore Enterprises. Available: http://www.spirulinasource.com/earthfoodch1a.html [Accessed 2009].
HENZE, M., VAN LOOSDRECHT, M. C. M., EKAMA, G. A. & BRDJANOVIC, D. 2008. *Biological Wastewater Treatment - Principles, Modelling and Design,* London, IWA.
HIDALGO, P., TORO, C., CIUDAD, G. & NAVIA, R. 2013. Advances in Direct Transesterification of Microalgal Biomass for Biodiesel Production. *Reviews in Environmental Science and Bio/Technology*, 1-21.
HIERHOLTZER, A. & AKUNNA, J. C. 2012. Modelling Sodium Inhibition on the Anaerobic Digestion Process. *Water Science and Technology,* 66,(7), 1565-1573.

HIRANO, A., HON-NAMI, K., KUNITO, S., HADA, M. & OGUSHI, Y. 1998. Temperature Effect on Continuous Gasification of Microalgal Biomass: Theoretical Yield of Methanol Production and Its Energy Balance. *Catalysis Today,* 45,(1-4), 399-404.

HONDZO, M. M., KAPUR, A. & LEMBI, C. A. 1997. The Effect of Small-Scale Fluid Motion on the Green Alga Scenedesmus Quadricauda. *Hydrobiologia,* 364,(2), 225-235.

HONEYWELL. 2011. *Unisim Design* [Online]. Available: http://hpsweb.honeywell.com/Cultures/en-US/Products/ControlApplications/simulation/UniSimDesign/default.htm [Accessed 27/09/2011].

HOUSE OF COMMONS ENERGY AND CLIMATE CHANGE COMMITTEE 2011. Energy and Climate Change - Eighth Report.UK Energy Supply: Security or Independence? . London.

HOWE, C. 2012. Direct Electricity Generation from Microalgae Using Biophotovoltaics. *Algal biotechnology; biofuels and beyond.* UCL, London.

HUNTLEY, M. E. & REDALJE, D. J. 2007. CO2 Mitigation and Renewable Oil from Photosynthetic Microbes: A New Appraisal. *Mitigation and Adaptation Strategies for Global Change.*

HUTCHINSON, N., BINGHAM, N., MURRELL, N., FARID, S. & HOARE, M. 2006. Shear Stress Analysis of Mammalian Cell Suspensions for Prediction of Industrial Centrifugation and Its Verification. *Biotechnology and Bioengineering,* 95,(3), 483-491.

IDEL'CHIK, I. E. 1986. *Handbook of Hydraulic Resistance* Washington, Hemisphere Pub. Corp.

IFRF. INTERNATIONAL FLAME RESEARCH FOUNDATION 2004. *Online Combustion Handbook. Method from Combustion File 24*.

ILLMAN, A. M., SCRAGG, A. H. & SHALES, S. W. 2000. Increase in Chlorella Strains Calorific Values When Grown in Low Nitrogen Medium. *Enzyme and Microbial Technology,* 27,(8), 631-635.

INTERNATIONAL WATER ASSOCIATION. 2010. *Coagulation and Flocculation in Water and Wastewater Treatment* [Online]. Available: http://www.iwawaterwiki.org/xwiki/bin/view/Articles/CoagulationandFlocculationinWaterandWastewaterTreatment#Information [Accessed 23/01 2013].

IRISH ENVIRONMENTAL PROTECTION AGENCY 1997. Waste Water Treatment Manuals: Primary, Secondary and Tertiary Treatment. Wexford.

JAGO, C. F., KENNAWAY, G. M., NOVARINO, G. & JONES, S. E. 2007. Size and Settling Velocity of Suspended Flocs During a Phaeocystis Bloom in the Tidally Stirred Irish Sea, Nw European Shelf. *Marine Ecology-Progress Series,* 345, 51-62.

JAMES, S. C. & BORIAH, V. 2010. Modeling Algae Growth in an Open-Channel Raceway. *Journal of Computational Biology,* 17,(7), 895-906.

JANA, A. K. 2009. *Process Simulation and Control Using Aspen,* New Dehli, PHI.

JANELT, G., BOLT, P., GERBSCH, N., BUCHHOLZ, R. & CHO, M. G. 1997. The Lamellar Settler – a Low-Cost Alternative for Separating the Micro-Alga Chlorella Vulgaris from a Cultivation Broth? *Applied microbiology and biotechnology,* 48,(1), 6-10.

JANSSEN, M., SLENDERS, P., TRAMPER, J., MUR, L. R. & WIJFFELS, R. 2001. Photosynthetic Efficiency of Dunaliella Tertiolecta under Short Light/Dark Cycles. *Enzyme and Microbial Technology,* 29,(4–5), 298-305.

JENA, U. & DAS, K. C. 2011. Comparative Evaluation of Thermochemical Liquefaction and Pyrolysis for Bio-Oil Production from Microalgae. *Energy & Fuels,* 25,(11), 5472-5482.

JENSSEN, T. K. & KONGSHAUG, G. Year. Energy Consumption and Greenhouse Gas Emissions in Fertiliser Production. *In:* International Fertiliser Society, 2003 London.
JIMENEZ, C., COSSIO, B. R. & NIELL, F. X. 2003. Relationship between Physicochemical Variables and Productivity in Open Ponds for the Production of Spirulina: A Predictive Model of Algal Yield. *Aquaculture,* 221,(1-4), 331-345.
JOHNSON, D. A., WEISSMAN, J. C. & GOEBEL, R. P. 1988. An Outdoor Test Facility for Large Scale Production of Microalgae. Golden, Colorado: SERI.
JOLHNKAI, G. 1997. Basic River Water Quality Models. Paris: UNESCO.
JONKER, J. G. G. & FAAIJ, A. P. C. 2013. Techno-Economic Assessment of Micro-Algae as Feedstock for Renewable Bio-Energy Production. *Applied Energy,* 102,(0), 461-475.
JORQUERA, O., KIPERSTOK, A., SALES, E. A., EMBIRUÇU, M. & GHIRARDI, M. L. 2010. Comparative Energy Life-Cycle Analyses of Microalgal Biomass Production in Open Ponds and Photobioreactors. *Bioresource Technology,* 101,(4), 1406-1413.
JOSHI, J. B., ELIAS, C. B. & PATOLE, M. S. 1996. Role of Hydrodynamic Shear in the Cultivation of Animal, Plant and Microbial Cells. *Chemical Engineering Journal and the Biochemical Engineering Journal,* 62,(2), 121-141.
KADAM, K. L. 2002. Environmental Implications of Power Generation Via Coal-Microalgae Cofiring. *Energy,* 27,(10), 905-922.
KANELLOS, M. 2009. *Algae Biodiesel: It's $33 a Gallon* [Online]. Greentech Media Available: http://www.greentechmedia.com/articles/algae-biodiesel-its-33-a-gallon-5652.html [Accessed 03/02/2009].
KAZAMIA, E., ALDRIDGE, D. C. & SMITH, A. G. 2012. Synthetic Ecology – a Way Forward for Sustainable Algal Biofuel Production? *Journal of Biotechnology,* 162,(1), 163–169.
KELLLY, M. S. & DWORJANYN, S. 2008. The Potential of Marine Biomass for Anaerobic Biogas Production a Feasibility Study with Recommendations for Further Research. Scotland: The Crown Estate on behalf of the Marine Estate.
KELLY, J. 2011. *The Great Biofuel Boondoggle* [Online]. Levittown, PA. : Calkins Media, Inc. Available: http://www.phillyburbs.com/news/local/courier_times_news/opinion/oped/the-great-biofuel-boondoggle/article_1d667241-4ec4-5e40-b74c-6a6b6ccd1b00.html?cbst=83 [Accessed 23/01/2012].
KEMP, I. C. 2007. *Pinch Analysis and Process Integration : A User Guide on Process Integration for the Efficient Use of Energy* Oxford Butterworth-Heinemann.
KESTIN, J., SOKOLOV, M. & WAKEHAM, W. A. 1978. Viscosity of Liquid Water in the Range -8 to 150 °C. *Journal of Physical and Chemical Reference Data,* 7,(3), 941-948.
KETCHUM, B. H. & REDFIELD, A. C. 1949. Some Physical and Chemical Characteristics of Algae Growth in Mass Culture. *Journal of Cellular and Comparative Physiology,* 33,(3), 281-299.
KETHEESAN, B. & NIRMALAKHANDAN, N. 2011. Development of a New Airlift-Driven Raceway Reactor for Algal Cultivation. *Applied Energy,* In Press, Corrected Proof.
KHOO, H. H., SHARRATT, P. N., DAS, P., BALASUBRAMANIAN, R. K., NARAHARISETTI, P. K. & SHAIK, S. 2011. Life Cycle Energy and CO2 Analysis of Microalgae-to-Biodiesel: Preliminary Results and Comparisons. *Bioresource Technology,* 102,(10), 5800-5807.

KLEMES, J., FRIEDLER, F., BULATOV, I. & VARBANOV, P. 2010. *Sustainability in the Process Industry : Integration and Optimization: Integration and Optimization*, McGraw-Hill Publishing.

KLEMM, D., HEUBLEIN, B., FINK, H.-P. & BOHN, A. 2005. Cellulose: Fascinating Biopolymer and Sustainable Raw Material. *ChemInform,* 44,(22), 3358-3393.

KNOPF, F. C. 2011. *Modeling, Analysis and Optimization of Process and Energy Systems*, Wiley.

KNOTHE, G., VAN GERPEN, J. & KRAHL, J. (eds.) 2005. *The Biodiesel Handbook,* Champaign, Illinois: AOCS.

KNUCKEY, R. M., BROWN, M. R., ROBERT, R. & FRAMPTON, D. M. F. 2006. Production of Microalgal Concentrates by Flocculation and Their Assessment as Aquaculture Feeds. *Aquacultural Engineering,* 35,(3), 300-313.

KOVALYOVA, S. 2009. *European Body Sees Algae Fuel Industry in 10-15 Years* [Online]. Thomson Reuters. Available: http://www.reuters.com/article/idUSTRE5526HY20090603 [Accessed 03/06/2009].

KROMKAMP, J. & WALSBY, A. E. 1990. A Computer-Model of Buoyancy and Vertical Migration in Cyanobacteria. *Journal of Plankton Research,* 12,(1), 161-183.

KRUSE, O. & HANKAMER, B. 2010. Microalgal Hydrogen Production. *Current Opinion in Biotechnology,* 21,(3), 238-243.

KRUSE, O., RUPPRECHT, J., MUSSGNUG, J. R., DISMUKES, G. C. & HANKAMER, B. 2005. Photosynthesis: A Blueprint for Solar Energy Capture and Biohydrogen Production Technologies. *Photochemical & Photobiological Sciences,* 4,(12), 957-970.

LARDON, L., HELIAS, A., SIALVE, B., STAYER, J. P. & BERNARD, O. 2009. Life-Cycle Assessment of Biodiesel Production from Microalgae. *Environmental Science & Technology,* 43,(17), 6475-6481.

LAWS, E. A. & BERNING, J. L. 1991. A Study of the Energetics and Economics of Microalgal Mass Culture with the Marine Chlorophyte Tetraselmis Suecica: Implications for Use of Power Plant Stack Gases. *Biotechnology and Bioengineering,* 37,(10), 936-947.

LEE, A., LEWIS, D. & ASHMAN, P. 2009. Microbial Flocculation, a Potentially Low-Cost Harvesting Technique for Marine Microalgae for the Production of Biodiesel. *Journal of Applied Phycology,* 21,(5), 559-567.

LEE, D. H. 2011. Algal Biodiesel Economy and Competition among Bio-Fuels. *Bioresource Technology,* 102,(1), 43-49.

LEFEBVRE, O. & MOLETTA, R. 2006. Treatment of Organic Pollution in Industrial Saline Wastewater: A Literature Review. *Water Research,* 40,(20), 3671-3682.

LEMON, D. 2008. Biofuels: The Motor Industry Perspective. *Biofuels: Technology meets strategy.* London: SCI.

LEWIS, D. 2009. *Clean Algae Biofuel Project Leads World in Productivity* [Online]. The American Association for the Advancement of Science, . Available: http://www.eurekalert.org/pub_releases/2009-11/uoa-cab110409.php [Accessed 10/12/2009].

LI, J., XU, N. S. & SU, W. W. 2003. Online Estimation of Stirred-Tank Microalgal Photobioreactor Cultures Based on Dissolved Oxygen Measurement. *Biochemical Engineering Journal,* 14,(1), 51-65.

LIU, J., HUANG, J., FAN, K. W., JIANG, Y., ZHONG, Y., SUN, Z. & CHEN, F. 2010. Production Potential of Chlorella Zofingienesis as a Feedstock for Biodiesel. *Bioresource Technology,* 101,(22), 8658–8663.

LIU, J., MUKHERJEE, J., HAWKES, J. J. & WILKINSON, S. J. 2013. Optimization of Lipid Production for Algal Biodiesel in Nitrogen Stressed Cells of Dunaliella Salina Using Ftir Analysis. *Journal of Chemical Technology & Biotechnology*, n/a-n/a.

LIU, X., CLARENS, A. F. & COLOSI, L. M. 2011. Meta-Model of Algae Bio Energy Life Cycles (Mabel). *LCA XI Conference.* Chicago.

LJUNGGREN, M. 2011. *Biological Production of Hydrogen and Methane. Process Evaluation and Design through Modeling..* Ph.D., Lund University.

LOVE, J. 2011. Enhanced Algal Growth in Microlensed Photobioreactors. *Algae for renewable energy.* University of Bath.

LOWE, S. A. 2003. Omission of Critical Reynolds Number for Open Channel Flows in Many Textbooks. *Journal of Professional Issues in Engineering Education and Practice,* 129,(1), 58-59.

LUNDQUIST, T. J., WOERTZ, I. C., QUINN, N. W. T. & BENEMANN, J. R. 2010. A Realistic Technology and Engineering Assessment of Algae Biofuel Production. Berkeley, California: Energy Biosciences Inst.

MAAS, C. 2009. Greenhouse Gas and Energy Co-Benefits of Water Conservation. Canadian Water and Wastewater Association.

MADDI, B., VIAMAJALA, S. & VARANASI, S. 2011. Comparative Study of Pyrolysis of Algal Biomass from Natural Lake Blooms with Lignocellulosic Biomass. *Bioresource Technology,* 102,(23), 11018-11026.

MAF NZ. 2007. *Light Nature and Measurement* [Online]. Wellington NZ: Sustainable Farming Fund Ministry of Agriculture and Forestry. Available: www.maf.govt.nz/sff/about-projects/search/L05-006/light.pdf [Accessed 01/12/2010].

MALETSKY, C. 2004. Kudu Gas Power in Five Years. *The Namibian*, 21/10/2004.

MANNWEILER, K. & HOARE, M. 1992. The Scale-Down of an Industrial Disk Stack Centrifuge. *Bioprocess Engineering,* 8,(1-2), 19-25.

MARQUEZ, F. J., SASAKI, K., NISHIO, N. & NAGAI, S. 1995. Inhibitory Effect of Oxygen Accumulation on the Growth of Spirulina Platensis. *Biotechnology Letters,* 17,(2), 225-228.

MARTINEZ, O. S. 1996. *Light Limited Growth of Chromatium Vinosium.* PhD, Autonomous University of Barcelona.

MARYLAND, U. O. 2007. *Photosynthetically Active Radiation (Par) as Observed from Satellites* [Online]. University of Maryland. Available: http://www.atmos.umd.edu/~srb/par/01project.htm [Accessed 2009].

MATA, T. M., MARTINS, A. A. & CAETANO, N. S. 2010. Microalgae for Biodiesel Production and Other Applications: A Review. *Renewable & Sustainable Energy Reviews,* 14,(1), 217-232.

MAYHEW, Y. R. & ROGERS, G. F. C. 1972. *Thermodynamic and Transport Properties of Fluids,* Oxford, Blackwell.

MCGHEE, J. E., STJULIAN, G. & DETROY, R. W. 1982. Continuous and Static Fermentation of Glucose to Ethanol by Immobilized Saccharomyces-Cerevisiae Cells of Different Ages. *Applied and Environmental Microbiology,* 44,(1), 19-22.

MCGINN, P. J., DICKINSON, K. E., BHATTI, S., FRIGON, J. C., GUIOT, S. R. & O'LEARY, S. J. B. 2011. Integration of Microalgae Cultivation with Industrial Waste Remediation for Biofuel and Bioenergy Production: Opportunities and Limitations. *Photosynthesis Research,* 109,(1-3), 231-247.

MCKENDRY, P. 2002. Energy Production from Biomass (Part 2): Conversion Technologies. *Bioresource Technology,* 83,(1), 47-54.
MCKINLAY, J. B. & HARWOOD, C. S. 2010. Photobiological Production of Hydrogen Gas as a Biofuel. *Current Opinion in Biotechnology,* 21,(3), 244-251.
MCKINNEY, R. E. 2004. *Environmental Pollution Control Microbiology,* New York, Marcel Decker.
MEDINA, M. & NEIS, U. 2007. Symbiotic Algal Bacterial Wastewater Treatment: Effect of Food to Microorganism Ratio and Hydraulic Retention Time on the Process Performance. *Water Science and Technology,* 55,(11), 165-171.
MELCHING, C. S. & FLORES, H. E. 1999. Reaeration Equations Derived from US Geological Survey Database. *Journal of Environmental Engineering-Asce,* 125,(5), 407-414.
MENDOZA, J. L., GRANADOS, M. R., DE GODOS, I., ACIÉN, F. G., MOLINA, E., BANKS, C. & HEAVEN, S. 2013a. Fluid-Dynamic Characterization of Real-Scale Raceway Reactors for Microalgae Production. *Biomass and Bioenergy,* 54, 267-275.
MENDOZA, J. L., GRANADOS, M. R., DE GODOS, I., ACIÉN, F. G., MOLINA, E., HEAVEN, S. & BANKS, C. J. 2013b. Oxygen Transfer and Evolution in Microalgal Culture in Open Raceways. *Bioresource Technology,* 137,(0), 188-195.
MERONEY, R. N. & COLORADO, P. E. 2009. Cfd Simulation of Mechanical Draft Tube Mixing in Anaerobic Digester Tanks. *Water Research,* 43,(4), 1040-1050.
MERRILL, A. L. & WATTS, B. K. 1955. Energy Values of Foods: Basis & Duration. Slight Revised February 1973. US Department of Agriculture.
METEOTEST. 2009. *Maps of Global Horizontal Radiation and Temperature* [Online]. Meteotest. Available: http://www.meteonorm.com/pages/en/downloads/maps.php [Accessed 2009].
MIAO, X. L. & WU, Q. Y. 2004. High Yield Bio-Oil Production from Fast Pyrolysis by Metabolic Controlling of Chlorella Protothecoides. *Journal of Biotechnology,* 110,(1), 85-93.
MIAO, X. L., WU, Q. Y. & YANG, C. Y. 2004. Fast Pyrolysis of Microalgae to Produce Renewable Fuels. *Journal of Analytical and Applied Pyrolysis,* 71,(2), 855-863.
MILLEDGE, J. J. 2010a. The Challenge of Algal Fuel : Economic Processing of the Entire Algal Biomass. *Condensed Matter -Materials Engineering Newsletter* [Online], 1. Available: http://mse.mcmaster.ca/Condensed%20Matter/January2010.pdf.
MILLEDGE, J. J. 2010b. The Potential Yield of Microalgal Oil. *Biofuels International* 4,(2), 44 45.
MILLEDGE, J. J. 2011a. Commercial Application of Microalgae Other Than as Biofuels: A Brief Review. *Reviews in Environmental Science and Biotechnology,* 10,(1), 31-41.
MILLEDGE, J. J. 2011b. The Potential Energy Challenges of Micro-Algal Process Operations for Fuel Production *Algae for Renewable Energy.* University of Bath.
MILLEDGE, J. J. 2012a. Micro-Algal Process Flow-Sheet Energy Balance Optimisation: Initial Software Evaluation. *Algal Biotechnology: Biofuels and Beyond.* UCL.

MILLEDGE, J. J. 2012b. Microalgae – Commercial Potential for Fuel, Food and Feed. *Food Science & Technology,* 26,(1), 26-28.
MILLEDGE, J. J. 2013. Micro-Algal Biorefineries. *Towards Establishing Value Chains for Bioenergy.* Swakopmund, Namibia: The African, Caribbean and Pacific Group of States (ACP) Science and Technology Programme.
MILLEDGE, J. J. & HEAVEN, S. 2011. Disc Stack Centrifugation Separation and Cell Disruption of Microalgae: A Technical Note. *Environment and Natural Resources Research,* 1,(1), 17-24.
MILLER, F. P., VANDOME, A. F. & MCBREWSTER, J. 2010. *Hydrogen Pinch : Hydrogen, Pinch Analysis, Petrochemical, Oil Refinery, Water Pinch Analysis, Timeline of Hydrogen Technologies, Chemical Element,* Beau Bassin, Mauritius, Alphascript.
MILLERO, F. J. & LEPPLE, F. K. 1973. The Density and Expansibility of Artificial Seawater Solutions from 0 to 40°C and 0 to 21 Chlorinity. *Marine Chemistry,* 1,(2), 89-104.
MINISTRY OF ENVIRONMENT AND TOURISM NAMIBIA. 2002. *Digital Atlas of Namibia* [Online]. Available: http://www.uni-koeln.de/sfb389/e/e1/download/atlas_namibia/main_namibia_atlas.html [Accessed 03/01 2012].
MINOWA, T. & SAWAYAMA, S. 1999. A Novel Microalgal System for Energy Production with Nitrogen Cycling. *Fuel,* 78,(10), 1213-1215.
MINOWA, T., YOKOYAMA, S., KISHIMOTO, M. & OKAKURA, T. 1995. Oil Production from Algal Cells of Dunaliella-Tertiolecta by Direct Thermochemical Liquefaction. *Fuel,* 74,(12), 1735-1738.
MIRANDA, J. R., PASSARINHO, P. C. & GOUVEIA, L. 2012. Pre-Treatment Optimization of Scenedesmus Obliquus Microalga for Bioethanol Production. *Bioresource Technology,* 104, 342-348.
MISRA, M. K., RAGLAND, K. W. & BAKER, A. J. 1993. Wood Ash Composition as a Function of Furnace Temperature. *Biomass & Bioenergy,* 4,(2), 103-116.
MOHEIMANI, N. & BOROWITZKA, M. 2006. The Long-Term Culture of the Coccolithophore & Leurochrysis Carterae (Haptophyta) in Outdoor Raceway Ponds. *Journal of Applied Phycology,* 18,(6), 703-712.
MOHEIMANI, N. R. 2005. *The Culture of Coccolithophorid Algae for Carbon Dioxide Bioremediation.* PhD, Murdoch University.
MOHN, F. H. 1988. Harvesting of Micro-Algal Biomass. *In:* BOROWITZKA, M. A. & BOROWITZKA, L. J. (eds.) *Micro-Algal Biotechnology* Cambridge: Cambridge University Press.
MOLINA-GRIMA, E., BELARBI, E.-H., ACIEN-FERNANDEZ, F. G., ROBLES-MEDINA, A. & YUSUF, C. 2003. Recovery of Microalgal Biomass and Metabolites: Process Options and Economics. *Biotechnology Advances,* 20, 491-515.
MOLINA, E. M., FERNANDEZ, F. G., CAMACHO, F. G., RUBIO, F. C. & CHISTI, Y. 2000. Scale-up of Tubular Photobioreactors. *Journal of Applied Phycology,* 12,(3-5), 355-368.
MOLINA GRIMA, E. 2011. Microalgal Mass Culture, Key Aspects in Design Open Outdoor Culture and Photobioreactors. *Biodiesel production from microalgae: science, engineering and economic evaluation.* Almeria.
MOLINA GRIMA, E., ACIEN FERNANDEZ, F. G., GARCIA CAMACHO, F. & CHISTI, Y. 1999. Photobioreactors: Light Regime, Mass Transfer, and Scaleup. *Journal of Biotechnology,* 70,(1-3), 231-247.
MOLINA GRIMA, E., BELARBI, E.-H., ACIEN-FERNANDEZ, F. G., ROBLES-MEDINA, A. & YUSUF, C. 2003. Recovery of Microalgal Biomass and Metabolites: Process Options and Economics. *Biotechnology Advances,* 20,(7-8), 491-515.

MOLINA GRIMA, E., FERNANDEZ, J., ACIEN, F. G. & CHISTI, Y. 2001. Tubular Photobioreactor Design for Algal Cultures. *Journal of Biotechnology,* 92,(2), 113-131.
MOLINA GRIMA, E., FERNANDEZ, J. M., ACIEN FERNANDEZ, S. & ACIEN FERNADEZ, F. 2010. Microalgae, Mass Culture Methods. *In:* FLICKINGER, M. C. (ed.) *Encyclopedia of Industrial Biotechnology, Bioprocess, Bioseparation and Cell Technology.* John Wiley.
MONTEIRO, J., SILVA, P. A. C., ARAUJO, O. Q. F. & MEDEIROS, J. L. 2010. Pareto Optimization of an Industrial Ecosystem: Sustainability Maximization. *Brazilian Journal of Chemical Engineering,* 27,(3), 429-440.
MONTY, J. S. 2005. *Developments in Smooth Wall Turbulent Duct Flows.* Ph.D., University of Melbourne.
MOODY, L. F. 1944. Friction Factors for Pipe Flow. *Trans. A. S. M. E.,* 66, 671-684.
MORAINE, R., SHELEF, G., MEYDAN, A. & LEVI, A. 1979. Algal Single Cell Protein from Wastewater-Treatment and Renovation Process. *Biotechnology and Bioengineering,* 21,(7), 1191-1207.
MULDER, K. & HAGENS, N. J. 2008. Energy Return on Investment: Toward a Consistent Framework. *Ambio,* 37,(2), 74-79.
MÜLLER, G., DENCHFIELD, S., MARTH, R. & SHELMERDINE, R. Year. Stream Wheels for Applications in Shallow and Deep Water. *In:* Proc. 32nd IAHR Congress, 2007 Venice. Paper 291.
NASA. 2009. *Surface Meteorology and Solar Energy* [Online]. Available: http://eosweb.larc.nasa.gov/sse/SurfaceSurface [Accessed 2009].
NREL. 2009. *U.S. Solar Radiation Resource Maps:* [Online]. NREL. Available: http://rredc.nrel.gov/solar/old_data/nsrdb/redbook/atlas/Table.html [Accessed 2009].
NREL. 2011. *Best Research Cell Efficiencies* [Online]. Available: http://www.nrel.gov/pv/ [Accessed 21/11/2011].
NURDOGAN, Y. & OSWALD, W. J. 1996. Tube Settling of High-Rate Pond Algae. *Water Science and Technology,* 33,(7), 229-241.
OFFICE OF GAS AND ELECTRICITY MARKETS 2009. Biodiesel, Glycerol and the Renewables Obligation. Decision Document.
OLGUIN, E. J. 2012. Dual Purpose Microalgae-Bacteria-Based Systems That Treat Wastewater and Produce Biodiesel and Chemical Products within a Biorefinery. *Biotechnology Advances,* 30,(5), 1031-1046.
OLGUÍN, E. J. 2003. Phycoremediation: Key Issues for Cost-Effective Nutrient Removal Processes. *Biotechnology Advances,* 22,(1-2), 81-91.
ORIGIN OIL 2010. Algae Harvesting, Dewatering and Extraction. *World Biofuel Markets.* Amsterdam.
OSWALD, W. J. 1988. Large-Scale Algal Culture Systems (Engineering Aspects). *In:* BOROWITZKA, M. A. & BOROWITZKA, L. J. (eds.) *Micro-Algal Biotechnology.* Cambridge: Cambridge University Press.
OZKURT, I. 2009. Qualifying of Safflower and Algae for Energy. *Energy Education Science and Technology Part a-Energy Science and Research,* 23,(1-2), 145-151.
PACKER, M. 2009. Algal Capture of Carbon Dioxide; Biomass Generation as a Tool for Greenhouse Gas Mitigation with Reference to New Zealand Energy Strategy and Policy. *Energy Policy,* 37,(9), 3428-3437.
PAINE, R. & VADAS, R. 1969. Calorific Values of Benthic Marine Algae and Their Postulated Relation to Invertebrate Food Preference. *Marine Biology,* 4, 79-86.

PAPAZI, A., MAKRIDIS, P. & DIVANACH, P. 2010. Harvesting Chlorella Minutissima Using Cell Coagulants. *Journal of Applied Phycology,* 22,(3), 349-355.
PARK, J. B. K., CRAGGS, R. J. & SHILTON, A. N. 2011. Wastewater Treatment High Rate Algal Ponds for Biofuel Production. *Bioresource Technology,* 102,(1, Sp. Iss. SI), 35-42.
PARK, S. & LI, Y. B. 2012. Evaluation of Methane Production and Macronutrient Degradation in the Anaerobic Co-Digestion of Algae Biomass Residue and Lipid Waste. *Bioresource Technology,* 111, 42-48.
PARLIAMENTARY OFFICE OF SCIENCE & TECHNOLOGY 2010. Global Carbon Trading. Postnote.
PARLIAMENTARY OFFICE OF SCIENCE & TECHNOLOGY 2011. Biofuels from Algae. Postnote.
PATE, R., KLISE, G. & WU, B. 2011. Resource Demand Implications for US Algae Biofuels Production Scale-Up. *Applied Energy,* 88,(10), 3377-3388.
PENG, W. M., WU, Q. Y. & TU, P. G. 2000. Effects of Temperature and Holding Time on Production of Renewable Fuels from Pyrolysis of Chlorella Protothecoides. *Journal of Applied Phycology,* 12,(2), 147-152.
PEPERZAK, L., COLIJN, F., KOEMAN, R., GIESKES, W. W. C. & JOORDENS, J. C. A. 2003. Phytoplankton Sinking Rates in the Rhine Region of Freshwater Influence. *Journal of Plankton Research,* 25,(4), 365-383.
PERALTA, Y., SANCHEZ, E. & KAFAROV, V. 2010. Exergy Analysis for Third Generation Biofuel Production from Microalgae Biomass. *Chemical Engineering Transactions,* 21, 1363-1368.
PERRY, R. H. & CHILTON, C. H. 1973. *Chemical Engineers' Handbook,* Tokyo, McGraw Hill.
PERSSON, S. P. E., BARTLETT, H. D., BRANDING, A. E. & REGAN, R. W. 1979. Agricultural Anaerobic Digesters. Pennsylvania State University.
PFEIFFER, T. J., LAWSON, T. B. & CHURCH, G. E. 2007. Engineering Considerations for Water Circulation in Crawfish Ponds with Paddlewheel Aerators. *Aquacultural Engineering,* 36,(3), 239-249.
PFROMM, P. H., AMANOR-BOADU, V. & NELSON, R. 2011. Sustainability of Algae Derived Biodiesel: A Mass Balance Approach. *Bioresource Technology,* 102,(2), 1185-1193.
PIENKOS, P. T. & DARZINS, A. 2009. The Promise and Challenges of Microalgal-Derived Biofuels. *Biofuels, Bioprod, Bioref*, 431-440.
PIRES, J. C. M., ALVIM-FERRAZ, M. C. M., MARTINS, F. G. & SIMOES, M. 2012. Carbon Dioxide Capture from Flue Gases Using Microalgae: Engineering Aspects and Biorefinery Concept. *Renewable & Sustainable Energy Reviews,* 16,(5), 3043-3053.
PLATT, T. & IRWIN, B. 1973. Caloric Content of Phytoplankton. *Limnology and Oceanography,* 18,(2), 306-310.
POELMAN, E., DE PAUW, N. & JEURISSEN, B. 1997. Potential of Electrolytic Flocculation for Recovery of Micro-Algae. *Resources, Conservation and Recycling,* 19,(1), 1-10.
POKOO-AIKINS, G., NADIM, A., EL-HALWAGI, M. M. & MAHALEC, V. 2010. Design and Analysis of Biodiesel Production from Algae Grown through Carbon Sequestration. *Clean Technologies and Environmental Policy,* 12,(3), 239-254.
PORTEOUS, G. C. 1983. Dewatering Sewage Sludge by Centrifuge. DEFRA.
POWELL, E. E., EVITTS, R. W., HILL, G. A. & BOLSTER, J. C. 2011. A Microbial Fuel Cell with a Photosynthetic Microalgae Cathodic Half Cell Coupled to a

Yeast Anodic Half Cell. *Energy Sources Part a-Recovery Utilization and Environmental Effects,* 33,(5), 440-448.
PUCHAJDA, B. & OLESZKIEWICZ, J. 2008. Impact of Sludge Thickening on Energy Recovery from Anaerobic Digestion. *Water Science and Technology,* 57,(3), 395-401.
PURCHAS, D. B. 1981. *Solid-Liquid Separation Technology* London, Uplands Press.
PUSHPARAJ, B., PELOSI, E., TORZILLO, G. & MATERASSI, R. 1993. Microbial Biomass Recovery Using a Synthetic Cationic Polymer. *Bioresource Technology,* 43,(1), 59-62.
PUTT, R. 2007. Algae as a Biodiesel Feedstock:A Feasibility Assessment. Department of Chemical Engineering, Auburn University,Alabama.
QUASCHNING, V. 2010. *Renewable Energy and Climate Change,* Chichester, Wiley.
RABE, A. E. & BENOIT, R. J. 1962. Mean Light Intensity - a Useful Concept in Correlating Growth Rates of Dense Cultures of Microalgae. *Biotechnology and Bioengineering,* 4, 377-390.
RAJA, R., HEMAISWARYA, S., ASHOK KUMAR, N., SRIDHAR, S. & RENGASAMY, R. 2008. A Perpespective on Biotechnological Potential of Microalgae. *Critical Review in Microbiology,* 34, 34-77.
RAMAKRISHNAN, B., KUMARASWAMY, S., MALLICK, K., ADHYA, T. K., RAO, V. R. & SETHUNATHAN, N. 1998. Effect of Various Anionic Species on Net Methane Production in Flooded Rice Soils. *World Journal of Microbiology and Biotechnology,* 14,(5), 743-749.
RAS, M., LARDON, L., BRUNO, S., BERNET, N. & STEYER, J.-P. 2011. Experimental Study on a Coupled Process of Production and Anaerobic Digestion of Chlorella Vulgaris. *Bioresource Technology,* 102,(1), 200-206.
RAWAT, I., RANJITH KUMAR, R., MUTANDA, T. & BUX, F. 2013. Biodiesel from Microalgae: A Critical Evaluation from Laboratory to Large Scale Production. *Applied Energy,* 103,(0), 444-467.
RAZON, L. F. & TAN, R. R. 2011. Net Energy Analysis of the Production of Biodiesel and Biogas from the Microalgae: Haematococcus Pluvialis and Nannochloropsis. *Applied Energy,* 88,(10), 3507-3514.
REIJNDERS, L. 2009. Microalgal and Terrestrial Transport Biofuels to Displace Fossil Fuels. *Energies,* 2,(1), 48-56.
RENEWABLE FUELS AGENCY 2008. The Gallagher Review of the Indirect Effects of Biofuels Production,.
RENNIE, G. 1853. The Propulsion of Vessels by Paddlewheels. *In:* TREDGOLD, T. (ed.) *The Steam Engine.* London: John Weale.
REYNOLDS, C. S. 1984. *The Ecology of Freshwater Phytoplankton,* Cambridge, Cambridge University Press.
RICHARDSON, J. F., HARKER, J. H. & BACKHURST, J. R. 2002. *Coulson & Richardson's Chemical Engineering; Volume 2, Fifth Edition, Particle Technology & Separation Processes,* Oxford, Elsevier Butterworth-Heinemann.
RITTMANN, B. E. & MCCARTY, P. L. 2001. *Environmental Biotechnology: Principles and Applications,* Singapore, McGraw-Hill.
ROBERTS, K. 1974. Crystalline Glycoprotein Cell Walls of Algae: Their Structure, Composition and Assembly. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences,* 268,(891), 129-146.
RODRIGUEZ, M. E., SAMO, A. L., HOZBOR, D. F. & YANTORNO, O. M. 1993. Effect of Hydromechanical Forces on the Production of Filamentous Haemagglutinin and Pertussis Toxin of≪I≫Bordetella

Pertussis≪/I&Gt. *Journal of Industrial Microbiology & Biotechnology,* 12,(2), 103-108.
ROESIJADI, G., JONES, S. B., SNOWDEN-SWAN, L. J. & ZHU, Y. 2010. Macroalgae as a Biomass Feedstock: A Preliminary Analysis. Washington: U.S. Department of Energy.
ROGERS, G. F. C. & MAYHEW, Y. R. 1992. *Engineering Thermodynamics - Work & Heat Transfer,* New York, Longman.
ROSENBAUM, M., HE, Z. & ANGENENT, L. T. 2010. Light Energy to Bioelectricity: Photosynthetic Microbial Fuel Cells. *Current Opinion in Biotechnology,* 21,(3), 259-264.
ROSSI, N., JAOUEN, O., LEGENTILHOMME, P. & PETIT, I. 2004. Harvesting of Cyanobacterium Arthrospira Platensis Usmg Organic Filtration Membranes. *Food and Bioproducts Processing,* 82,(C3), 244-250.
SAIDUR, R., ABDELAZIZ, E. A., DEMIRBAS, A., HOSSAIN, M. S. & MEKHILEF, S. 2011. A Review on Biomass as a Fuel for Boilers. *Renewable & Sustainable Energy Reviews,* 15,(5), 2262-2289.
SALTER, A. & BANKS, C. 2008. Anaerobic Digestion: Overall Energy Balances – Parasitic Inputs & Beneficial Outputs. *Sustainable Organic Resources Partnership - Advances in Biological Processes for Organics and Energy recycling.* Birmingham.
SAMSON, R. & LEDUY, A. 1983. Improved Performance of Anaerobic Digestion of Spirulina Maxima Algal Biomass by Addition of Carbon-Rich Wastes. *Biotechnology Letters,* 5,(10), 677-682.
SANCHEZ, E., OJEDA, K., EL-HALWAGI, M. & KAFAROV, V. 2011. Biodiesel from Microalgae Oil Production in Two Sequential Esterification/Transesterification Reactors: Pinch Analysis of Heat Integration. *Chemical Engineering Journal,* 176-77, 211-216.
SANDER, K. & MURTHY, G. S. 2010. Life Cycle Analysis of Algae Biodiesel. *International Journal of Life Cycle Assessment,* 15,(7), 704-714.
SAWAYAMA, S., MINOWA, T. & YOKOYAMA, S. Y. 1999. Possibility of Renewable Energy Production and CO2 Mitigation by Thermochemical Liquefaction of Microalgae. *Biomass and Bioenergy,* 17,(1), 33-39.
SAZDANOFF, N. 2006. *Modeling and Simulation of the Algae to Biodiesel Fuel Cycle.* BSc, Ohio State University.
SCHEFFLAN, R. 2011. *Teach Yourself the Basics of Aspen Plus,* Hoboken, NJ, Wiley.
SCHENK, P., THOMAS-HALL, S., STEPHENS, E., MARX, U., MUSSGNUG, J., POSTEN, C., KRUSE, O. & HANKAMER, B. 2008. Second Generation Biofuels: High-Efficiency Microalgae for Biodiesel Production. *BioEnergy Research,* 1,(1), 20-43.
SCHLARB-RIDLEY, B. 2011. Algal Research in the UK. A Report for Bbsrc. BBSRC.
SCHLESINGER, A., EISENSTADT, D., BAR-GIL, A., CARMELY, H., EINBINDER, S. & GRESSEL, J. 2012. Inexpensive Non-Toxic Flocculation of Microalgae Contradicts Theories; Overcoming a Major Hurdle to Bulk Algal Production. *Biotechnology Advances,* 30,(5), 1023-1030
SENIOR, J., SAENGER, N. & MULLER, G. 2010. New Hydropower Converters for Very Low-Head Differences. *Journal of Hydraulic Research,* 48,(6), 703-714.
SERVICE, R. F. 2011. Algae's Second Try. *Science,* 333, 1238-39.
SHEEHAN, J., DUNAHAY, T., BENEMANN, J. & ROESSLER, P 1998. A Look Back at the US Department of Energy's Aquatic Species Program - Biodiesel from Algae. National Renewable Energy Laboratory NREL.

SHELEF, G., SUKENIK, A. & GREEN, M. 1984a. Microalgae Harvesting and Processing: A Literature Review. Golden, Colorado: Solar Energy Research Institute.

SHELEF, G., SUKENIK, A. & GREEN, M. Year. Separation and Harvesting of Marine Microalgal Biomass. *In:* BARCLAY, W. R. & MCINTOSH, R. P., eds. Algal biomass technology, 1984b Colorado. J. Cramer.

SHEN, Y., YUAN, W., PEI, Z. J., WU, Q. & MAO, E. 2009. Microalgae Mass Production Methods. *Transactions of the Asabe,* 52,(4), 1275-1287.

SHENOY, A. U. & SHENOY, U. V. 2012. Targeting and Design of Energy Allocation Networks with Carbon Capture and Storage. *Chemical Engineering Science,* 68,(1), 313-327.

SHIRVANI, T., YAN, X., INDERWILDI, O. R., EDWARDS, P. P. & KING, D. A. 2011. Life Cycle Energy and Greenhouse Gas Analysis for Algae-Derived Biodiesel. *Energy & Environmental Science,* 4,(10), 3773-3778.

SIALVE, B., BERNET, N. & BERNARD, O. 2009. Anaerobic Digestion of Microalgae as a Necessary Step to Make Microalgal Biodiesel Sustainable. *Biotechnology Advances,* 27,(4), 409-416.

SILLS, D. L., PARAMITA, V., FRANKE, M. J., JOHNSON, M. C., AKABAS, T. M., GREENE, C. H. & TESTER, J. W. 2012. Quantitative Uncertainty Analysis of Life Cycle Assessment for Algal Biofuel Production. *Environmental Science & Technology,* 47,(2), 687-694.

SIMAPRO UK. 2011. *About Simapro* [Online]. Available: http://www.simapro.co.uk [Accessed 18/07/2011].

SIMS, R. E. H. 2002. *The Brilliance of Bioenergy,* London, James & James.

SINGH, A., NIGAM, P. S. & MURPHY, J. D. 2011. Mechanism and Challenges in Commercialisation of Algal Biofuels. *Bioresource Technology,* 102,(1), 26-34.

SINGH, A. & OLSEN, S. I. 2011. A Critical Review of Biochemical Conversion, Sustainability and Life Cycle Assessment of Algal Biofuels. *Applied Energy,* 88,(10), 3548–3555.

SINGH, J. & GU, S. 2010. Biomass Conversion to Energy in India-a Critique. *Renewable & Sustainable Energy Reviews,* 14,(5), 1367-1378.

SINGH, S., BHUSHAN, K. N. & BANERJEE, U. C. 2005. Bioactive Compounds from Cyanobacteria and Microalgae: An Overview. *Critical Review Biotechnology,* 25,(3), 73-95.

SINGH, U. & AHLUWALIA, A. 2013. Microalgae: A Promising Tool for Carbon Sequestration. *Mitigation and Adaptation Strategies for Global Change,* 18,(1), 73-95.

SINNOTT, R. K. 2005. *Coulson & Richardson's Chemical Engineering; Volume 6, Fourth Edition,Chemical Engineering Design,* Oxford, Elsevier Butterworth-Heinemann.

SIPPULA, O. 2010. *Fine Particle Formation and Emissions in Biomass Combustion.* Ph.D., University of Eastern Finland.

ŞIRIN, S., TROBAJO, R., IBANEZ, C. & SALVADÓ, J. 2012. Harvesting the Microalgae; Phaeodactylum Tricornutum with Polyaluminum Chloride, Aluminium Sulphate, Chitosan and Alkalinity-Induced Flocculation. *Journal of Applied Phycology*, 1-14.

SMAYDA, T. J. 1970. The Suspension and Sinking of Phytoplankton in the Sea. *In:* BARNES, H. (ed.) *Oceanography and Marine Biology Annual Review.* London: George Allen & Unwin.

SMIL, V. 1999. *Energies,* Cambridge MA, MIT.

SMITH, V. H., STURM, B. S. M., DENOYELLES, F. J. & BILLINGS, S. A. 2010. The Ecology of Algal Biodiesel Production. *Trends in Ecology & Evolution,* 25,(5), 301-309.
SOLAZYME. 2012. *Meeting the Growing Need for Renewable Fuels* [Online]. Available: http://solazyme.com/fuels [Accessed 20/02/2012].
SOMPECH, K., CHISTI, Y. & SRINOPHAKUN, T. 2012. Design of Raceway Ponds for Producing Microalgae. *Biofuels,* 3,(4), 387-397.
SORENSEN, B. 2012. *Hydrogen and Fuel Cells Emerging Technologies and Applications.,* Oxford, Elsevier.
SORGUVEN, E. & OZILGEN, M. 2010. Thermodynamic Assessment of Algal Biodiesel Utilization. *Renewable Energy,* 35,(9), 1956-1966.
SOURNIA, A. (ed.) 1978. *Phytoplankton Manual,* Paris: UNESCO.
SPEIGHT, J. 2005. *Lange's Handbook of Chemistry*, McGraw-Hill
SPIRAX SARCO. 2012. *Energy Consumption of Tanks and Vats* [Online]. Available: http://www.spiraxsarco.com/resources/steam-engineering-tutorials/steam-engineering-principles-and-heat-transfer/energy-consumption-of-tanks-and-vats.asp [Accessed 10/12 2012].
SPOLAORE, P., JOANNIS-CASSAN, C., DURAN, E. & ISAMBET, A. 2006. Commercial Applications of Microalgae. *Journal Of Biosciences & Bioengineering,* 101,(2), 87-96.
SRINIVAS, T. 2008. *Environmental Biotechnology* New Delhi, New Age International
ST JOHN, J. 2009. *Algae Company Number 56: Plankton Power* [Online]. Greentech Media Inc. Available: http://www.greentechmedia.com/articles/read/plankton-power-another-algae-biodiesel-maker-emerges [Accessed 10/12/2009].
STEPHENSON, A. L. 2009. *The Sustainability of First- and Second-Generation Biofuels Using Life Cycle Analysis.* PhD, University of Cambridge.
STEPHENSON, A. L., KAZAMIA, E., DENNIS, J. S., HOWE, C. J., SCOTT, S. A. & SMITH, A. G. 2010. Life-Cycle Assessment of Potential Algal Biodiesel Production in the United Kingdom: A Comparison of Raceways and Air-Lift Tubular Bioreactors. *Energy & Fuels,* 24 (7), 4062–4077.
STEPHENSON, P. G., MOORE, C. M., TERRY, M. J., ZUBKOV, M. V. & BIBBY, T. S. 2011. Improving Photosynthesis for Algal Biofuels: Toward a Green Revolution. *Trends in Biotechnology,* 29,(12), 615–623.
STRIK, D., TERLOUW, H., HAMELERS, H. & BUISMAN, C. 2008. Renewable Sustainable Biocatalyzed Electricity Production in a Photosynthetic Algal Microbial Fuel Cell (Pamfc). *Applied Microbiology and Biotechnology,* 81,(4), 659-668.
STUCKI, S., VOGEL, F., LUDWIG, C., HAIDUC, A. G. & BRANDENBERGER, M. 2009. Catalytic Gasification of Algae in Supercritical Water for Biofuel Production and Carbon Capture. *Energy & Environmental Science,* 2,(5), 535-541.
SUBHADRA, B. & GRINSON, G. 2011. Algal Biorefinery-Based Industry: An Approach to Address Fuel and Food Insecurity for a Carbon-Smart World. *Journal of the Science of Food and Agriculture,* 91,(1), 2-13.
SUKENIK, A., BILANOVIC, D. & SHELEF, G. 1988. Flocculation of Microalgae in Brackish and Sea Waters. *Biomass,* 15,(3), 187-199.
SYMONS, G. E. & BUSWELL, A. M. 1933. The Methane Fermentation of Carbohydrates1,2. *Journal of the American Chemical Society,* 55,(5), 2028-2036.
TAMIYA, H. 1957. Mass Culture of Algae. *Annual Review of Plant Physiology,* 8,(1), 309-334.

TARCHEVSKY, I. A. & MARCHENKO, G. N. 1991. *Cellulose: Biosynthesis and Structure,* Berlin, Springer.
TAYLOR, G. 2008. Biofuels and the Biorefinery Concept. *Energy Policy,* 36**,**(12), 4406-4409.
TCHOBANOGLOUS, G. & SCHROEDER, E. 1985. *Water Quality: Characteristics, Modeling and Modification*, Prentice Hall.
TERRY, K. L. & RAYMOND, L. P. 1985. System-Design for the Autotrophic Production of Microalgae. *Enzyme and Microbial Technology,* 7**,**(10), 474-487.
THIMIJAN, R. W. & HEINS, R. D. 1983. Photometric, Radiometric, and Quantum Light Units of Measure - a Review of Procedures for Interconversion. *Hortscience,* 18**,**(6), 818-822.
THORNE, R., HU, H. N., SCHNEIDER, K., BOMBELLI, P., FISHER, A., PETER, L. M., DENT, A. & CAMERON, P. J. 2011. Porous Ceramic Anode Materials for Photo-Microbial Fuel Cells. *Journal of Materials Chemistry,* 21**,**(44), 18055-18060.
TOKUSOGLU, O. & UNAL, M. K. 2003. Biomass Nutrient Profiles of Three Microalgae: Spirulina Platensis, Chlorella Vulgaris, and Isochrisis Galbana. *Journal of Food Science,* 68**,**(4), 1144-1148.
TORRI, C., ALBA, L. G., SAMORI, C., FABBRI, D. & BRILMAN, D. W. F. 2012. Hydrothermal Treatment (Htt) of Microalgae: Detailed Molecular Characterization of Htt Oil in View of Htt Mechanism Elucidation. *Energy & Fuels,* 26**,**(1), 658-671.
TREDICI, M. R. 2010. Photobiology of Microalgae Mass Cultures: Understanding the Tools for the Next Green Revolution. *Biofuels,* 1**,**(1), 143-162.
TREDICI, M. R. 2012. Energy Balance of Microalgae Cultures in Photobioreactors and Ponds. *The EU Workshop Life Cycle Analysis of Algal Based Biofuels.* Brussels.
TWIDELL, J. & WEIR, T. 2006. *Renewable Energy Sources,* London, Taylor & Francis.
UDUMAN, N., QI, Y., DANQUAH, M. K., FORDE, G. M. & HOADLEY, A. 2010. Dewatering of Microalgal Cultures: A Major Bottleneck to Algae-Based Fuels. *Journal of Renewable and Sustainable Energy,* 2**,**(1), 012701-15.
US CONGRESS 2007. Energy Independence and Security Act. 110th Congress ed. USA.
US DEPARTMENT OF AGRICULTURE NATURAL RESOURCES CONSERVATION SERVICE. 2012. *National Soil Survey Handbook - Part 618 – Soil Properties and Qualities* [Online]. Available: http://soils.usda.gov/technical/handbook/contents/part618.html#43 [Accessed 12/03/2012].
US ENVIROMENTAL PROTECTION AGENCY 1979. Process Design Manual for Sludge Treatment and Disposal.
US ENVIRONMENTAL PROTECTION AGENCY. 2013. *Combined Heat and Power Partnership* [Online]. Available: http://www.epa.gov/chp/index.html [Accessed 07/02 2013].
UZIR, M. H. & MAT DON, M. 2007. Biochemical Engineering - a Concise Introduction. Penang: Universiti Sains Malaysia.
VAN DEN HENDE, S., VERVAEREN, H., DESMET, S. & BOON, N. 2011. Bioflocculation of Microalgae and Bacteria Combined with Flue Gas to Improve Sewage Treatment. *New Biotechnology,* 29**,**(1), 23-31.
VAN LERLAND, E. T. & PEPERZAK, L. 1984. Separation of Marine Seston and Density Determination of Marine Diatoms by Density Gradient Centrifugation. *Journal of Plankton Research,* 6**,**(1), 29-44.

VANDAMME, D., FOUBERT, I., MEESSCHAERT, B. & MUYLAERT, K. 2010. Flocculation of Microalgae Using Cationic Starch. *Journal of Applied Phycology,* 22,(4), 525-530.
VANDAMME, D., PONTES, S. C. V., GOIRIS, K., FOUBERT, I., PINOY, L. J. J. & MUYLAERT, K. 2011. Evaluation of Electro-Coagulation-Flocculation for Harvesting Marine and Freshwater Microalgae. *Biotechnology and Bioengineering,* 108,(10), 2320-2329.
VARDON, D. R., SHARMA, B. K., BLAZINA, G. V., RAJAGOPALAN, K. & STRATHMANN, T. J. 2012. Thermochemical Conversion of Raw and Defatted Algal Biomass Via Hydrothermal Liquefaction and Slow Pyrolysis. *Bioresource Technology,* 109, 178-187.
VARFOLOMEEV, S. D. & WASSERMAN, L. A. 2011. Microalgae as Source of Biofuel, Food, Fodder, and Medicines. *Applied Biochemistry and Microbiology,* 47,(9), 789-807.
VASUDEVAN, V., STRATTON, R. W., PEARLSON, M. N., JERSEY, G. R., BEYENE, A. G., WEISSMAN, J. C., RUBINO, M. & HILEMAN, J. I. 2012. Environmental Performance of Algal Biofuel Technology Options. *Environmental Science & Technology,* 46,(4), 2451-2459.
VELASQUEZ-ORTA, S. B., CURTIS, T. P. & LOGAN, B. E. 2009. Energy from Algae Using Microbial Fuel Cells. *Biotechnology and Bioengineering,* 103,(6), 1068-1076.
VELASQUEZ-ORTA, S. B., LEE, J. G. M. & HARVEY, A. 2012. Alkaline in Situ Transesterification of Chlorella Vulgaris. *Fuel,* 94,(0), 544-550.
VELDHUIS, M. J. W., FUHR, F., BOON, J. P. & TEN HALLERS-TJABBERS, C. 2006. Treatment of Ballast Water; How to Test a System with a Modular Concept? *Environmental Technology,* 27,(8), 909-921.
VENNARD, J. K. & STREET, R. L. 1976. Elementary Fluid Mechanics. 5th , SI version. ed. London: Wiley, 1976. .
VERMA, N. M., MEHROTRA, S., SHUKLA, A. & MISHRA, B. N. 2010. Prospective of Biodiesel Production Utilizing Microalgae as the Cell Factories: A Comprehensive Discussion. *African Journal of Biotechnology,* 9,(10), 1402-1411.
WAGENINGEN UNIVERSITY. 2011. *Research on Microalgae within Wageningen Ur* [Online]. Available: http://www.algae.wur.nl/UK/technologies/biorefinery [Accessed 25/01 2013].
WAITE, A. M., THOMPSON, P. A. & HARRISON, P. J. 1992. Does Energy Control the Sinking Rates of Marine Diatoms. *Limnology and Oceanography,* 37,(3), 468-477.
WALKER, D. 2009. Biofuels, Facts, Fantasy, and Feasibility. *Journal of Applied Phycology,* 21,(5), 509-517.
WALKER, D. A. 2010. Biofuels - for Better or Worse? *Annals of Applied Biology,* 156,(3), 319-327.
WANG, L. K., PEREIRA, N. C. & HUNG, T. T. 2009. *Biological Treatment Processes,* New York, Humana.
WANNINKHOF, R. 1992. Relationship between Wind-Speed and Gas-Exchange over the Ocean. *Journal of Geophysical Research-Oceans,* 97,(C5), 7373-7382.
WATER TREATMENT. 2010. *Waste Stabilization Ponds* [Online]. [Accessed 06/03/2012].
WEAST, R. C. (ed.) 1985. *Handbook of Chemistry and Physics,* Boca Raton Fl: CRC.

WEILAND, P. 2010. Biogas Production: Current State and Perspectives. *Applied Microbiology and Biotechnology,* 85,(4), 849-860.
WEISSMAN, J. C. & GOEBEL, R. P. 1987. Design and Analysis of Microalgal Open Pond Systems for the Purpose of Producing Fuels: A Subcontract Report.
WEISSMAN, J. C., TILLETT, D. M. & GOEBEL, R. P. 1989. Design and Operation of an Outdoor Microalgae Test Facility. Golden, Colorado: SERI.
WELLINGER, A. & LINDBERG, A. 2001. Biogas Upgrading and Utilisation - Iea Bioenergy, Task 24 - Energy from Biological Conversion of Organic Waste. *In:* IEA (ed.).
WEYER, K. M., BUSH, D. R., DARZINS, A. & WILLSON, B. D. 2010. Theoretical Maximum Algal Oil Production. *Bioenergy Research,* 3,(2), 204-213.
WIJFFELS, R. H. 2007. Potential of Sponges and Microalgae for Marine Biotechnology. . *Trends in Biotechnology,* 26,(1), 26-31.
WILEMAN, A., OZKAN, A. & BERBEROGLU, H. 2012. Rheological Properties of Algae Slurries for Minimizing Harvesting Energy Requirements in Biofuel Production. *Bioresource Technology,* 104, 432-439.
WILHELM, C. & JAKOB, T. 2011. From Photons to Biomass and Biofuels: Evaluation of Different Strategies for the Improvement of Algal Biotechnology Based on Comparative Energy Balances. *Applied microbiology and biotechnology,* 92,(5), 909-19.
WILKIE, A. C. 2005. Effect of Ambient Temperature on the Energy Balance of Anaerobic Digestion Plants. *Dairy Manure Conference.* Cornell University.
WILLIAMS, P. J. L. B. & LAURENS, L. M. L. 2010. Microalgae as Biodiesel & Biomass Feedstocks: Review & Analysis of the Biochemistry, Energetics & Economics. *Energy & Environmental Science,* 3,(5), 554-590.
WOOLEY, R. J. & PUTSCHE, V. 1996. Development of an Aspen Plus Physical Property Database for Biofuels Components. Golden, Colorado: NREL.
WORLD COMMISSION ON ENVIRONMENT AND DEVELOPMENT 1987. *Our Common Future,* Oxford, Oxford University Press.
WORLD ENERGY COUNCIL 2010 Survey of Energy Resources. London.
WU, B. X. 2012. Integration of Mixing, Heat Transfer, and Biochemical Reaction Kinetics in Anaerobic Methane Fermentation. *Biotechnology and Bioengineering,* 109,(11), 2864-2874.
XU, X., SONG, C., WINCEK, R. T., ANDRESEN, J. A., MILLER, B. G. & SCARONI, A. W. 2003. Separation of CO2 from Power Plant Flue Gas Using a Novel CO2 "Molecular Basket" Adsorbent. *Abstracts of Papers American Chemical Society,* 225,(1-2), FUEL 76.
YANG, C., JIA, L. S., CHEN, C. P., LIU, G. F. & FANG, W. P. 2011a. Bio-Oil from Hydro-Liquefaction of Dunaliella Sauna over Ni/Rehy Catalyst. *Bioresource Technology,* 102,(6), 4580-4584.
YANG, J., XU, M., ZHANG, X. Z., HU, Q. A., SOMMERFELD, M. & CHEN, Y. S. 2011b. Life-Cycle Analysis on Biodiesel Production from Microalgae: Water Footprint and Nutrients Balance. *Bioresource Technology,* 102,(1), 159-165.
YANG, Y. F., FENG, C. P., INAMORI, Y. & MAEKAWA, T. 2004. Analysis of Energy Conversion Characteristics in Liquefaction of Algae. *Resources, Conservation and Recycling,* 43,(1), 21-33.
ZAMALLOA, C., VULSTEKE, E., ALBRECHT, J. & VERSTRAETE, W. 2011. The Techno-Economic Potential of Renewable Energy through the Anaerobic Digestion of Microalgae. *Bioresource Technology,* 102,(2), 1149-1158.
ZEMKE, P. E., WOOD, B. D. & DYE, D. J. 2010. Considerations for the Maximum Production Rates of Triacylglycerol from Microalgae. *Biomass & Bioenergy,* 34,(1), 145-151.

ZHAO, B., MA, J., ZHAO, Q. & FREAR, C. 2012. Anaerobic Digestion of Algal Biomass Residues with Nutrient Recycle.
ZIMMERMAN, W. B., HEWAKANDAMBY, B. N., TESAR, V., BANDULASENA, H. C. H. & OMOTOWA, O. A. 2009. On the Design and Simulation of an Airlift Loop Bioreactor with Microbubble Generation by Fluidic Oscillation. *Food and Bioproducts Processing,* 87,(3), 215-227.

Appendices

Appendix 1 Files on accompanying CD

a. maxmin simple flow 1 fix vol ad.xlsx
b. oxygen relative outgassing.xlsx
c. whey ad energy inputs.xlsx
d. bacteria n and CO_2 .xlsx

Appendix 2 Related publications 2010 to 2013

Peer Review Journals

HEAVEN, S., MILLEDGE, J. & ZHANG, Y. 2011. Comments on 'Anaerobic Digestion of Microalgae as a Necessary Step to Make Microalgal Biodiesel Sustainable'. *Biotechnology Advances,* 29**,** (1), 164-7.

MILLEDGE, J. J. 2011. Commercial Applications of Microalgae other than as Biofuels: A Brief Review. *Reviews in Environmental Science and Biotechnology,* 10**,** (1), 31-4

MILLEDGE, J. J. & HEAVEN, S. 2011. Disc Stack Centrifugation Separation and Cell Disruption of Microalgae: A Technical Note. *Environment and Natural Resources Research,* 1**,** (1), 17-24.

MILLEDGE, J. J. & HEAVEN, S. 2013. A Review of the Harvesting of Micro-Algae for Biofuel Production. *Reviews in Environmental Science and Biotechnology,* 12**,** (2), 165-178.

Invited Contributions

MILLEDGE, J. J. 2010. The Challenge of Algal Fuel: Economic Processing of the Entire Algal Biomass. *Condensed Matter -Materials Engineering Newsletter,* 1, (6), 4-6.

MILLEDGE, J. J. 2010. The Potential Yield of Microalgal Oil. *Biofuels International* 4**,** (2), 44 45.

MILLEDGE, J. J. 2012. Microalgae – Commercial Potential for Fuel, Food and Feed. *Food Science & Technology,* 26**,** (1), 26-28.

Conference Presentations

MILLEDGE, J. J. 2013. Micro-Algal Biorefineries. *Towards Establishing Value Chains for Bioenergy.* Swakopmund, Namibia.

MILLEDGE, J. J. 2013. Horizontal biorefineries. *Sustainable Chemical Manufacture through Industrial Biotechnology*, Birmingham.

Conference Posters

Appendix 2

MILLEDGE, J. J. 2011. The Potential Energy Challenges of Micro-Algal Process Operations for Fuel Production *Algae for Renewable Energy*. University of Bath.
MILLEDGE, J. J. 2012. Micro-Algal Process Flow-Sheet Energy Balance Optimisation: Initial Software Evaluation. *Algal Biotechnology: Biofuels and Beyond.* UCL.