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***Biofuels from microalgae. Technological
aspects and economic analysis***

Biocarburanti da microalghe. Panoramica della tecnologia ed analisi economica

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Summary

This thesis explores the potential of producing biofuels from microalgae. The unsustainability of today's energy systems is the issue that originates the interest in the argument. In particular, transportation sector offers uncertain for the future, because traditional biofuels compete for land use with food production.

This work examines the feasibility of producing biofuels from microalgae with a deep technical exploration of all the different stages of production. Alternatives are proposed for the required treatments, with consideration for costs, possibility of scale-up and results achieved.

Concerning biofuels from microalgae, no large-scale facility exists: there is limited technical expertise and the production has been made only in laboratories. Here we analyze a production in large scale, assuming costs and revenues for twelve different algal productions. An economic analysis is made for each scenario, with the use of economic indicators (such as Net Present Value, Internal Rate of Return and Payback Period) in order to demonstrate the viability of the project.

System optimization is designed and the integration with CO₂-emitting industries, wastewater treatment facilities and cogeneration systems is considered to enhance the feasibility of the project.

Introduction

This thesis discusses biofuels from microalgae cultivation. Technological aspects are discussed with the aim of analyzing the process from an economical perspective.

An overview of biofuels production from microalgae is presented in chapter 1. Attention is given to the factors that motivate the study, such as the increase in crude oil costs, environmental concerns, and the limits of traditional biofuels that use land suitable for agriculture and food production.

The second chapter describes the characteristics and features of algae. Algae are classified for different strains, lipid contents and productivities. Then we treat algal physiology, with photosynthesis and light utilization and lipid synthesis and regulation aspects.

Chapter 3 discusses the algae cultivation, with particular attention to open pond systems and photobioreactors. These two different techniques are described, with consideration to their design, operational parameters, operating and plant costs and achievable productivity. Then the technologies are compared and also hybrid systems and alternatives are reported.

Chapters 4 and 5 are more technical and describe the downstream processes and the biofuels conversion techniques. The treated processes include: harvesting through sedimentation, flocculation and centrifugation; dewatering through filtration and drying; extraction of oil and other products; conversion technologies.

In the chapters 6 and 7 the biofuels production from algae is analyzed in depth. Chapter six contains an economic analysis of the technologies, with regards to open ponds. Different scenario and several economic aspects are considered, such as assumptions, potential costs and revenues, source of the used capital, interest and inflation rates, amortization and different economic indicators. Results of different scenarios are presented and commented.

Chapter 7 discusses how to optimize the system. Attention is given to: combined processes (the integration with water treatment facilities and with CO₂-emitting industries); obtainable co-products; combined heat and power cogeneration and to other important economical aspects as distribution and utilization of biofuels, intangibles and investments in the algae industry.

Chapter 8 presents our conclusions, with the achieved results and the future perspectives and technological developments.

CHAPTER 1

Biofuel from microalgae, an overview.

1. Biofuels from microalgae, some motivations.

Renewable energies are gaining everyday wider public and scientific attentions. Renewable energy sources such as biomass, hydro, wind, solar (both thermal and photovoltaic), geothermal and marine energy sources will play an important role in the world's future energy supply. Each alternative source of energy has its own advantages and disadvantages, including political, economical and technical issues. Renewable energy is a promising solution because it is clean and environmentally safe.

Today's energy system is unsustainable for different aspects. The development of renewable energies is driven by several factors. Some major reasons are:

- The energy availability from the non-renewable sources is limited, and beyond that, the exploration, the processing and the use of energy impose considerable impacts on the environment (Balat 2010).
- Renewables produce lower levels of greenhouse gases and other pollutants when compared to the fossil energy sources they replace (Dincer 2008). Today, over 80% of the energy we use comes from three fossil fuels: petroleum, coal and natural gas. About 98% of carbon emissions results from fossil fuels' combustion.
- Petroleum fuels have an uneven distribution in the world; for example, the Middle East has 63% of the global reserves and is the dominant supplier of petroleum (Hacisalihoglu et Al. 2009).

This energy system is unsustainable because of equity issues as well as environmental, economic, and geopolitical concerns that will have implications far into the future. Interestingly, the renewable energy resources are more evenly distributed than fossil or nuclear resources.

In this scenario, the situation is particularly acute in the transportation sector, where currently there are no relevant alternatives to fossil fuels. Finding clean and renewable energy sources, in order to substitute petroleum ranks as one of the most challenging problems facing mankind in the medium to long-term.

Biofuels are obtaining great consideration driven by aspects such as oil price spikes, the need for increase energy security and concern over greenhouse gas (GHG) emissions from fossil fuels. Although biofuels are still more expensive than fossil fuels, their production is increasing in countries around the world. Encouraged by policy measures and biofuels targets for transport, its global production is estimated to be over 35 billion liters (COM,

2006). In Europe the main alternative to petrol fuels is biodiesel, representing 82% of total European biofuels production (Bozbas, 2008) and is still growing in Europe, Brazil and United States, based on political and economic objectives. The U.S. Department of Agriculture (USDA) generated a strategy to help recharge the rural American economy with the goal to develop a successful market for biofuels. The U.S. renewable fuels mandate, part of the 2007 American energy law, requires the use of 21 billion gallons of advanced biofuels a year by 2022 and of 15 billion gallons of corn-based fuels (USDA 2010).

Since the biomasses that are used to produce biofuels may also be used for human consumption, the consequence is that the cost of both food and biofuel could increase. This situation can lead to prevent biofuels usage, even if they have advantages over traditional fuels. The available quantities of waste oils, animal fats and other non-food crops are not enough to match the today demands for biofuels. Additionally, biofuels need to have lower environmental impacts and ensure the same level of performance of existing fuels (Reinhardt et al. 2008).

A transition to “advanced” biofuels, such as microalgae, can contribute to a reduction in land requirements due to their presumed higher energy yields per hectare as well as to their non-requirement of agricultural land. There are some barriers to the development of advanced biofuel production. These barriers will be discussed more in depth and are mainly technological, economical and concerning supply, storage, safety and policy. Reducing them is one of the driving factors for the future research and development. For decades, microalgae have been studied and cultivated on a commercial base for the production of high value compounds for food, feed, cosmetics and pharmaceutical products. The need for alternative renewable energy feedstocks that are not in competition to food production has drawn the attention to microalgae. Microalgae can be cultivated in open or closed ponds or in photobioreactors. Production costs are uncertain and vary with the feedstocks available. Some researches calculate that with microalgae it is potentially achievable a biodiesel cost per barrel of only \$20 (Demirbas 2009). Despite all the claims and research dating from the early 1970s to date, none of the projected algae and oil yields has been achieved (Patil, 2005).

There is a very controversial debate about whether microalgae can provide short-term economic advantages in substitution of oil or not. Algae fuel can reach price parity with oil in 2017 if granted production tax credits, according to the head of the Algal Biomass Organization. The current cost of a barrel of algae biofuel ranges from \$140 to \$900 (Reuters, 2010).

This work focuses its attention on microalgae and how they can be used for biofuel production, with a primary consideration on biodiesel. Questions associated with production and processing of microalgae are considered in detail; not only those directly related with

biofuels production, but also the possibilities of combining it with pollution control, in particular with biological sequestration of greenhouse gases emissions, or wastewater treatment. The current status of biofuels production from microalgae, concerning their growth, harvest and processing is reviewed. Other potential applications and how to combine them with biofuels production are also described.

The aim of this research is to analyze the production of energy and biofuels with microalgae and to find out how feasible the process is, from an economic point of view.

2. Future prospects for the fuel sector.

The transportation sector is based on fossil fuels, which constitute the great majority of the energy consumption. Debates regarding the future of energy demand and supply have taken place worldwide. The main forces driving the oil transition are: high-energy prices due to increase in global energy demand, security of supply and climate change (Belkin, 2008; Greene, 2007). It is expected that with the development of new growing economies, such as India and China, the global consumption of energy will raise and lead to more environmental damage. Global primary energy demand is predicted to rise by 40% between 2007 and 2030 (IEA, 2010) putting additional pressure on the fossil fuel dependent countries. The relatively stable situation with crude oil prices changed drastically in the beginning of the 21st century. The price escalation to over \$140 per barrel in 2008 broke all records causing fears of the impact on world economy.

Oil dependency cost has been assessed by studies in which security of supply is regarded as one of the major issues (Maibach et al., 2007). Furthermore, it is well known that the reserves of fossil fuels are finite. The reduction of crude oil availability and difficulties in their extraction and processing, will eventually lead to an increase of fuel's cost.

The third driver of the oil transition is the increase in anthropogenic GHG concentrations, which very likely caused the increase of global average temperatures over the past 50 years (IPCC, 2007). Carbon dioxide is the main anthropogenic greenhouse gas; its concentration has risen from 250 ppm in a preindustrial era to present 379 ppm (IPCC, 2007). GHG contribute not only to global warming but also to other influences on the environment and human life.

Oceans absorb approximately one-third of the CO₂ emitted each year by human activities and as its levels increase in the atmosphere, the amount dissolved in oceans will also

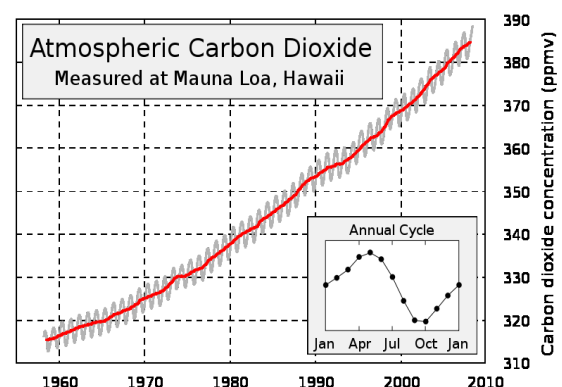


Fig. 1. Curve of the atmospheric CO₂ concentrations. (Global Warming Art, 2010)

increase turning the water pH gradually to more acidic. This pH change may cause the quick loss of coral reefs and of marine ecosystem biodiversity, with huge implications in ocean life and consequently in earth life (Ormerod, 2002).

Many countries and regions around the world established targets for CO₂ reduction in order to meet the sustainability goals agreed under the Kyoto Protocol. This shows how finding clean and renewable energy sources has associated issues connected with economic development and prosperity, quality of life, global stability, and requires from all stakeholders tough decisions and long term strategies.

Reducing the use of fossil fuels would considerably reduce the amount of carbon dioxide and other pollutants produced. This can be achieved by either using less energy altogether or by replacing fossil fuel with renewable fuels. Hence, the future trend is towards using alternative energy sources. Fortunately, technological developments are making the transition possible. Biofuels are one of the potential substitutes for fossil fuels in the road transportation sector. They are commonly considered to generate environmental (climate change mitigation), economic (rural development, trade deficit reduction) and political benefits (security of supply, energy independency). Studies conducted on the environmental impacts, however, show considerable differences (Quirin et al., 2004). The fear is that large-scale biofuels production accelerates climate change, causes deforestation, impoverishment and dispossession of local communities, biodiversity loss, water and soil degradation, loss of food sovereignty and security (Biofuelwatch, 2010). Therefore a comprehensive scientific study on the environmental risks and benefits is suggested.

The criticism is, however, aimed at first generation biofuels (also known as agro-biofuels).

2.1. Price of petroleum: future projections.

The price of petroleum as quoted in news generally refers to the spot price per barrel (159 liters). The price is highly dependent on both its location and its grade, determined by factors such as its specific gravity or API and its sulphur content. There are many different varieties and grades of crude oil and, in order to make referencing types of oil easier for sellers and buyers, benchmarks are used. Crude oil benchmarks, also known as oil markers, were first introduced in the mid 1980s. There are three primary benchmarks: WTI, Brent Blend and Dubai. The Energy Intelligence Group has identified 161 different oil markers in total (EIA 2006).

The demand for oil is highly dependent on global macroeconomic conditions. According to the International Energy Agency, high oil prices generally have a large negative impact on the global economic growth (IEA, 2004). The price of oil is highly variable. Like the price of all commodities, it is subject to major swings over time, particularly tied to the overall business cycle. When demand for a commodity like oil exceeds production capacity, the price will rise

quite sharply because both demand and supply are fairly inelastic in the short run. Users of oil have commitments and habits that determine their energy use and these take time to adjust. Instead, on the supply side, adding new capacity is time-consuming and expensive. The US Energy Information Agency (EIA) uses the imported refiner acquisition cost, the weighted average cost of all oil imported into the US, as its "world oil price". In December 2009, EIA released a forecast for the future trends of oil price (Table 1; Fig. 2). The forecast use 2008 dollars and predicts prices for both Imported Low-Sulfur Light Crude Oil (represented in blue in the tables and graphs) and Imported Crude Oil (represented in orange). The analysis covers a time span of 29 years, starting from 2007, until the year 2035.

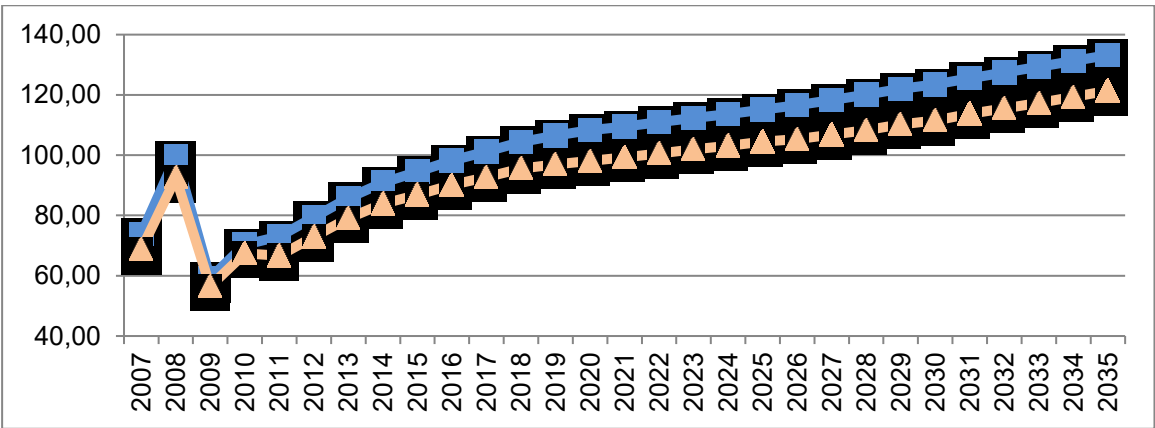


Fig. 2. Crude oil price forecast, 2008 dollars per barrel for Imported Low-Sulfur Light Crude Oil (blue) and Imported Crude Oil (orange).

Table 1. US Energy Information Agency forecast for the future trends of oil price.

| Crude Oil Prices (2008 dollars per barrel) | | | | | | |
|---|-------------|-------------|-------------|-------------|-------------|-------------|
| Year | 2007 | 2008 | 2009 | 2010 | 2011 | |
| Imported Low-Sulfur Crude Oil | 73,93 | 99,57 | 59,21 | 70,30 | 73,06 | |
| Imported Crude Oil | 68,69 | 92,61 | 56,49 | 67,40 | 66,63 | |
| 2012 | 2013 | 2014 | 2015 | 2016 | 2017 | 2018 |
| 79,41 | 85,74 | 90,91 | 94,52 | 98,23 | 101,23 | 106,47 |
| 72,94 | 79,09 | 83,84 | 86,88 | 90,10 | 92,72 | 95,59 |
| 2020 | 2021 | 2022 | 2023 | 2024 | 2025 | 2026 |
| 108,28 | 109,52 | 110,92 | 112,32 | 113,63 | 115,09 | 116,61 |
| 98,14 | 99,33 | 100,54 | 101,94 | 103,12 | 104,49 | 105,35 |
| 2028 | 2029 | 2030 | 2031 | 2032 | 2033 | 2034 |
| 120,13 | 122,04 | 123,50 | 125,56 | 127,43 | 129,29 | 131,25 |
| 108,28 | 110,26 | 111,49 | 113,70 | 115,49 | 117,35 | 119,35 |
| | | | | | | 2035 |
| | | | | | | 133,22 |
| | | | | | | 121,37 |

It is relevant to underline that the prices in the table and in the graph are in 2008 dollars. These prices can drastically change if we take in consideration the different value of money over time; for this reason we can suppose different inflation rates. Being impossible to know the future inflation rates, and being them variable, I hypothesized three different scenarios,

with an average inflation rate of 1.5%, 3% and 5% (Table 2). The case above, with 2008 dollars, represents a case of a flat inflation, with a rate equals to zero.

Table 2. US Energy Information Agency forecast for the future trends of oil price, with inflation rates.

| Price increases with an inflation rate of 1,5%; 3%; 5% | | | | | |
|---|-------------|-------------|-------------|-------------|-------------|
| | 2015 | 2020 | 2025 | 2030 | 2035 |
| price, 2008 \$ | 94,52 | 108,28 | 115,09 | 123,50 | 133,22 |
| | 86,88 | 98,14 | 104,49 | 111,49 | 121,37 |
| i.r.=1,5% | 104,9 | 129,46 | 148,24 | 171,36 | 199,14 |
| | 96,42 | 117,34 | 134,59 | 154,7 | 181,42 |
| i.r.=3% | 116,25 | 154,38 | 190,23 | 236,64 | 295,92 |
| | 106,85 | 139,92 | 172,71 | 213,63 | 269,6 |
| i.r.=5% | 133 | 194,46 | 263,79 | 361,27 | 497,37 |
| | 122,25 | 176,25 | 239,49 | 326,14 | 453,13 |
| increment, i.r.=1,5% | 10,99% | 19,56% | 28,80% | 38,75% | 49,48% |
| increment, i.r.=3% | 22,99% | 42,58% | 65,28% | 91,61% | 122,13% |
| increment, i.r.=5% | 40,72% | 79,59% | 129,20% | 192,52% | 273,35% |

If we consider the different value of money over time, the forecast for the crude oil costs shows a quite significant increase. An accurate prevision of the future average inflation rate is almost impossible, because it is influenced by an incredibly high number of factors. In addition, several factors have an impact on the oil price as well. For this reason the calculation serves just to provide different hypothetical scenarios. I calculated the predicted future price increments choosing an inflation rate of 3% (Table 3; Fig. 3). An inflation rate of 3% is rather high; despite that, I assume that it could well describe the future oil prices increase, adjusting the rather conservative EIA projections.

Table 3. US EIA forecast for the future trends of oil price, with an inflation rate of 3%.

| Price with an inflation rate of 3% | | | | | |
|---|-------------|-------------|-------------|-------------|-------------|
| | 2015 | 2020 | 2025 | 2030 | 2035 |
| | 116,25 | 154,38 | 190,23 | 236,64 | 295,92 |
| | 106,85 | 139,92 | 172,71 | 213,63 | 269,6 |
| Increment from 2010 prices | | | | | |
| | 55,87% | 107,00% | 155,07% | 217,30% | 296,78% |
| | 49,44% | 95,69% | 141,55% | 198,78% | 277,06% |

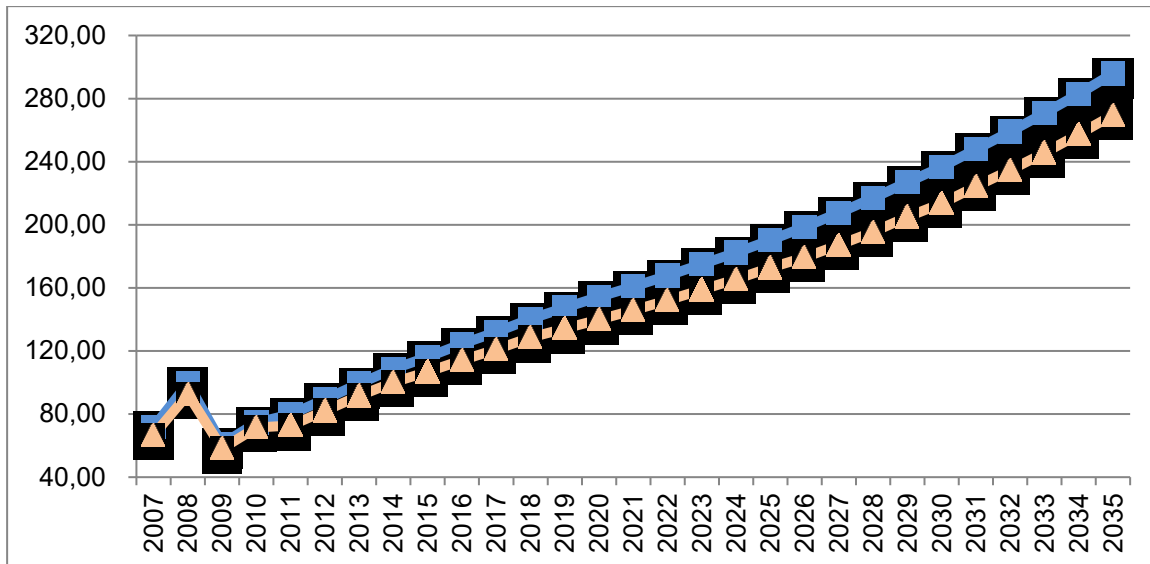


Fig. 3. Crude oil price forecast for Imported Low-Sulfur Light Crude Oil (blue) and Imported Crude Oil (orange) using an inflation rate of 3%.

Summarizing, the crude oil price could increase meaningfully. Projections show how the estimated increase is around 100% in ten years, and around 200% in twenty years. Furthermore, in the last years the price of oil has had some peaks and low points that can show its high variability. A brief history of the crude oil prices demonstrates how unstable the oil price could be. A recent \$17 low point was reached in January 1999, caused by increased oil production from Iraq, coincided with the Asian Financial Crisis, which reduced demand. Prices then increased rapidly: \$35 on September 2000, then a steadily increase, reaching \$50 by September 2004. Crude oil prices rose to a record high above \$60 in June 2005. Crude oil peaked at \$77 in July 2006. Oil broke through \$110 on March 12, 2008, \$135 on May 22, 2008, \$140 on June 26, 2008 and \$145 on July 3, 2008 (BBC, 2008). On July 11, 2008, oil prices rose to a new record of \$147.27 following concern over recent Iranian missile tests (BBC, 2008).

The drop in demand for oil contributes to the oil prices decline. On September 15, oil price fell below \$100 for the first time in seven months. Oil traded below \$70 on October 16, 2008. On December 21, 2008, oil was trading at \$34 a barrel, less than one fourth of the peak price reached four months earlier. By August 2009, prices returned to \$70 a barrel. EIA expects the price of West Texas Intermediate (WTI) crude oil to average about \$84 per barrel this winter (October 1, 2010 to March 31, 2011) (EIA, 2010).



Fig. 4. New York Mercantile Exchange prices for West Texas Intermediate 1996-2010 (Public Domain)

This price is approximately \$15 higher than the one projected by the same agency in December 2009. In conclusion, the very uncertain situation should be an incentive to investments in other fuels than crude oil. Even with the forecasts made by EIA, which are in my opinion rather conservative, high inflation or energy, financial or economic crisis could dramatically increase the crude oil prices.

3. Biofuels.

Biofuels are a wide range of combustible materials directly or indirectly derived from biomass, commonly produced from plants, animals and microorganisms but also from organic wastes. Investment into biofuels production capacity exceeded \$4 billion worldwide in 2007 and is growing (UNEP, 2009).

Biofuels production is expected to offer new opportunities to diversify income and fuel supply sources, to promote employment in rural areas, to develop long term replacement of fossil fuels, and to reduce GHG emissions, boosting the decarbonization of transportation fuels and increasing the security of energy supply. Most transportation fuels are liquids, because vehicles usually require high energy density. The fuels that are easiest to burn cleanly are typically liquids and gases. Thus liquids (and gases that can be stored in liquid form) meet the requirements of being both portable and clean burning. The term “biofuel” covers liquid fuels and various biogases.

The most common biofuels are biodiesel and bioethanol, which can replace diesel and gasoline, respectively, in today cars with little or none modifications of vehicle engines. They contribute to lower combustion emissions with respect to fossil fuels per equivalent power output (Delucchi, 2003). They can be produced using existing technologies and be distributed through the available distribution system. For this reasons biofuels are currently pursued as a fuel alternative that can be easily applied until other options harder to implement, such as hydrogen, will be available.

So far, biomass use constitutes only a negligible share of total global energy consumption. For example, first generation biofuels for transport provided only 0.3% of global final energy consumption in 2006 and 1.8% of total transport fuels in 2007 (UNEP, 2009).

Biofuels for transport are commonly addressed according to their current or future availability as first, second or third generation biofuels (IEA 2010). Second and third generation biofuels are also called “advanced” biofuels.

3.1. Biofuels classification.

First-generation biofuels are commercially produced using conventional technology. The basic feedstocks are seeds, grains, or whole plants from crops such as corn, sugar cane, rapeseed, wheat, sunflower seeds or oil palm. These plants were originally selected as food

or fodder and most are still mainly used to feed people. The most common first-generation biofuels are bioethanol (currently over 80% of liquid biofuels production by energy content), followed by biodiesel, vegetable oil and biogas. The bioethanol production methods used are enzyme digestion (to release sugars from stored starches), fermentation of the sugars, distillation and drying. The distillation process requires significant energy input for heat. The basic technology for biodiesel is the transesterification of oils and fats, in order to provide fatty acid methyl ester (FAME).

Second-generation biofuels can be produced from a variety of non-food sources. These include waste biomass, the stalks of wheat, corn stover, wood and special energy or biomass crops. Second-generation biofuels use biomass to liquid technology, by thermochemical conversion (mainly to produce biodiesel) or fermentation (mainly to produce cellulosic ethanol) (Inderwildi and King, 2009). Many second-generation biofuels are under development such as bio-hydrogen or bio-methanol.

Algae fuel is a biofuel from algae and is addressed as a third-generation biofuel (IEA 2010). Algae are feedstocks from aquatic cultivation, rich of triglycerides used to produce biodiesel. The processing technology is basically the same as for biodiesel from second-generation feedstocks. Other third-generation biofuels include alcohols like bio-propanol or bio-butanol, which due to lack of production experience are usually not considered to be relevant as fuels on the market before 2050 (IEA 2010).

4. Environmental aspects: advanced biofuels versus first-generation biofuels.

4.1. Carbon capture.

The Earth is continuously bombarded by energy from the Sun. The sunlight provides an average of 236 W for every square meter reaching the Earth's surface (Sension, 2007). Of this, most is absorbed directly and drives the ocean currents and our weather. A fraction (0.1–0.5%) is captured from biological systems via photosynthesis. Light energy captured in this way drives most living systems. The energy is used to capture atmospheric carbon dioxide, fixing it into the biomass of life on Earth.

Algae, like corn, soybeans, sugar cane, wood and other plants, use photosynthesis to convert solar energy into chemical energy. They store this energy in the form of oils, carbohydrates, and proteins through the absorption of CO₂ from the atmosphere. The plant oil can be converted to biofuels; hence biofuels are a form of solar energy. For these reasons, all biofuels share the great advantages of being both renewables and more environmental friendly than fossil fuels. The more efficient a particular plant is at converting

solar energy into chemical energy, the better it is from a biofuel perspective, and algae are among the most photosynthetically efficient plants on earth. The high photosynthetic efficiency of microalgae will have a wide treatment in chapter two.

One of the most interesting advantages of algal systems is that they allow the biological capture of CO₂ for generation of useful biomass. Carbon capture is a method of mitigating the contribution of fossil fuel emissions to global warming, based on capturing carbon dioxide from large point sources such as fossil fuel power plants, and storing it in such a way that it does not enter the atmosphere.

Presently most carbon capture and sequestration discussions are about geological storage of CO₂. The oil and gas industry has successfully injected CO₂ into reservoirs, although this has mainly been for increased yield of fossil hydrocarbon reserves and not for long-term storage. Even if this is proven safe, the biggest difficulty with this approach is the added cost of separation of the CO₂ from the emission streams (Packer, 2009).

In recent years, there has been increasing interest in greenhouse gas mitigation technologies. As a consequence, there has been renewed interest in microalgae mass culture and fuels production from the perspective of CO₂ utilization. Microalgae mass culture and fuels production from the perspective of CO₂ utilization is not a new concept, as Oswald and Golueke (1960) had previously emphasized the potential for microalgae systems to reduce and avoid CO₂ emissions and thus reduce the global warming. There are many reports on the potential of algal biomass to generate fuels and most of these are based on the premise of utilizing the CO₂ emitted from fossil fuel power stations or other industrial sources of CO₂ such as cement processing (Benemann, 1997; Hughes and Benemann, 1997; Vunjak et al., 2005; Greque de Morais and Costa, 2007; Ratledge and Cohen, 2008). There are also valuable studies in the literature exploring algal capture of either simulated or actual flue gas CO₂.

Microalgae have a rather unique attribute: they can utilize concentrated CO₂ for growth, rather than the air-levels of CO₂ used by higher plants. This allows the culture of microalgae utilizing power plant flue gases, probably the only method for the direct use of such CO₂ sources (Sheehan et Al. 1998). Of course, once the microalgae biomass is converted to, and used as fuel, this CO₂ is again released. However, an equivalent amount of fossil fuel is not burned, reducing overall CO₂ emissions. On the contrary, carbon capture for first-generation biofuels is mitigating only by recycling carbon from the atmosphere.

Algae's high CO₂ absorption can be demonstrated through different examples: it is estimated that of the 160 gigatonnes of CO₂ captured each year from the atmosphere, 60% occurs in surface waters to support the growth of diverse microalgae species. Fig. 5 is a simple demonstration of how biomass generation is dramatically stimulated by the presence of additional low levels of CO₂. The algae absorb the extra CO₂ present, capturing it as biomass through increased growth.

Using a combination of algal biomasses and fossil fuels combustion to generate energy in major power plants allows a further environmental benefit: a relevant CO₂ emissions saving is achievable. Jarvis shows a reduction of more than 70 % in the CO₂ emissions of a coal plant when fueled with a combination of coal and microalgae (Jarvis 2008).

The algal potential for capture of CO₂ from fossil power plants has as major benefit the fact that direct CO₂ capture processes are preferable to indirect ones.

As microalgae grow in aqueous environments, directly passing flue gases through this medium is a very efficient way of capturing the CO₂ in those streams (Benemann, 1997). The application of CO₂ directly to terrestrial crops via enclosures is likely to be prohibitively expensive though indirect stimulation of land species by flue gases is an alternative approach, which may be cost-effective despite being very much less direct and less efficient (Packer 2009). For these reasons many believe that microalgae are the only economic and environmental friendly route to biodiesel (Chisti, 2007; Schenk et al., 2008), though there is robust discussion about it (Ratlidge and Cohen, 2008).

4.2. Food against fuel.

The major concern about biofuels is linked to land use: restrictions or prior claims on use of land (food, energy, amenity use, housing, commerce, industry, leisure or designations as areas of natural beauty, special scientific interest, etc.), as well as the environmental and ecological effects of large areas of monoculture are the main non-technical barriers to biofuels development.

The potential market for biodiesel far surpasses the availability of plant oils not designated for other markets. For example, in Europe to fulfill a 10% fuel target from domestic

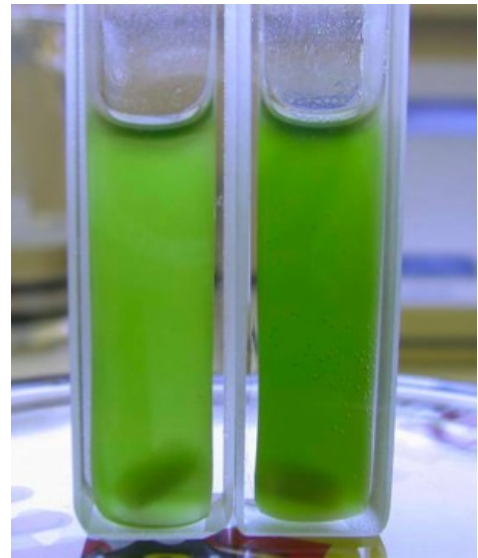


Fig. 5. Both the culture were bubbled with air; the left one with air containing 0.038 % of CO₂, the right one with an additional 1% of CO₂. The image shows the growth after 36 hours. This growth was greatly stimulated by the additional CO₂. The difference in biomass is confirmed by measurement of chlorophyll a (Chl a). The right culture contains twice the biomass of the left one: 31 µg/ml versus 16 µg/ml of Chl a (Packer, 2009).

production, the actual feedstocks supply is not enough to meet the current demand; the land requirements for biofuels production would be more than the potential available arable land for bio-energy crops (Scarlat et Al. 2008). The extensive plantation, pressure for land use change and increase of cultivated fields may lead to land competition and biodiversity loss, due to the cutting of existing forests and the utilization of ecological importance areas. Biodiesel may also be disadvantageous when replacing crops used for human consumption or if its feedstocks are cultivated in forests and other critical habitats with associated biological diversity.

The earth has a limited area of arable land, and grain reserves are limited as well. There is typically a 30 days supply of wheat in storage at any given time. When the supply is 33 days, it is considered a glut and prices drop; at 27 days the prices skyrocket. The small amount of grain (mainly sugarcane, maize and oilseeds) now being used for biofuel has had a domino effect, causing all grain prices to double (Gressel, 2008). This will soon trickle through the food chain and all food prices should soon double with littler grain available for emergency food aid. Moreover there is the bioethical issue of burning grain for fuel to run luxury automobiles when people are undernourished. The developed world is near top yields that are economically achievable. It would be possible to slightly increase yields by increasing pesticide and fertilizer use, but this would be environmentally undesirable and not overly cost effective. The long-term increases in yield to sustain human nutritional needs will have to come from the parts of the developing world that practices subsistence agriculture with yields lower than world averages. The doubled grain prices should allow these developing areas to produce competitively. The governments of developing country directly dealing with this turnabout could be a key for realizing this scenario.

Many proponents discuss about cultivating biofuels on marginal lands. Marginal lands are those with least economic value. However the environmental, ecological and economic impacts should be fully considered. The land may be less marginal after these other aspects are factored into the equation.

All these aspects gave the spin for the development of second-generation biofuels. Nevertheless concerns about them exist. They addressed the problem of land use, and are considered environmental friendly, because the source biomasses often are waste products. However, the available quantities of waste oils and animal fats are not enough to match the today request for biofuels. For this reason second-generation biofuels can substitute fossil fuels only in a small percentage.

Microalgae do not need fertile land to grow and have a very high yield. They can yield several times more energy per unit area than other biofuel crops. The United States Department of Energy estimates that if all the petroleum fuel in the United States is replaced with algae fuel, it would require 15,000 square miles (40,000 km²) (Hartman and Eviana,

2008). This is less than 14% of the area of corn harvested in the United States in 2000. However, these claims remain unrealized, commercially.

Yield and productivity are essential aspects concerning the production of biofuels from algae. Scientists, government agencies and companies are making efforts with the aim to increase them and to discover more details about the biological processes that regulate them.

In conclusion each biofuel technology has its own advantages and problems and, depending on the area of application, different options will be better suited. To address and solve the major problem of first-generation biofuels – the land use – is the goal of advanced biofuels. However, there are still several technical limits to the full development of third-generation biofuels technologies; limits that will be discussed in depth in the next chapters.

5. Historical perspective.

5.1. Early researches.

Historically, algae have been seen as a promising source of protein and have been actively cultured by man for centuries, mainly for food. German scientists first conceived growing algae in open ponds on a large scale, during World War II as a source of protein (Soeder, 1986). The first attempt to translate the biological requirements for algal growth into engineering specifications for a large scale plant was made at the Stanford Research Institute (1948–1950). Important results were achieved in the late 1940s and early 1950s. Nitrogen limitation was reported to significantly increase microalgal lipid storage. In 1949 Spoehr and Milner published detailed information on the effects of environmental conditions on algal composition and on the consequences after varying nitrogen supply on the lipid and chlorophyll content of *Chlorella* and some diatoms. Proposals to use algae as a means of producing energy started in the late 1950s when Meier (1955) and Oswald and Golueke (1960) suggested the utilization of the carbohydrate fraction of algal cells for the production of methane gas via anaerobic digestion.

Microalgae were first mass cultured on rooftop at MIT during the early 1950s, the first mention of algae biofuels appeared in the report of that project. Methane from algae was studied at U.C. Berkeley during the 1950s, the initial conceptual process and systems analysis were published in the 1960 (Benemann, 2008).

With the advent of the oil embargo in the early 1970s and the consequent energy shocks, a search for alternative energy sources set the stage for a research effort on algal lipids, microalgae biofuels, H₂ and methane in combination with wastewater treatment. Around the 1978 the American President Carter and his administration consolidated all federal energy activities under the support of the newly established U.S. Department of Energy (DOE). The Aquatic Species Program was a research effort intended to look at the use of aquatic plants

as source of energy. The program lasted from 1978 until 1996 and supported research primarily at DOE's National Renewable Energy Laboratory (NREL, formerly the Solar Energy Research Institute). Approximately \$25 million (Sheehan, 1998) was invested during the program. During the early years, the emphasis was on using algae to produce hydrogen, but the focus changed to liquid fuels (mostly biodiesel) in 1982.

5.2. The Aquatic Species Program.

The Aquatic Species Program represents one of the most comprehensive research efforts to date on fuels from microalgae. The program created the basis to all the following researches and studies about microalgae biofuels. Advances were made through algal strain isolation and characterization, studies of algal physiology and biochemistry, genetic engineering, process development, and demonstration-scale algal mass culture. Techno-economic analyses and resource assessments were also important aspects of the program. In 1998, a comprehensive overview of the project was completed (Sheehan et al., 1998). Some of the highlights are described briefly below.

The Aquatic Species Program researchers collected more than 3,000 strains of microalgae. The isolated were screened for their tolerance to variations in salinity, pH, and temperature, and also for their ability to produce neutral lipids. The collection was narrowed to the 300 most promising strains and afterwards further studies examined the ability of many strains to induce lipid accumulation under conditions of nutrient stress (Jarvis, 2008).

Under inducing conditions, some species in the collection were shown to accumulate as much as 60% of their dry weight in the form of lipid, primarily triacylglycerides (TAGs) (Chisti, 2007).

To discover the mechanisms that regulate the lipids accumulation was one of the major attempts by the ASP researchers. They discovered that the enzyme acetyl-CoA carboxylase (ACCase) catalyzes the first step in the biosynthesis of fatty acids used for TAG synthesis. ACCase activity was found to increase under the nutrient stress conditions (Roessler, 1988), suggesting that it may play a role in controlling lipid synthesis. With the advent of the first successful transformation of microalgae, it became possible to manipulate the expression of ACCase in an attempt to increase oil yields (Dunahay et al., 1995). These initial attempts at metabolic engineering identified a pathway to modify the gene encoding in the ACCase enzyme. However, no effect was seen on lipid production in these preliminary experiments (Sheehan et al., 1998). Additional studies focused on storage carbohydrate production, as biosynthesis of these compounds competes for fixed carbon units that might otherwise be used for lipid formation. The beginning of genetic engineering on algae is a great credit of the ASP, this is one of the subjects that gained the major attention from researchers and scientists involved in energy and biofuels researches in the last years.

The work of the Aquatic Species Program research included all the aspects involving the production of algae; biological productivity and various problematic process steps were addressed. Cost-effective methods of harvesting and dewatering algal biomass and lipid extraction, purification, and conversion to fuel are critical to successful commercialization of the technology. In particular, harvesting, extraction of oil droplets from the cells and purification of the oil were found to be highly energy and capital-intensive processes. Among various techniques, harvesting via flocculation was deemed particularly encouraging (Sheehan et al., 1998). Conversion of algal oils to biodiesel was successfully demonstrated in the Aquatic Species Program and shown to be one of the less challenging aspects of the technology. In addition, other biofuel process options were evaluated but no further fuel characterization, scale-up, or engine testing was carried out.

Under Aquatic Species Program subcontracts, demonstration-scale outdoor microalgal cultivation was conducted in California, Hawaii and New Mexico (Sheehan et al., 1998). The ASP concentrated his efforts on paddlewheel-mixed raceway ponds; the raceway design was based on the high rate pond system developed at University of California-Berkeley. The ponds in California and Hawaii were of small dimensions, from 48 to 200 m²; instead the facility in New Mexico utilized two 1,000 m² outdoor ponds. A major difference was on the ponds' depth, which was 60 cm in the Hawaiian experiment and diminished to 15-25 cm in New Mexico (Jarvis, 2008). The systems were successful in that long-term, stable production of algal biomass was demonstrated, and the efficiency of CO₂ utilization (bubbled through the algae culture) was shown to be more than 90% with careful pH control. Low nighttime and winter temperatures limited productivity in New Mexico, but overall biomass productivity averaged around 10 g/(m² day) with occasional periods approaching 50 g/(m² day). One serious problem encountered was that the desired starting strain was often outgrown by faster reproducing, but lower oil producing, strains from the wild (DOE, 2010).

Detailed techno-economic analyses underlined the necessity for very low-cost culture systems, such as unlined open ponds (Benemann and Oswald, 1996). The high operation and capital costs of photobioreactors were the reason that directed the ASP towards open pond technology. The techno-economic analyses showed clearly how biological productivity has the single largest influence on fuel cost. Different cost analyses led to differing conclusions on fuel cost, but even with optimistic assumptions about CO₂ credits and productivity improvements, estimated costs for unextracted algal oil were determined to range from \$59 - \$186 per barrel (Sheehan et al., 1998). It was concluded that algal biofuels would not be cost-competitive with petroleum, which was trading at less than \$20 for barrel in 1995.

Altogether, the Aquatic Species Program successfully demonstrated the feasibility of algal culture as source of oil and resulted in important advances in the technology. However, it

also became clear that in order to achieve an economically feasible process, significant barriers would need to be overcome. The overall conclusion from this review of 2 decades of DOE and ASP research and development in microalgal mass culture for biodiesel and other renewable fuels is that this technology still requires relatively long-term R&D for practical realization (Sheehan et al., 1998). In particular, the work highlighted the need to understand and optimize the biological mechanisms of algal lipid accumulation and to find creative, cost-effective solutions for the culture and process engineering challenges.

5.3. Other relevant programs.

The Aquatic Species Program was not the only algae related program sponsored by the U.S. DOE. From 1968 to 1990 the Marine Biomass Program, a research initiative to determine the technical and economic feasibility of macroalgae cultivation and conversion to fuels, was developed. The main aim was to substitute natural gas via anaerobic digestion. Similar to the findings of the Aquatic Species Program, researchers concluded that algal-derived natural gas would not be cost-competitive with fossil fuel gas (DOE, 2010).

In Japan, the Research for Innovative Technology of the Earth program (RITE) has carried out an extensive program for microalgal CO₂ utilization. The program was established in 1990 as a 10-year effort, carried out by approximately two dozen private companies, with some supporting work at various national laboratories and academic institutions (Sheehan et al., 1998).

Contrary to the U.S. Aquatic Species Program approach, the Japanese effort has focused on closed photobioreactors and on higher-value products. The main reason was that photobioreactors require less land area than open ponds, because of much higher productivities. The higher productivities were assumed to be possible by using optical fibers to diffuse light into the reactors and by greater control over environmental conditions (such as the ability to supply high CO₂ levels to the cultures). One major emphasis of the Japanese program has been on developing high-value coproducts, from animal feeds to antibiotics to specialty chemicals. According to Sheehan, the Japanese RITE Biological CO₂ Fixation Program, and other Japanese R&D activities have not significantly advanced the technology for biofuels production or CO₂ utilization, despite large investments. Another part of the project demonstrated actual increased productivity in optical fiber bioreactors. However, these devices were changed with more conventional, air-lift tubular reactors due to the complication and costs of the former. Benemann classified the RITE project on optical fiber bioreactors as a complete failure from an economic point of view, with a cost of photobioreactors around \$1000 for m² (Benemann, 2008).

5.4. Results after the ASP.

In the overview of the ASP project Sheenan et Al. foreshadowed the future revival of the biodiesel from algae field: “this report should be seen not as an ending, but as a beginning. When the time is right, we fully expect to see renewed interest in algae as a source of fuels and other chemicals. The highlights presented here should serve as a foundation for these future efforts.”

Since the end of DOE’s Aquatic Species Program in 1996, funding for algal research has come from several sources worldwide. A number of laboratories increasingly focused on algal biofuels research. Private investment in algal biofuels has been increasing at a dramatic rate over the last few years, significantly outpacing government funding.

Different aspects changed since 1996. First of all oil prices did not stay flat. Secondly environmental concerns about GHG increased. In addition the technology advanced in material science and new photobioreactor designs were developed. Last but not least the research in biotechnology reach amazing results, with advances in metabolic engineering, in genomics, bioengineering, etc. (Jarvis, 2008).

The U.S. DOE’s Biomass Program organized the National Algal Biofuels Technology Roadmap Workshop in December, 2008. The workshop was a clear clue of the renewed interest in the microalgae. Recently, the U.S. Energy Secretary Steven Chu announced \$80 million in government funding for biofuels research and development, the great majority are for algae research (Cnet, 2011). A number of R&D companies is worldwide competing in bringing the technology to the market and potential of algae in terms of biomass production has become the focus of a lively debate recently (Kovacevic and Wesseler, 2010).

According to the head of the Algal Biomass Organization, Mary Rosenthal, the technology is mature and is going through the same nascent issues of any emerging industry, trying to emerge from lab to pilot and than from pilot to scale. Currently, more than 100 companies worldwide are at work to bring algae to market (Reuters, 2010).

CHAPTER 2

Characteristics and Features of Algae.

1. Algae classification.

Algae are a diverse mix of organisms that have been unified based on their ability to carry out photosynthesis and live in aquatic habitats. Algae can be single or multi-cellular and prokaryotic or eukaryotic. Algae can be either freshwater or marine; some grow optimally at intermediate saline levels and some in hypersaline conditions.

Algae include macroalgae, or seaweeds, and microalgae; both are eukaryotic organisms and their cells display a high degree of internal organization, including a membrane-bound nucleus containing the genetic material and several other internal parts, surrounded by membranes as well. Algae include cyanobacteria also known as blue-green algae, which are prokaryotic (those cells that lack a distinct nucleus). Often the cyanobacteria are referred to be part of microalgae. Algae are the most diverse organisms in the world (Mayfield, 2008) and a very high number of different species exist. More than 300000 species are estimated, whose diversity is much greater than that of land plants (Scott et Al., 2010). According to Benemann there are 30000 described species of microalgae and this number is estimated to be less than 10 % of the existing species; in this case just the microalgae species would be more than 300000. Microalgae are classified in 11 divisions, divided into 29 classes, versus the two division and twelve classes of vascular plants (Benemann, 2008). However, algae degree of specialization or differentiation of cell types is much less than for terrestrial vascular plants. All algae contain proteins, carbohydrates, lipids and nucleic acids in varying proportions.

Microalgae are categorized into four main groups: diatoms (Bacillariophyceae), green algae (Chlorophyceae), blue-green algae (Cyanophyceae) and golden algae (Chrysophyceae). As the name suggests these are microscopic algae and many are unicellular. Like higher plants, they produce storage lipids in the form of triacylglycerol lipid molecules (TAGs) as energy storage molecules. Many species exhibit rapid growth and high productivity and many microalgal species can be induced to accumulate substantial quantities of lipids, often greater than 60% of their dry biomass (Sheenan et Al., 1998). Microalgae assume particular relevance, because they can be used for bioenergy generation; in particular TAGs can be easily transesterified to biodiesel. Among microalgae, green algae contain complex long-chain sugars (polysaccharides) in their cell walls. These carbohydrate cell walls account for a large proportion of the carbon contained in these organisms, though many species contain quite high levels of various lipids. Diatoms probably represent the largest group of biomass

producers on earth. It is estimated that more than 100,000 species exist. They have silicate cell walls and have been of considerable interest in the area because they can accumulate very high levels of lipid.

Macroalgae are multicellular plants growing in salt or fresh water. Depending on their pigmentation, they are classified into three broad groups: brown seaweed (Phaeophyceae); red seaweed (Rhodophyceae) and green seaweed (Chlorophyceae). Even if macroalgae can accumulate lipids as microalgae, their lipid content often is considerably lower. Nevertheless they are high in carbohydrates, which can be converted to various fuels (DOE, 2010). In this work we will mainly consider microalgae, due to better properties that make them very interesting for the production of biofuels. According to Mayfield, the reasons that make algae interesting as biofuels platform are scalability, sustainability, the achievability of superior fuels and potential low cost (Mayfield, 2008).

2. Algae lipid content and productivities.

Microalgae can be used for bioenergy generation (biodiesel, biomethane, biohydrogen, etc.). The technology can be associated with CO₂ mitigation and wastewater treatment. Thanks to the Aquatic Species Program we have learnt that many microalgae can accumulate neutral lipids and that diatoms and green algae are the most promising groups. Moreover a perfect strain does not exist and several factors, mainly climate and water type, influence the strain choice (Jarvis, 2008).

Algal biofuels have gained high interests, however nowadays macro and microalgae are mainly used for food, in animal feed, in feed for aquaculture and as bio-fertilizer. Biomass from microalgae can have multi-faceted health promoting effects; it is dried and marketed in the human health food market in form of powders or pressed in the form of tablets. Aquatic biomass could also be used as raw material for co-firing to produce electricity, for example in combination with coal in traditional power plants. As presented in the introduction, microalgae appear to be the only source of renewable biodiesel that is capable of meeting the global demand for transport fuels. This fact will probably drive their future uses and increase their production.

Microalgae naturally accumulate lipid during their growth. Many species can be induced to accumulate substantial quantities of lipids thus achieving a high oil yield. The average lipid content varies between 1 and 70%, however laboratory tests have found species that under certain conditions can reach 90% of dry weight (Li et Al. 2008; Spolaore et Al. 2006; Christi 2007). Lipid content and biomass productivities of different marine and freshwater microalgae species are reported in Table 4, showing significant differences between the various species. A critical concept is that high oil content does not mean high oil productivity. As shown in Table 4, high oil content is associated with low productivities. Most common

algae (*Chlorella*, *Cryptocodinium*, *Cylindrotheca*, *Dunaliella*, *Isochrysis*, *Nannochloris*, *Nannochloropsis*, *Neochloris*, *Nitzschia*, *Phaeodactylum*, *Porphyridium*, *Schizochytrium*, *Tetraselmis*) have oil levels between 20 and 50% but higher productivities can be reached (Mata et Al., 2010).

Table 4. Lipid content and productivity of different microalgae species (Mata et Al., 2010).

| Marine and freshwater microalgae species | Lipid content (% dry weight biomass) | Lipid productivity (mg/L/day) | Volumetric productivity of biomass (g/L/day) | Areal productivity of biomass (g/m ² /day) |
|--|--------------------------------------|-------------------------------|--|---|
| <i>Ankistrodesmus</i> sp. | 24.0–31.0 | – | – | 11.5–17.4 |
| <i>Botryococcus braunii</i> | 25.0–75.0 | – | 0.02 | 3.0 |
| <i>Chaetoceros muelleri</i> | 33.6 | 21.8 | 0.07 | – |
| <i>Chaetoceros calcitrans</i> | 14.6–16.4/39.8 | 17.6 | 0.04 | – |
| <i>Chlorella emersonii</i> | 25.0–63.0 | 10.3–50.0 | 0.036–0.041 | 0.91–0.97 |
| <i>Chlorella protothecoides</i> | 14.6–57.8 | 1214 | 2.00–7.70 | – |
| <i>Chlorella sorokiniana</i> | 19.0–22.0 | 44.7 | 0.23–1.47 | – |
| <i>Chlorella vulgaris</i> | 5.0–58.0 | 11.2–40.0 | 0.02–0.20 | 0.57–0.95 |
| <i>Chlorella</i> sp. | 10.0–48.0 | 42.1 | 0.02–2.5 | 1.61–16.47/25 |
| <i>Chlorella pyrenoidosa</i> | 2.0 | – | 2.90–3.64 | 72.5/130 |
| <i>Chlorella</i> | 18.0–57.0 | 18.7 | – | 3.50–13.90 |
| <i>Chlorococcum</i> sp. | 19.3 | 53.7 | 0.28 | – |
| <i>Cryptocodinium cohnii</i> | 20.0–51.1 | – | 10 | – |
| <i>Dunaliella salina</i> | 6.0–25.0 | 116.0 | 0.22–0.34 | 1.6–3.5/20–38 |
| <i>Dunaliella primolecta</i> | 23.1 | – | 0.09 | 14 |
| <i>Dunaliella tertiolecta</i> | 16.7–71.0 | – | 0.12 | – |
| <i>Dunaliella</i> sp. | 17.5–67.0 | 33.5 | – | – |
| <i>Ellipsoidon</i> sp. | 27.4 | 47.3 | 0.17 | – |
| <i>Euglena gracilis</i> | 14.0–20.0 | – | 7.70 | – |
| <i>Haematococcus pluvialis</i> | 25.0 | – | 0.05–0.06 | 10.2–36.4 |
| <i>Isochrysis galbana</i> | 7.0–40.0 | – | 0.32–1.60 | – |
| <i>Isochrysis</i> sp. | 7.1–33 | 37.8 | 0.08–0.17 | – |
| <i>Monodus subterraneus</i> | 16.0 | 30.4 | 0.19 | – |
| <i>Monallanthus salina</i> | 20.0–22.0 | – | 0.08 | 12 |
| <i>Nannochloris</i> sp. | 20.0–56.0 | 60.9–76.5 | 0.17–0.51 | – |
| <i>Nannochloropsis oculata</i> | 22.7–29.7 | 84.0–142.0 | 0.37–0.48 | – |
| <i>Nannochloropsis</i> sp. | 12.0–53.0 | 37.6–90.0 | 0.17–1.43 | 1.9–5.3 |
| <i>Neochloris oleoabundans</i> | 29.0–65.0 | 90.0–134.0 | – | – |
| <i>Nitzschia</i> sp. | 16.0–47.0 | – | – | 8.8–21.6 |
| <i>Oocystis pusilla</i> | 10.5 | – | – | 40.6–45.8 |
| <i>Pavlova salina</i> | 30.9 | 49.4 | 0.16 | – |
| <i>Pavlova lutheri</i> | 35.5 | 40.2 | 0.14 | – |
| <i>Phaeodactylum tricornutum</i> | 18.0–57.0 | 44.8 | 0.003–1.9 | 2.4–21 |
| <i>Porphyridium cruentum</i> | 9.0–18.8/60.7 | 34.8 | 0.36–1.50 | 25 |
| <i>Scenedesmus obliquus</i> | 11.0–55.0 | – | 0.004–0.74 | – |
| <i>Scenedesmus quadricauda</i> | 1.9–18.4 | 35.1 | 0.19 | – |
| <i>Scenedesmus</i> sp. | 19.6–21.1 | 40.8–53.9 | 0.03–0.26 | 2.43–13.52 |
| <i>Skeletonema</i> sp. | 13.3–31.8 | 27.3 | 0.09 | – |
| <i>Skeletonema costatum</i> | 13.5–51.3 | 17.4 | 0.08 | – |
| <i>Spirulina platensis</i> | 4.0–16.6 | – | 0.06–4.3 | 1.5–14.5/24–51 |
| <i>Spirulina maxima</i> | 4.0–9.0 | – | 0.21–0.25 | 25 |
| <i>Thalassiosira pseudonana</i> | 20.6 | 17.4 | 0.08 | – |
| <i>Tetraselmis suecica</i> | 8.5–23.0 | 27.0–36.4 | 0.12–0.32 | 19 |
| <i>Tetraselmis</i> sp. | 12.6–14.7 | 43.4 | 0.30 | – |

As several species appear to be efficient and productive, the selection of the most adequate strains needs to take into account other parameters, such as resistance to other species invasion, the ability to develop using the nutrients available, the ability to growth under specific environmental conditions, etc. *Chlorella* seems to be a good option for biodiesel production, also because it was deeply studied during the ASP and it is already successfully mass cultivated for nutrient purposes.

The composition of fatty acids of the different microalgae species is another relevant factor. These are saturated and unsaturated fatty acids with 12–22 carbon atoms and they have significant consequences on the characteristics of biodiesel produced (Mata et Al, 2010). Fatty acids can constitute up to 40% of the algae overall mass (Becker, 1994). The composition of fatty acids can be influenced from disparate aspects, both nutritional and

environmental. In particular it is possible to alter the accumulation and composition acting on cultivation conditions and growth phases. For example it is acknowledged that nitrogen deficiency and salt stress enhance the accumulation of long fatty acid chains (Mata et Al, 2010).

Both the lipid content and the productivity of a strain drive the oil yield, which can be considered the most significant distinguishing characteristic of an algal species cultivated with the target of biofuels production. The microalgae oil yield is generally much greater than other vegetable oil crops. The yield is strain-dependent and according to different authors, for some strains it is over 200 times the yield from the best-performing vegetable oil plant, using the same land size (Sheenan et Al., 1998; Christi 2007). Table 5 compares the biodiesel production efficiencies and land use of microalgae and other vegetable oil crops. Microalgae are the fastest-growing photosynthesizing organisms; some strains can complete an entire growing cycle in few days (Sheenan et Al., 1998). This is the reason of the significant variations in the overall biomass productivity, resulting oil yield and biodiesel productivity between microalgae and other plants. Indeed analyzing the table we can see that the oil contents between seed plants and microalgae are similar.

Table 5. Comparison of microalgae with other biofuels feedstocks (Mata et Al., 2010).

| Plant source | Seed oil content (% oil by wt in biomass) | Oil yield (L oil/ha year) | Land use (m ² year/kg biodiesel) | Biodiesel productivity (kg biodiesel/ha year) |
|---|--|------------------------------|--|--|
| Corn/Maize (<i>Zea mays</i> L.) | 44 | 172 | 66 | 152 |
| Hemp (<i>Cannabis sativa</i> L.) | 33 | 363 | 31 | 321 |
| Soybean (<i>Glycine max</i> L.) | 18 | 636 | 18 | 562 |
| Jatropha (<i>Jatropha curcas</i> L.) | 28 | 741 | 15 | 656 |
| Camelina (<i>Camelina sativa</i> L.) | 42 | 915 | 12 | 809 |
| Canola/Rapeseed (<i>Brassica napus</i> L.) | 41 | 974 | 12 | 862 |
| Sunflower (<i>Helianthus annuus</i> L.) | 40 | 1070 | 11 | 946 |
| Castor (<i>Ricinus communis</i>) | 48 | 1307 | 9 | 1156 |
| Palm oil (<i>Elaeis guineensis</i>) | 36 | 5366 | 2 | 4747 |
| Microalgae (low oil content) | 30 | 58,700 | 0.2 | 51,927 |
| Microalgae (medium oil content) | 50 | 97,800 | 0.1 | 86,515 |
| Microalgae (high oil content) | 70 | 136,900 | 0.1 | 121,104 |

We have seen that diatoms and green algae are the most promising algal groups. However, algal specie that can be said to be the best in terms of biofuels production does not exist among diatoms and greens. We have underlined the huge diversity among algae species; the type and strain of algae being cultivated will ultimately affect every step of the algae to biofuels supply chain (DOE, 2010). Consequently the choice of algal strain is another critical aspect and it has to take under consideration a wide range of factors, from the climate and growth conditions to the final product that is desired.

3. Strain Isolation, Screening and Selection.

The large number of different algal strains makes algae isolation and screening an important challenge. The goals are to identify and maintain promising algal specimens for cultivation.

Because the technology is not yet developed to a mass scale, the cultivation mechanisms are not fully understood. For this reason, new strains should be isolated from a wide variety of environments to provide the largest range in metabolic versatility possible. Screening different natural habitats, with the broadest coverage of environments should lead to successful results.

New strains can be isolated from natural habitats using traditional cultivation techniques, such as enrichment cultures. However, this solution often is very time consuming; some algal strains take weeks to months to be isolated by traditional methods. New isolation techniques have proven to be extremely useful. An example is the fluorescence-activated cell sorting (FACS) (Sieracki et al., 2004). Strain identification can also be based on molecular methods such as rRNA sequence comparison, in order to use morphological similarities in the algae. Using infrared light to sort out the best oil-producing algae is new promising technology. NREL uses near infrared spectroscopy to determine the oil content in the algae in a matter of minutes (NREL, 2010). Furthermore, this technology is better than the solvent extraction method, commonly used for the lipid content determination, for the lower amount of chemicals required. The principle that drives the process is that different molecules in the algal biomass reflect or absorb the light in a different way. In this way the oil content of that particular strain of algae is estimated. Furthermore the technique is non-destructive to the cell. Future plans are to apply these infrared methods to growing cultures to see if researchers can do real-time monitoring and determine when a culture is ready to be harvested (NREL, 2010).

According to DOE, an ideal screen would cover three major areas: growth physiology, metabolite production, and strain robustness. The term growth physiology encompasses a number of parameters that require significant experimental effort, such as maximum specific growth rate, maximum cell density, tolerance to environmental variables (temperature, pH, salinity, oxygen levels, CO₂ levels), and nutrient requirements (DOE, 2010). Screening for metabolite production is linked with the cellular composition of proteins, lipids, and carbohydrates. Strain robustness is a critical detail, especially considering the results obtained during the ASP, when the strains tested in the laboratory often performed differently in outdoor mass cultures (Sheenan et Al., 1998).

Concerning the problem of identifying strains with the desired traits for biofuel production, there are three recommendations. First of all one strain would unlikely have everything we want and grow everywhere we need. Secondly, a potential strain need to easily adapt itself to genetic modifications, in order to produce high level of desired molecules and fit harvest and fuel recovery requirements (Mayfield, 2008). The third consideration is that the strain's choice needs to consider what the desired final product is. Producing biodiesel, biomass for co-firing in power plants, human high-protein nutrition products or animal feed

biomass require algae with different features. Moreover different algae and different outputs require different processes, so the entire supply chain is affected. As example, if the desired product is volatile, such as ethanol, a strain that is capable to secrete fuel precursors is appealing, because it could reduce or skip the cell harvesting step (DOE, 2010; Algenol Biofuels, 2011). The technology needed is influenced as well, because only a closed cultivation system, such as a photobioreactors one, permits the condensation of the ethanol and water vapors in atmosphere and their collection.

According to DOE, in relation to biofuels there are two types of algal model systems to consider studying: species or strains amenable to providing information on basic cellular processes regarding the synthesis of fuel precursors and species or strains with characteristics useful for large-scale growth. Species with sequenced genomes and transgenic capabilities are the most amenable to investigating cellular processes since the basic tools are in place (DOE, 2010).

There are several useful algal characteristics to consider while selecting strains. We have already discussed about the algal lipid content and composition; culture stability and rapid growth are other key features to low cost and successful production of biofuels. The ability to compete with contaminating strains is another major aspect. It can be positively influenced by rapid growth and by the resistance to predators and viruses. Furthermore, the ability to grow in continuous culture where a high cell density exists may help a strain to be the culture dominant, while at the same time reducing the amount of water to be processed daily. Heterotrophic growth capabilities (when carbon is not fixed but used for growth) may be another attractive attributes of algal strains. Addition of supplemental carbon can result in increased lipid accumulation (Xu et al. 2006). Also there are factors that could reduce the costs of harvest. The ability to flocculate without addition of chemical flocculating agents is a positive algal attribute, as long as it could be controlled to avoid settling in the cultivation system (DOE, 2010).

4. Algal Physiology.

Algae are photosynthetic organisms that grow using CO₂, nutrients and water. They harvest solar energy and use it to convert CO₂ and nutrients to O₂ and organic macromolecules such as carbohydrates proteins and lipids. We have seen that some microalgae accumulate lipids such as triacylglycerols as their main carbon storage compounds. Certain microalgal species can naturally accumulate large amounts of TAGs, and exhibit photosynthetic efficiency and lipid production at least an order of magnitude greater than terrestrial crop plants as seen in table 5. Stress conditions such as high light or nutrient starvation influence and increase the lipid growth. We have also underlined as lipids and carbohydrates are potential biofuels

precursors. Achieving advanced biofuels production is possible only if we understand the metabolic pathways and processes that generate lipids and carbohydrates.

4.1. Photosynthesis and Light Utilization.

Algal productivity is crucially influenced by photosynthesis efficiency; it affects growth rate, biomass production and, potentially, the percent of biomass that is the desired fuel precursor. The light conditions deeply influence the algae cultivation: absence of light, too bright conditions or variations in lighting intensity could be causes of algal stress. Regarding light utilization, it is possible to research algal photosystems mechanisms; acting in the field could increase the algal yield and productivity. The majority of light that falls on a photosynthetic algal culture is not utilized.

In laboratory scale good light utilization can be achieved, but, at larger scale with high cell density cultures, cells nearer to the light source tend to absorb all the incoming light, preventing it from reaching more distant cells (Christi, 2007). Light is a source of energy for plants and algae. However, only a fraction of light can be effectively used: when exposed to high light, algal photosystems have built-in strategies to prevent the over-absorption of light energy. A fraction up to 95% of incident light is dissipated as heat and could be considered wasted; the process prevents oxidative damages (Bassi, 2008). Heat dissipation is the main photoprotection system. Even with this, under certain light regimes, photoinhibition or the reduction of photosynthesis due to light damage can still occur. With photoinhibition, the algal photosystems are damaged and the biomass yield becomes lower. Too long highlight exposure can create permanent damages and end the culture (Bassi, 2008).

The chlorophyll antenna or Lhc (Light harvesting complexes) regulate the light absorption. Lhc are proteins with a double role. During good light regimes Lhc increase the light absorption efficiency and transfer the captured energy to the photosystems. With higher incoming light the antenna proteins activate heat dissipation photoprotection mechanisms, and the majority of energy is wasted. Fig. 6 shows the light saturation curve, which describes the connection between light intensity and photosynthesis efficiency.

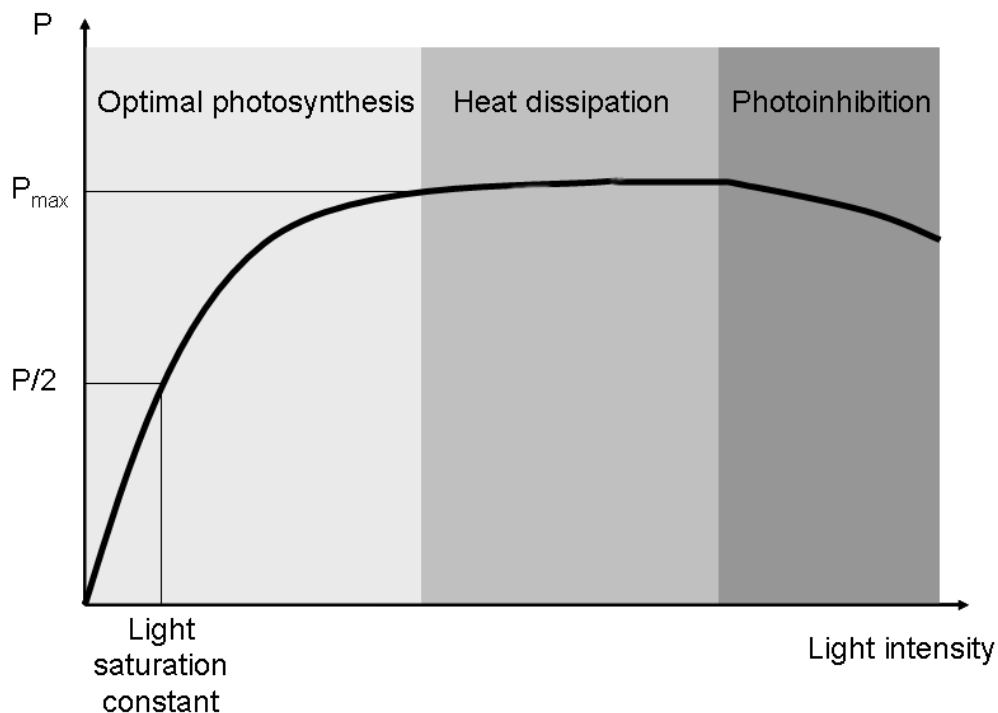


Fig. 6. Light saturation curve. Photosynthesis / Absorbed energy (P) increases linearly with the light intensity until saturation. P_{max} is the maximum photosynthetic rate. Upon saturation of photosynthesis, excess absorbed energy is dissipated as heat and if light further increases beyond the capacity of photoprotective mechanisms, photosynthetic rate decreases because of photoinhibition mechanisms (Bassi, Formighieri, 2010).

Efficiency increase is required in order to realize successfully economic biofuels from microalgae. The wasted light dissipated in heat influences the process efficiency. Bioengineering is working in an effort to overcome this barrier. It was shown that reducing the size of the chlorophyll antenna could increase the efficiency of light utilization (Polle et Al., 2002). As consequence the algae on the culture's surface have an inferior light absorption and there is a deeper light penetration in the culture. The better light distribution will activate photosynthesis reactions also in the deeper algae. This genetic mutation presents an enormous potential impact on the plants productivity. Estimates evaluate an increase of the biomass yield by a factor of 3 or 4 for cultures with reduced antenna mutants (Bassi, R., 2008; Polle et Al., 2002).

The use of reduced antenna mutants assumes a vast interest for photobioreactors systems. A patchy light distribution creates convective fluxes that mix the microalgae; this is marked in particular photobioreactors designs such as the tubular one. Rapid uncontrolled variations could increase algal stress and affect productivity. Microalgal daily theoretical photosynthetic productivity in full lightening conditions is of 75 g dry weight for m^2 . Experiments with pilot photobioreactors have revealed a lower productivity in wild type microalgae, around to 20-30 $g/(m^2 \text{ day})$ (Lee, 1997). The use of mutant algae could increase the productivity with a better

control over stress conditions (Bassi, Formighieri, 2010). The light penetration in a tubular photobioreactor with wild type and mutant microalgae is represented in Fig. 7.

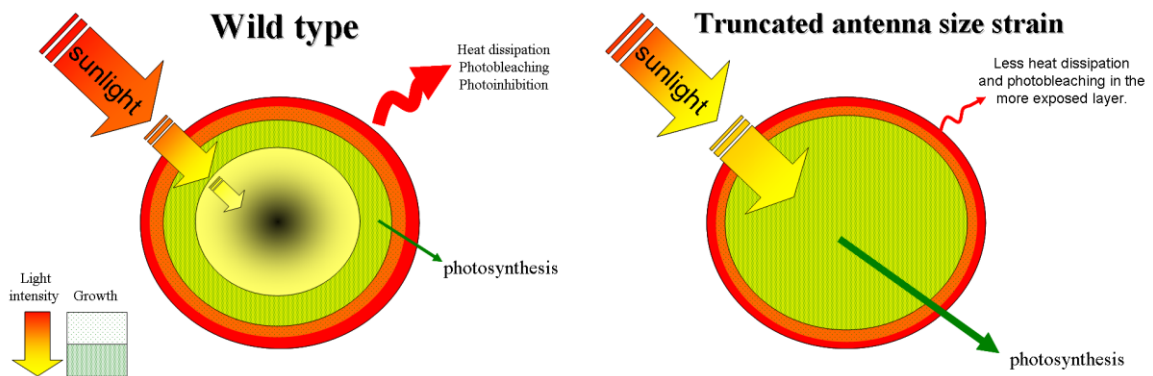


Fig. 7. Schematic representation of a tubular photobioreactor transversal section. In wild-type (left) most of light is absorbed by cells in surface layers. Excess light is transformed into heat by physiologic dissipation mechanisms. An algal strain with truncated antenna (right) is less efficient in absorbing light, thus allowing penetration of the irradiance deeper into the culture. As a consequence, a greater number of cells is photosynthetically active and accumulates biomass and a smaller fraction of the incident energy is dissipated into heat due to the lower number of photons intercepted by each photosystem (Bassi, Formighieri, 2010).

However, since antenna proteins, besides light harvesting, have an important role in photoprotection, it is essential that the reduction in the antenna system does not compromise photoprotection capacity of the strain in use. Cells without antenna proteins have a larger photoinhibition sensibility and do not survive if exposed to full solar light (Bassi, Formighieri, 2010). For this reason the choice of the best mutant specie requires accurate analysis and studies.

4.2. Lipid synthesis and regulation.

Knowing how and when carbon is partitioned into lipids and carbohydrates could be very useful for biofuels strain development and designing cultivation strategies. According to DOE, research on how algal cells control the flux and partitioning of photosynthetically fixed carbon into carbohydrates, proteins and lipids is critically needed. Here we report the main research results in the topic, with particular attention for the lipid synthesis and regulation.

For a wider coverage of the argument please consult the DOE algal biofuels roadmap (DOE, 2010).

Carbon partitioning in algae is poorly understood. We know that starvation and stress conditions can increase the lipids formation. Algae use different kind of macromolecules for carbon storage: table 6 shows the dissimilarities of chemical composition between different species of microalgae.

The composition and structure of the algal cell walls is another important consideration.

These structures are diverse strain by strain. Cell walls can be an important storage of carbohydrates. It can also be a technical barrier, for example, when trying to access DNA for genetic manipulations, or efficiently extracting biofuel precursors from cells in mass culture (DOE, 2010). Further, a link between starch and lipid metabolism has been established and it is possible that TAG and starch could be inter-convertible, with important potential implications for biofuel production (DOE, 2010).

The lipid yields obtained from algal mass culture efforts is lower than the high values observed in the laboratory, as proved during the ASP. One reason may be the regulation mechanisms of fatty acids and TAG synthesis in algae. The argument is poorly understood and solving this lack of understanding can enhance lipid production and strain improvement. Fatty acids are common precursors for the synthesis of both membrane lipids and TAG. The correlation between lipids and fatty acids need to be elucidated. If the ability to control the fate of fatty acids varies among algal groups or even between isolates or strains, the basal lipid and TAG content may represent an intrinsic property of individual species or strains (DOE, 2010).

Lipid Synthesis and Regulation occur in different way. There is poor comprehension of the different mechanisms and of the relations between them. The argument needs an extensive research effort to be fully understood.

The major pathway for the formation of TAG in plants and algae is known as primary pathway for TAG synthesis. It involves new fatty acid synthesis. However, algae may possess multiple pathways for TAG synthesis, and the relative contribution of these individual pathways to overall TAG formation may depend on environmental or culture

Table 6. Chemical composition of algae on a dry matter basis (Demirbas, 2011).

| Species of sample | Proteins | Carbohydrates | Lipids | Nucleic acid |
|----------------------------------|----------|---------------|--------|--------------|
| <i>Scenedesmus obliquus</i> | 50-56 | 10-17 | 12-14 | 3-6 |
| <i>Scenedesmus quadricauda</i> | 47 | - | 1.9 | - |
| <i>Scenedesmus dimorphus</i> | 8-18 | 21-52 | 16-40 | - |
| <i>Chlamydomonas reinhardtii</i> | 48 | 17 | 21 | - |
| <i>Chlorella vulgaris</i> | 51-58 | 12-17 | 14-22 | 4-5 |
| <i>Chlorella pyrenoidosa</i> | 57 | 26 | 2 | - |
| <i>Spirogyra</i> sp. | 6-20 | 33-64 | 11-21 | - |
| <i>Dunaliella bioculata</i> | 49 | 4 | 8 | - |
| <i>Dunaliella salina</i> | 57 | 32 | 6 | - |
| <i>Euglena gracilis</i> | 39-61 | 14-18 | 14-20 | - |
| <i>Prymnesium parvum</i> | 28-45 | 25-33 | 22-38 | 1-2 |
| <i>Tetraselmis maculata</i> | 52 | 15 | 3 | - |
| <i>Porphyridium cruentum</i> | 28-39 | 40-57 | 9-14 | - |
| <i>Spirulina platensis</i> | 46-63 | 8-14 | 4-9 | 2-5 |
| <i>Spirulina maxima</i> | 60-71 | 13-16 | 6-7 | 3-4.5 |
| <i>Synechococcus</i> sp. | 63 | 15 | 11 | 5 |
| <i>Anabaena cylindrica</i> | 43-56 | 25-30 | 4-7 | - |

conditions. Organelle interactions and lipid body formation are other important factors. Chloroplast membranes are the primary location for fatty acid biosynthesis in plants and control the exchange of metabolites inside the algal cells. The relations between oxidative stress and lipids storage is the last aspect to be comprehended. Under environmental stress conditions (such as nutrient starvation), some algal cells stop division and accumulate TAG as the main carbon storage compound. Synthesis of TAG and deposition of TAG into cytosolic lipid bodies may be, with exceptions, the default pathway in some algae under stress conditions (Hu et Al., 2008).

5. Algae Pros and Cons.

In the first chapter we have analyzed some reasons that make biofuels an important resource and given a view of the fuel and transportation sectors. Afterwards we have discussed about the history of algal fuels and about algae from different point of views. In the next chapters we will analyze and discuss the processes and technologies to cultivate algae and product biofuels. We now present the main advantages and disadvantages and biological limitation of algae as source of biofuels, as summary of the reasons that make algal biofuels a challenging prospect.

5.1 Advantages of microalgae over traditional biofuel crops:

- High growth rate and productivity, that makes possible to satisfy the demand on biofuels using limited land resources.
- Possibility of growing in non-agricultural land.
- Algae do not require herbicides or pesticides (Rodolfi et al., 2009).
- Cultivation that consumes less water than land crops. Furthermore, algae do not necessarily need fresh water (Rodolfi et al., 2009).
- Absence of non-photosynthetic supporting structures (roots, stems, fruit). Microalgae are often single-celled organisms and no other tissues have to be supported. Related to this the cells do not have to spend energy moving storage molecules like starch around between tissues (Benemann, 1997).
- Most species allow continuous production. This avoids establishment periods of conventional plants and means that under optimal conditions biomass can continuously increase.
- The high number of strains permits to choose the best suited to different environmental conditions or to specific growth characteristics.

5.2 Technology advantages of microalgae:

- Carbon neutral process; high-efficiency CO₂ mitigation. Possibility of removing CO₂ from industrial flue gases.
- It is easy to provide optimal nutrient levels due to the aqueous growth environment and to the simple nutrients required (Benemann, 1997).
- Harvesting efforts can be controlled to match productivity. Ability to adjust harvest rates to keep culture densities at optimal levels at all times (Benemann, 1997).
- Results of small-scale, cost-effective, experiments can be effectively translated to the mass scale (Sheehan et al., 1998).
- Microalgal research and development is inherently simpler and faster than terrestrial species, due to their high cell division rate. (Benemann, 1997).
- Algae biodiesel contains no sulfur and performs as well as petroleum diesel, while reducing emissions of particulate matter (Delucchi, 2003).
- Possibility of wastewater treatment using as nutrients water contaminants such as NH₄⁺, NO₃⁻, PO₄³⁻ (Wang et Al., 2008).
- Several biofuels can be obtained (biodiesel, methane, hydrogen, ethanol, among others); possibility of burning the biomass in power plants.
- Several valuable compounds can be extracted including fine chemicals and bulk products (fats, polyunsaturated fatty acids, oil, natural dyes, sugars, pigments, antioxidants, high-value bioactive compounds and others) (Li et Al., 2008; Raja et Al., 2008).

5.3. Disadvantages and biological limitation of microalgae:

- Microalgal farming facilities have high capital costs and require rather intensive care.
- Organism survival. Current mass systems have been unable to maintain the laboratory conditions; open ponds often become contaminated by undesired strains.
- Most algae either grow or alternatively produce lipids. This requires either batch culture or separate growing ponds and lipid producing ponds, increasing production costs. Hybrid systems could be an interesting solution.
- Light penetration is a biological limitation of wild type algal growth. As seen, transgenics with reduced antenna size allow a greater efficiency at high light intensities.
- Seasonality. Algal growth is a function of temperature. Future research will have much to offer to overcome this problem.
- Harvesting is a cost-prohibitive engineering factor. Various technologies have been tested, often with unsatisfying results (Sheenan et Al., 1998).

- Technology problems, such as harvesting, are species specific. There are not universal solutions (Benemann, 2008).
- Biosafety of transgenics is delicate concern. However generated transgenic algae/cyanobacteria have the characteristic of being so domesticated that the organisms become totally unfit to exist in the wild (DOE, 2010).

CHAPTER 3.

Algal cultivation: open ponds or photobioreactors?

In the first chapter the potential of microalgae for biofuels production has been illustrated. There are two main algal culture systems: open and closed systems. Open culture system can be done in natural waters such as lakes or in artificial ponds, while closed system are called photobioreactors (PBRs). Advantages and disadvantages of each technology will be analyzed in this chapter, but the major difference between them is the trade-off of cost versus control.

Even if the two systems and technologies are diverse, they grow microalgae similarly. The culture differences between ponds and PBRs are due mainly to the higher degree of control that is reached in photobioreactors, where nutrient supply and growth phases are better regulated.

1. Algal growth and cultivation.

1.1. Site and algae selection.

The first thing to consider in the realization of an algal plant is the site and algae selection. The importance of the strain choice has already been stressed. Common sources of microalgae include existing collection of microalgae, commercially available either from Universities or other foundations. Of the 3000 strains screened by the ASP (Aquatic Species Program), approximately half are still available in the University of Hawaii (Sheenan et Al. 1998). According to Mata et Al. some of the main microalgae collection in the world can be found in the: University of Coimbra (Portugal); Goettingen University (Germany); University of Texas Algal Culture Collection (TX, US); CSIRO Collection of Living Microalgae (CCLM, Australia); National Institute for Environmental Studies Collection (NIES, Japan). Other options are to contact companies specifically devoted to algae growth or to research the desired strain utilizing genetic engineering. This last choice has the advantages of in house expertise and of intellectual properties ownership, even if lengthy in time and cost intensive. The criteria for a site selection were analyzed in depth during the ASP program. The site selection process goes in parallel with the resource evaluation and has to take into concern nutrients supply and geographic location. Easiness to access to nutrients and carbon supply source is necessary. Vast water amounts, considering its salinity and chemistry, are necessary as well. Furthermore it is important to find an inexpensive land in a position with optimal climatic conditions. Temperature, insolation, precipitation are all factors of great

importance. An ideal total incident solar energy could be around 150 W/m^2 (Lehr and Posten, 2009). This value is an approximation, because we will see that different systems could have diverse illumination needs. In addition different strains have diverse climate needs. Climatic regions most suitable for microalgae have an annual average temperature of over $15 \text{ }^\circ\text{C}$, with a night temperature between 4 and $10 \text{ }^\circ\text{C}$ and a day temperature between 10 and $22 \text{ }^\circ\text{C}$ (Ben-Amotz, 2008). We can identify some examples of suitable region for algal mass cultivation. In its location studies, ASP was focused on the US south west desert as the best region in the United States (Jarvis, 2008; Sheenan et Al. 1998). There are several example of successful algal cultivation, even if not for biofuels production, in Israel (Ben-Amotz, 2008), Hawaii (Benemann, 2008) and Australia, where huge artificial shallow lagoon are used (Packer, 2009). In Europe, southern Spain and Puglia in Italy have the characteristic of high incident solar energy, with low seasonal variations. In addition both the regions have large power plants that can provide substantial amounts of CO_2 and are close to the see, for an easy supply of water.

1.2. Algae cultivation.

Microalgae are able to adapt themselves to the growth conditions and this factor allows the growth control over them. A sufficient supply of a carbon source and light to carry out photosynthesis are required for biomass growth. Microalgae may assume many types of metabolism and are capable of a metabolic shift as a response to changes in the environmental conditions. Examples of algal metabolisms are: autotrophic, heterotrophic, mixotrophic and photoheterotrophic (DOE, 2010). The aim of this work does not include a deep analysis of algal metabolisms and we remand at works specifically on the argument. In DOE, 2010 the topic is well treated with several references for further studies. Our purpose is to identify the recurrent growth phases, which describe the growth of most species both in open ponds and in photobioreactors.

Mata et Al. represent the algal culture in a batch culture (Fig. 8). Five reasonably well defined growth phases can be recognized: (1) lag phase; (2) exponential growth phase, representing the maximum growth rate under the specific conditions; (3) linear growth phase; (4) stationary growth phase; (5) decline or death phase. The dashed curve indicates the nutrients concentration, showing a depletion during the stationary phase and onwards (Mata et Al., 2010).

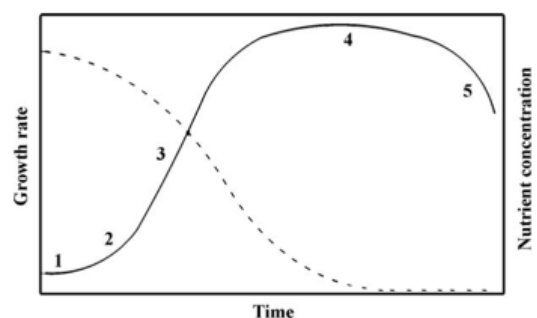


Fig. 8. Schematic representation of algae growth rate in batch culture (solid line) and nutrients concentration (dashed line) (Mata et Al., 2010).

Under suitable climatic conditions and sufficient nutrients, microalgae can grow profusely. Commonly they double their biomass within 24 h or within 3.5 h during the exponential growth phase (Chisti, 2007). Generally algal cultures in the exponential growth phase contain more protein, while cultures in the stationary phase have more carbohydrates and glycogen. There are several factors influencing algal growth. The next paragraph will take into account nutrients, their concentration, supply and effects on the culture.

- Light is the most important limiting factor for culturing algae in both closed and open outdoor systems.
- Temperature is the second factor to consider. Many microalgae can easily tolerate temperatures up to 15 °C lower than their optimal, but exceeding the optimum temperature by only 2–4 °C may result in the total culture loss (Mata et A., 2010). The effects of temperature reduce the possibility of algal cultivation in all the climates with a large seasonal temperature variation or with a considerable temperature span between night and day.
- The control over contaminants is particularly critical in open systems. Common biological contaminants observed include unwanted algae, mould, yeast, fungi, and bacteria.
- Salinity, in both open and closed systems, can affect the growth and cell composition of microalgae; every alga has a different optimum salinity range. Salinity can increase during hot weather conditions due to high evaporation and the easiest way for its control is by adding fresh water or salt as required.
- Mixing is another important growth parameter since it homogenizes the cells distribution, heat, metabolites, and facilitates transfer of gases. In photobioreactors it is important to provide a sufficient level of aeration, which lead to a better mixing of the microalgal culture. Prevented sedimentation, homogeneous conditions, and better contact between cells and nutrients are the results of a good mixing.

2. Nutrients.

Nutrients can be easily provided in a controlled way to the culture through runoff water. The main nutrients, in addition to water are: CO₂, nitrogen, phosphorous and iron. For diatoms, also silicate is essential. Minimal nutritional requirements can be estimated using the approximate molecular formula of the microalgal biomass that is CO_{0.48}H_{1.83}N_{0.11}P_{0.01} (Chisti, 2007).

Not only organic carbon, nitrogen, phosphorous and other nutrients as vitamins and salts are vital for algal growth, but also equilibrium between operational parameters (oxygen, carbon dioxide, pH, temperature, light intensity, product and byproduct removal, etc.) When

considering algal use for biodiesel production, it is important to quantitatively define the influence of these operational parameters and their interrelation. This allows to manipulate them and to obtain successfully a certain control over the composition of microalgae populations, even on a large scale (Sheenan, 1998). The ASP experience has repeatedly shown that properly managed algal cultures are quite resistant and that infections are often an indication of poor culture conditions.

The large-scale demand for microalgae may result in fertilizer shortages. The availability of phosphates and nitrates below some concentrations, depending on the cultivated species, will be a growth-limiting factor on the first growth phases (Bruton et al., 2009). Limitation of a key nutrient may also be desirable if under control. Nitrogen, phosphorous, or silicon limitation proved to induce oil accumulation in the cells in the stationary growth phase (Sheehan et al., 1998).

2.1. Flue gas utilization.

Most studies indicate that CO₂ addition to algal cultures stimulates growth (Fig. 5). If the cells utilize the carbon source, growth in both light and dark periods is possible, and high cell densities can be achieved. A potential disadvantage of the addition of external carbon sources is the possibility of increased contamination by undesired microbes living off the carbon source. (DOE, 2010)

To demonstrate that flue gases can be used for microalgae culture, NREL set up an experimental apparatus to supply controlled and measured amounts of such gases to the algal cultures. Fig. 9 shows a typical result, with no detectable difference between flue gas culture and the control gas (similar CO₂ levels, but without oxides of nitrogen (NO_x) and sulphur (SO_x)). The primary emission in flue gases is CO₂ at between 3% and 15% concentration depending on fuel source and design of the plant; coal-fired plants generally having higher CO₂ emissions (Packer, 2009). Other constituents of the flue gases however have to be considered, especially SO_x and NO_x and also metals present at much lower levels, nickel (Ni), vanadium (V) and mercury (Hg), again depending on the fuel used in the power plant that generates flue gases. Researches suggest that NO_x levels present in flues gases pose no problem for algal growth.

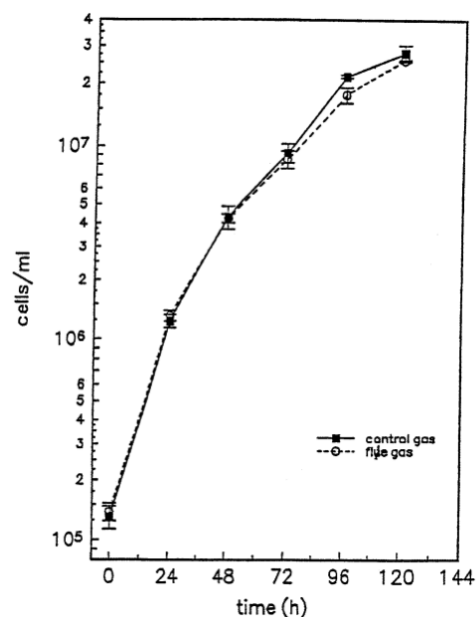


Fig. 9. Effects of flue gas components on microalgal growth (Sheenan et al., 1998).

On the contrary, SO_x can become a problem for some species if its concentration is above 400 ppm (Matsumoto et Al., 1997). Sheenan et Al. confirmed that SO_x and NO_x would not have a major effect on algal culture, but concluded also that the NO_x impurities could not provide the required nitrogen to the culture.

The experiment showed in fig. 9 and the studies reported above have an important consequence. Most of the laboratory and small-scale experiments in the field used control gas, that is purer and sometimes has different concentration of CO₂ than the flue gas from a power plant. It is likely that flue gas can be used in large-scale cultivation with results that are similar to the ones reached in laboratory experiments.

Nickel and V above 1.0 and 0.1 ppm, respectively, decrease algal productivity (Matsumoto et Al., 1997). These levels are higher than most flue emissions. There is no data to suggest that Hg has any detrimental effects on algal growth (Packer, 2009). Bioaccumulation of metals may be important if high-value nutritional oils were the desired final product. In addition the quality of the final biofuels may be affected by the presence of undesired metals.

Carbon is a key requirement; the composition of microalgae is of high percentage of carbon. For each kilogram of microalgae, different authors and companies suggest different required values of CO₂. As example, for every kilogram of dry weight algal biomass produced, Vertigo Algae Technologies affirms a use of 1,3–1,8 kg of CO₂ (Valcent, 2011), while PetroAlgae reports a value around 2,2 kg (PetroAlgae, 2011).

2.2. Nutrient source.

Nutrient supplies for algal cultivation have a sizeable impact on cost, sustainability, and production siting. Nitrogen, phosphorous, and iron additions represent a significant operating cost, accounting for 3–4 % of algal fuel cost (Benemann and Oswald, 1996). This calculation takes into account a 50% rate of nutrient recycle.

Phosphorous appears to be an especially important issue as there have been calculations that the world's supply of phosphate is in danger of running out (DOE, 2010). Nitrogen is typically supplied in one of three forms: ammonia, nitrate or urea. Because synthetic nitrogen fixation processes utilize fossil fuels, costs are tied to fossil fuel prices (DOE, 2010). Requirements for additional nutrients, such as sulfur, trace metals, vitamins, etc. must also be considered, but vary depending upon the specific strain and water source chosen.

Some authors suggest supplying the mineral supplements by a continuous fresh feed of water containing plant growth media, 30-10-10 fertilizer in detail (Powell and Hill, 2010). However, I consider this option too costly for mass production and searching for other more economically viable options is important. Therefore, utilizing the nutrient content of municipal, agricultural, or industrial waste streams is a very attractive alternative. Currently, algae are used in some wastewater treatment facilities because of their ability to provide oxygen for the

bacterial breakdown of organic materials and to sequester nitrogen and phosphorous into biomass for water cleanup (DOE, 2010).

Another approach to reduce nutrient costs is to pursue a diligent recycle. The final fuel product from algal oil contains no nitrogen, phosphorous, or iron; these nutrients end up primarily in the spent algal biomass. From a sustainability perspective, nutrient recycle may prove to be more valuable than using the spent biomass for products such as animal feed. According to Benemann and Oswald (1996) if the biomass residues are treated by anaerobic digestion to produce biogas, then most of the nutrients will remain in the digester sludge and can be returned to the growth system.

2.3. Water management.

We have seen algal ability to grow in water unsuitable for agricultural use, such as saline water from aquifers and seawater. At the same time, however, water management poses some of the largest issues for algal biofuels. An inadequate water use can easily make algal mass cultivation unfeasible. Water utilization could be an economic hurdle. Moreover polluting water could bring to the loss of public support due to perceived problems.

With large cultivation systems, water demands will be enormous. For example, a hypothetical 1 hectare, 15 cm deep open pond will require 1500 thousand liters to fill. In desert areas, according to evaporative losses demonstrated by Weissman and Tillet (1989) over 35 thousand liters of water per day can be lost from the 1 ha pond. Furthermore some actively growing algal cultures can double their biomass on a daily basis, meaning that half the culture volume must be processed daily. This is an enormous amount of water (750 thousand liters per day in the 1 ha example above).

Two considerations are relevant: first, the water being lost to evaporation is fresh water and replacing it with fresh water is a costly and often unsustainable option. On the other hand, adding low-quality water will concentrate salts, toxins, and other materials in the culture. A solution is to discard a portion of the pond volume each day. If this wasted water contains substantial nitrogen and phosphorous, disposal will become a problem. An advantage of closed photobioreactors over open ponds is a reduced rate of evaporation; evaporation however plays a critical role in temperature maintenance under hot conditions through evaporative cooling.

A second consideration is that moving around large volumes of water is a very energy-intensive process and can impose a significant cost. Water sterilization is energy-intensive and costly as well.

In conclusion, to contain costs, it is desirable to recycle most of that water back to the culture and treatment may be essential for water entering and exiting the process. For an optimal water management, developing cultivation systems with minimal water consumption,

studying methods to maximize water recycle and investigating ways to reduce the cost of water treatment and water movement will be important. Furthermore, selected a site, an analysis of the environmental laws regulating pollution and the water discharge is strongly suggested.

3. Open ponds.



Fig. 60. Cyanotech facility in Hawaii, where Natural Astaxanthin and Hawaiian Spirulina are cultivated in open ponds (Cyanotech, 2011).

3.1. Description.

Open ponds are the oldest and simplest systems for mass cultivation of microalgae. They have been used since the 1950s and they were accurately studied during the ASP. There are many different pond designs, from the round ponds with agitation provided by a rotating arm to inclined systems. Also natural lagoons are sometime used. However, the best option for developing algal biomass are shallow raceway ponds that allow efficient light exposure to the population of algal cells, good mixing and the highest productivity in open systems. The raceway pond has an elliptical shape and is mechanically mixed with a paddle wheel. This moves the water along the raceway, ensures vertical mixing of water to avoid algae settlement and to maximize gas exchange (Bruton et al., 2009). The ponds are kept shallow because of the need to keep the algae exposed to sunlight and the limited depth to which

sunlight can penetrate the pond water. The ponds are operated continuously; water and nutrients are constantly fed to the pond, while mature algae are continuously removed at one end. The size of these ponds is measured in terms of surface area, since this is so critical to capturing sunlight. Their productivity is measured in terms of biomass produced per day per unit of available surface area.

The raceway design is based on the high rate pond design (HRP, Fig. 11) developed in the Aquatic Species Program. Examination of subsequent literature (Ben-Amotz, 2008; Benemann, 2008; Huntley and Redalje, 2007; Chisti, 2007; Sheehan et al., 1998;) has shown that the basic design in Benemann and Oswald (1996) is still current, although some processing costs are likely to be lower due to modern technology.

A major handicap in the large-scale cultivation of algae using open systems is the inability to grow selected species in substantial volumes of hundreds of cubic meters. To date, only mass culture production of *Dunaliella*,

Chlorella and *Spirulina* in open pond systems was successful because of their tolerance to extreme conditions (salinity, alkalinity, etc.) that make these strains resistant to contaminants (Kovacevic and Wesseler, 2010). The world's largest open system *Spirulina* production, Earthrise Farm (44 ha), operates in the Imperial Valley USA and is shown in fig. 12. (Earthrise, 2011). The cultivation of more species is not feasible with nowadays technologies. To control and optimize the growth is very difficult and the products are algae with several different hydrocarbons. This algal product is complex to treat and convert to quality biofuels.

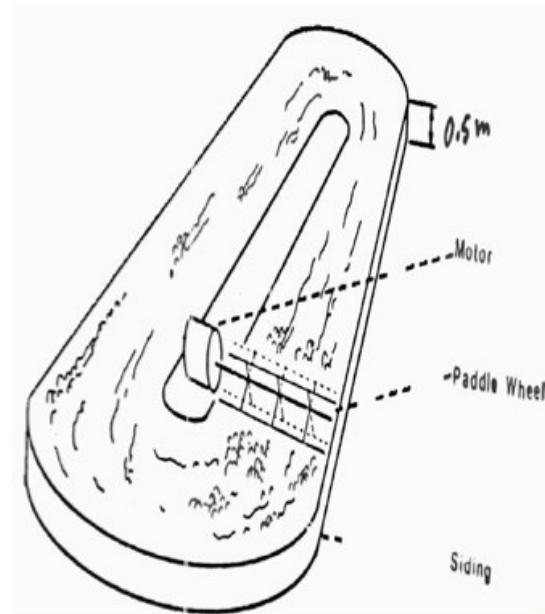


Fig. 11. High rate pond in the original 1984 design by Oswald (Ben-Amotz, 2008).

3.2. Pond design and operational parameters.

The dimensions of raceway ponds are not rigidly defined. Ben-Amotz (2008) reported a width of ten meters (two parallel channel, total width of 20 m) and a length of 150 m as the most common dimensions in open systems. Earthrise Farm has 30 ponds, each of 5000 m² and with a channel's width of 15 m. To obtain positive productivity results, the maximum depth of ponds is 30 cm, according to several studies (Sheenan et Al., 1998). Ben-Amotz suggests the minimum possible depth for large ponds: a good value could be around 15 cm (Richmond, 2004).



Fig. 17. The Earthrise spirulina farm (Earthrise, 2011).

Building an open pond of important dimension should be as economical as possible. The first step is the ground preparation. Several options are viable for pond lining (none, clay, concert, asphalt, fiberglass, plastic sheeting, others). Polyethylene (PE) or Polyvinyl chloride (PVC) are suggested because of their low Gauckler–Manning coefficient¹, that permits a better flow (Ben-Amotz, 2008). Outside walls and channel dividers should use the cheapest materials.

A paddle wheel mechanically mixes the pond. The paddle wheel location, design and parameters (number of blades, diameter, bottom to paddle distance, rpm, number of units per pond) are well treated by Ben-Amotz (2008) and by Sheenan et Al. 1998. The paddle influence on the pond's design is linked to the maximum area size served by it. The average operated area is around 1500 m², but depends on several factors. The paddle generates a flow velocity that has to be laminar and should be between 5 and 40 cm/s. 30 cm/s is a reference value. The continuous mixing of algal culture is required to prevent thermal stratification and to maintain carbonation.

The implementation of a computer system control is essential to regulate the site and the pH level. In particular, it can provide technology control of paddles, pumps, pipes, gases, sensors, liquid levels and feedbacks on biology information about pH, CO₂ flow, nutrient concentrations and water temperatures.

3.3. Operating and plant costs.

¹ The Gauckler–Manning coefficient indicates the opposition that a surface creates to a fluid flow. Smooth surfaces have a lower Gauckler–Manning coefficient and allow a better fluidity.

The raceway ponds require comparatively low capital investment. Operational costs are also low if the pond is controlled and regulated by a computer system. In this case weekly monitoring is enough to survey the biomass and nutrients. The pond cleaning is usually fast, simple and efficient. Energy is mainly consumed in the mixing operation.

It is very unlikely to imagine substantial technology improvements and cost reductions. Impressively the raceway pond implemented in New Mexico during the ASP, was designed in such a way to have a total power consumption of 0.04 W for m² (only 40 W for the entire 1000 m² pond) employing a counter-current injection system that allowed in 100% efficient CO₂ transfer to the water. Then the algae utilized over 90% of the injected CO₂ (Weissman and Goebel, 1987).

However, low cell concentrations in open cultivations make mixing and harvesting less effective. The downstream processes after an open pond cultivation are costly and energy intensive, harvesting and drying in particular. The ASP, concentrated exclusively on open systems, evaluated as too expensive the economical viability of biofuels from microalgae (Sheenan et Al., 1998; Benemann and Oswald, 1996). As viewed in the introduction, this situation has changed in the recent past, due to higher oil prices and better understanding of microalgae cultivation. Concerning open culture, it is important to underline that the possible economical feasibility is not due to a better technology. The change of perspective is more linked to better algal strains, with higher yield and productivity and to the increased price of fuels.

Open ponds plant's cost is strongly related to its size. Economies of scale are an important aspect in the creation of ponds. Plants of small dimension are very far from being feasible.

During the ASP techno-economic analyses projected detailed costs and revenues for open pond facilities (Benemann et Al, 1982; Weissmann and Goebel, 1987; Benemann and Oswald, 1996). However, whether the economic results of these works make still sense or can just provide a general indication, emerge as main question. Some costs are different because control technology and materials have been developed and the improved algal productivity provides higher revenues. However the main uncertainty is related with the difficulties to convert values in 1982 or 1987 dollars with nowadays currency. Notwithstanding in this paper the economic analysis of open pond facilities we will mainly follow the work made by Benemann and Oswald (1996).

3.4. Achievable productivity.

The low productivity is the main drawback of raceways. High light intensity causes cell mortality and contamination by fast growing microorganisms often happens. High biomass density cannot be achieved with these systems. The ASP program closeout report states the New Mexico plant, by the end of the research in 1996, were able to achieve a peak

performance of almost 300 t/(ha yr) dry weight biomass production, whereas at the beginning of the program they were producing around 50 t/(ha yr) biomass (Sheehan et al., 1998). The overall productivity was close to 36 t/(ha yr) or 10 g/(m² day) (Jarvis, 2008). However the results were far from achieving a 50% lipid concentration. In a recent report describing algal biomass for potential production in benign climates, with particular attention to New Zealand, Heubeck and Craggs say high rate algal pond production with CO₂ stimulation is between 40 and 75 t/(ha yr) (2007). Richmond reported obtainable values of 70-90 t/(ha yr), but only in perfectly managed pond (2004).

Large-scale open ponds lower productivity has one of the main causes in low night temperatures of the chose area. The coupling of waste heat from power plants and other industrial sources might help to overcome this problem. Chisti (2007) suggests about 1.5–3 times higher productivity is required for the technology to be really competitive. However, some companies claim impressively high yields compared to the ones achieved during the ASP; PetroAlgae for example claims that his open bioreactors have an overall biomass yield between 80 and 100 t/(ha yr) (PetroAlgae, 2011).

4. Photobioreactors.

4.1. Description.

PBRs are flexible systems that can be optimized according to the biological and physiological characteristics of the algal species being cultivated, allowing the cultivation of algal species that cannot be grown in open ponds. The reactor's walls limit the direct exchange of gases and contaminants between the cultivated cells and atmosphere are limited and this prevent evaporation, reduce CO₂ losses and offer a more safe and protected environment, preventing contamination or minimizing invasion by competing microorganisms.

Light does not impinge directly on the culture surface but has to cross the transparent reactor walls. Closed bioreactors can distribute the sun light over a larger surface area, which can be up to 10 times higher than the footprint area of the reactor. In existing commercial applications, artificial light and sometimes heat is used. During the RITE Japanese program optical fibers were used to diffuse light into the reactors (Sheehan et al., 1998); Siatem technology is testing photobioreactors with an internal LED illumination technology that uses only few light frequencies, in order to limit the use of energy. However, for economic reasons only solar light and waste heat are being nowadays considered for the biofuel production purposes.

Depending on their shape or design, PBRs are considered to have several advantages over open ponds: the main is better control over culture conditions and growth parameters (pH, temperature, mixing, CO₂ and O₂). Higher microalgae densities or cell concentrations with

higher volumetric productivities are achieved (Mata et Al., 2010) but PBRs require a continuous monitoring of nutrient and gas levels. The major drawbacks are the high capital and operating costs. Fouling and cleaning of PBRs external and internal walls is a big trouble as well. Over time accumulation of dirt (external) or algae (internal) will prevent light. Mixing to ensure optimum photosynthetic efficiency is also a major challenge. In order to maintain turbulent flow, energy needs to be supplied, generally for pumping, or for bubbling gases (Bruton et al., 2009), making the technology more energy-intensive than open ponds. Any parasitic energy load needs to be minimized in order to keep a positive energy balance on the overall process. The optimum degree of turbulence is a critical parameter; it promotes the fast circulation of microalgae cells from the dark to the light zone of the reactor but at the same time high liquid velocities and degrees of turbulence can damage microalgae due to shear stress (Mata et Al., 2010).

Until today only about 100 tons per year of microalgal biomass is derived from closed photobioreactors, while the biggest part of a few thousand tons is cultivated in open ponds (Lehr and Posten, 2009).

4.2. Photobioreactors design.

Closed systems consist of numerous designs: tubular, flat-plated, rectangular, continued stirred reactors, etc.

Tubular reactors (Fig. 13) are relatively cheap, have a large illumination surface area and have fairly good biomass productivities. They consist of transparent tubes that can be arranged vertically, horizontally, inclined, helical, or in a horizontal thin-panel design. The diameter of tubes is usually small (0.2 m diameter or less) and they are disposed to maximize the illumination surface-volume ratio of the reactor. The PBR is composed by this array of tubes or the solar collector, where the sunlight is captured, and by a reservoir. Growth medium circulates from the reservoir to the array and back to the reservoir.

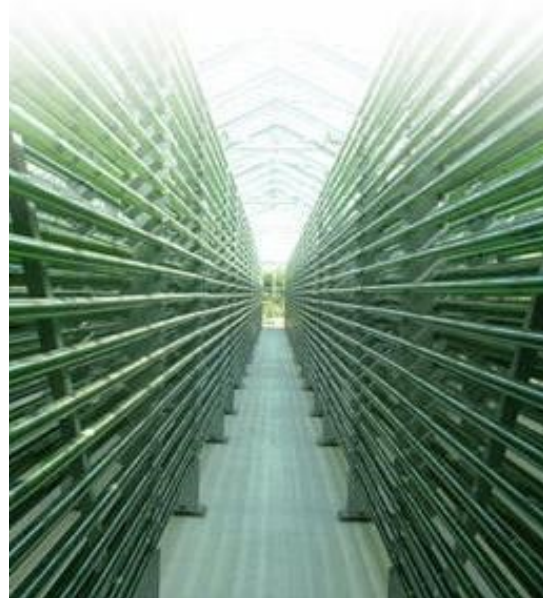


Fig. 13. Example of tubular photobioreactors (Benemann, 2008).

Disadvantages of this design include fouling, some degree of wall growth, dissolved oxygen and CO₂ along the tubes, and the pH gradients (Ugwu et al., 2008). Furthermore the land use is bigger than other PBR designs.

Column photobioreactors are compact with a low surface-volume ratio and have the advantage of high mass transfer. This design, with vertical bubble columns or airlift cylinders can attain good mixing with low shear stress, with substantially increased radial movement of fluid that is necessary for improved light–dark cycling. Consequently, cultures suffer less from photoinhibition and photooxidation, and experience a more adequate light–dark cycle. A much more chaotic gas–liquid flow than horizontal reactors, with low energy consumption is possible (Ugwu et al., 2008). Other advantages are high potential for scalability and easiness to sterilize, while limitations include their cost, small illumination surface area, sophisticated materials required in their construction. Since diameter and height cannot be much increased, a large number of units are needed to build a commercial plant. Vertical plate photobioreactors mixed by air bubbling seem even better than bubble columns in terms of productivity and ease of operation (Mata et Al., 2010).

Flat-plated photobioreactors have a large illumination surface area that allows high photosynthetic efficiency and low accumulation of dissolved oxygen concentration. The reactors are relatively cheap, easy to construct and easy to clean up and maintain. However, the large surface area presents scale-up problems, including difficulties in controlling culture temperature and carbon dioxide diffusion rate, and the tendency for algae adhering to the walls. It has been shown that vertical flat plates of 1000–2000 liters in volume can be successfully operated for long periods, hence having potential for scale up (Richmond, 2004). Nevertheless, scale-up requires many compartments and support materials (Ugwu et al., 2008).

Several other designs exist such as rectangular tanks reactors, continuous stirred tank reactors (CSTR), inclined triangular tubular photobioreactors, photobioreactors made by helical coils, etc.

The construction materials must lack toxicity, have high transparency, high mechanical strength, high durability and chemical stability and low cost. The ease of cleaning and loss of the plastics transparency exposed outdoors are operational issues to consider (Richmond, 2004).

The different designs have to consider technology and economics aspects. However, an ideal photobioreactors with perfect surface-volume ratio and that could reach high light conversion efficiency should take high trees as model (Fig. 14).



Fig. 14. Photobioreactor sculpture design by Charles Lee (BIOS, 2008).

4.3. Operating and plant costs.

Despite their advantages, it is not expected that PBR will have a significant impact in the near future on any product or process that can be attained in large outdoor raceway ponds. According to Benemann (2008), the advantages of PBRs are way overstated and for biofuels production PBRs are not affordable. The cost of biomass production in PBRs may be one order of magnitude higher than in ponds (Packer, 2009). While in some cases, for some microalgae species and applications it may be low enough to be attractive for aquaculture use, in other cases, the higher cell concentration and the higher productivity achieved in PBR may not compensate for its higher capital and operating costs. (Mata et Al., 2010)

Low cost designs are under research and may offer vast chances for the development of PBRs in mass scale. Several attempts have been undertaken to produce cheap reactors. Simple designs have been used for a long time. Novel attempts aim towards simplified installation, automation and maintenance (Lehr and Posten, 2009). One example is the V-shaped bag reactor from Novagreen, working as a bubble column (Fig. 15). Another low-cost, and scalable, variant is the bag system (Rodolfi, 2009). Other designs aim to higher efficiency in mixing, using airlift photobioreactor design implemented with the counter-current principle (Lehr and Posten, 2009). A photobioreactor that is cheap enough, energy efficient enough and stable enough to be used for commercial biofuel production over large areas does not presently exist. Nevertheless, the specific problems are addressed and partially solved (Lehr and Posten, 2009).



Fig. 15. V-shaped bag reactor from Novagreen (Novagreen, 2011).

The cost changes from photobioreactor to photobioreactor. Each photobioreactor also has its own productivity, influencing revenues. To compare the cost of different options is nowadays impossible because the companies do not provide cost information in their websites. The technology is in evolution but the market is still a niche and not developed enough. However, if in few years there will be significant technology improvements, the price would possibly be one of the leading factor in the comparison between different photobioreactors. From this point of view, the technology costs would be as advertised as the productivity is today. It is also important to underline that nowadays photobioreactors are costly because of the high research and development incurring costs of manufacturing companies. A significant future reduction may happen.

4.4. Achievable productivity.

The achievable productivity depends on the PBR type. Our intention is to show the potential that microalgae grown in photobioreactors have. Photobioreactors have higher efficiency and biomass concentration ($2\text{--}5\text{ g l}^{-1}$), shorter harvest time (2–4 weeks), and higher surface-volume ratio ($25\text{--}125\text{ m}^{-1}$) than open ponds (Wang et Al., 2008). High oil species of microalgae cultured in PBRs conditions have the potential to yield 19,000 – 57,000 l of microalgal oil per acre per year (Demirbas, 2010).

The commercial bioreactor supplier AlgaeLink claims for one of their systems a year round productivity in the order of 365 t/(ha yr) for several different species of algae. The productivity would be even greater, because the system requires an area of 0.83 ha and it is based on a

productivity rate equals to 500 g/m³ per day, productivity rate called “conservative” from the company.

Greenfuel Technologies Corporation, based in Massachusetts USA, who have several large-scale pilot plants operating and focus on CO₂ capture from industrial emitters, demonstrate dry weight productivities between 250 and 292 t/(ha yr) in their sunlight-powered algal bioreactors (Packer, 2009).

A widely stated claim is that microalgae are capable of producing over 30 times the amount of oil per unit area of land, compared to terrestrial oilseed crops (Sheehan et al., 1998). The actual global oilseed crop (sum of soy, rapeseed, sunflower and palm) oil production in 2007/2008 was 0.592 t ha⁻¹ for that year (Yu, 2008). If one assumes an oil concentration in algae of 42% the 365 t/(ha yr) productivity in the AlgaeLink bioreactors equates to 153.3 t/(ha yr) oil produced, which is about 259 times better productivity than the actual terrestrial oilseed crops. The reached productivities some years ago were thought to be impossible.

5. Open ponds versus photobioreactors.

For microalgae production a major decision to be made is whether to use closed photobioreactors or open ponds.

Open ponds of large area are relatively cheap to build, and easy to operate, but there is the impossibility of controlling contamination, the difficulty of maintaining a constant environment for the culture, particularly its temperature, and the low cell density that can be achieved. The latter point results in the need for extensive areas of land for the raceways and substantial costs for harvesting. To avoid microbial contamination, highly selective conditions have been used in some cases to guarantee dominance by the selected strain, but such conditions are not available for all species. Because of the drawbacks of open culture systems, much attention has been paid to closed photobioreactors, particularly with regard to the biomass productivity obtainable. However PBRs have considerably higher capital and operating costs than open ponds.

Regarding productivity, it is difficult to directly compare photobioreactors with open ponds; the evaluation depends on several factors, among which the algal species cultivated and the method adopted to compute productivity. Usually productivity per unit area is given for ponds where it is given as productivity per unit volume for enclosed bioreactors. As stated by Richmond, despite closed systems offer no advantage in terms of areal productivity, they largely surpass ponds in terms of volumetric productivity (8 times higher) and cell concentration (about 16 times higher). However the most useful way to express productivities for comparison between different production methods would be in biomass per unit light energy used or falling over a particular area (Packer, 2009; Richmond, 2004). This illuminated surface productivity can be expressed as g/(m² d).

Fig. 16 offers a comparison of the properties of various large-scale algal culture systems (Borowitzka, 1999). PBR and open ponds should not be viewed as competing technologies, because of the substantial differences and obtainable advantages that divide them. A potential solution is proposed in the next paragraph.

| Reactor type | Light utilization efficiency | Temperature control | Gas transfer | Mixing | Hydrodynamics stress on algae | Species control | Sterility | Scaleup |
|-----------------------------------|------------------------------|---------------------|--------------|-------------------|-------------------------------|-----------------|-------------------|----------------|
| Unstirred shallow ponds | Poor | None | Poor | Very poor | Very low | Difficult | None | Very difficult |
| Circular stirred ponds | Fair-good | None | Poor | Fair | Low | Difficult | None | Very difficult |
| Paddle-wheel | Fair-good | None | Poor | Fair-good | Low | Difficult | None | Very difficult |
| Raceway Ponds | | | | | | | | |
| Tanks | Very poor | None | Poor | Poor | Very low | Difficult | None | Very difficult |
| Stirred tank reactor | Fair-good | Excellent | Low-high | Largely uniform | High | Easy | Easily achievable | Difficult |
| Air-lift reactor | Good | Excellent | High | Generally uniform | Low | Easy | Easily achievable | Difficult |
| Bag culture | Fair-good | Good | Low-high | Variable | Low | Easy | Easily achievable | Difficult |
| Flat plate reactor | Excellent | Excellent | High | Uniform | Low-high | Easy | Achievable | Difficult |
| Tubular reactor (serpentine type) | Excellent | Excellent | Low-high | Uniform | Low-High | Easy | Achievable | Reasonable |
| Tubular reactor (biocoil type) | Excellent | Excellent | Low-high | Uniform | Low-high | Easy | Achievable | Easy |

Fig. 16. Comparison of the properties of various large-scale algal culture systems (Borowitzka, 1999).

6. Hybrid systems and alternative technologies.

Intermediate systems have been designed with the scope of a technology that has lower plant cost than photobioreactors but is more resistant to contaminants than open ponds.

There are simply systems such as open ponds under greenhouses for a more controlled environment or two-step cultivation processes that involve a combination of raceway and photobioreactor designs. It has been estimated that even adding simple plastic sheeting to cover over ponds would much more than double total systems capital and operating costs (Benemann and Oswald, 1996). The combination of raceway and photobioreactor has great potential. The first step is the fast cultivation of biomass in the PBR and the second step is stress cultivation in open ponds. A photobioreactor first step allows good protection of the growing biomass during early stages by maximizing the CO₂ capture. After the microalgae suspension is transferred to open ponds with low nitrogen nutrients and maintained high CO₂ levels. The open raceway in the second step often do not suffer of traditional open culture problems, because higher density of algal biomass is more resistant to external contamination and this nutrient deficit phase avoids the growth of contaminating species (Bruton et al., 2009). The combination of photobioreactor and open pond cultivation has proved efficient for astaxanthin production (Huntley and Redalje, 2007). Companies developing biofuel applications are currently testing it. The University of Florence has undertaken considerable research into this topic (Rodolfi et al., 2009). Literature data supports the approach, comparing the economics of the bioreactors coupled to ponds versus larger enclosed bioreactors on their own (Huntley and Redalje, 2007).

Most algae either grow or alternatively they produce lipids. This requires either batch culture or separate growing ponds and lipid producing ponds, increasing production costs. Hybrid

systems are a solution: photobioreactors are used for rapid cell division and then in open ponds algae rich in oil are produced in conditions of nutrient depletion. This combination is probably the most logical choice for cost-effective cultivation of high yielding strains for biofuels. Importantly, the size of the inoculum needs to be large enough for the desired species to establish in the open system before an unwanted species. Therefore to minimize contamination issues, cleaning or flushing the ponds should be part of the aquaculture routine, and as such, open ponds can be considered as batch cultures (Schenk et Al., 2008). For bioreactors when comparing batch or continuous systems, only continuous systems are realistically feasible for microalgal biomass (Packer, 2009). Hybrid systems are probably the only valuable exception.

CHAPTER 4

The downstream processes.

Conversion of algae to liquid transportation fuels requires processing steps such as harvesting, dewatering and extraction of fuel precursors (lipids and carbohydrates). Because algae are mostly water, these processes are difficult and energy-intensive. The critical importance of these processes has been recognized only recently, when concerns about reducing costs and energy use emerged in several feasibility studies. The biomass recovery from the culture medium may contribute to 20–30% of the total biomass production cost (Grima et Al., 2003). High costs related to harvesting are easily explained; microalgae are typically very small and cultures often have concentration lower than 0.5 g/l. In open ponds the algal concentration is even lower, sometimes cultures have as low as 0.02-0.06% algae total solid matter (Bruton et al., 2009) and natural oceanic microalgal levels are one to two orders of magnitude still lower. Before being further treated, these cultures must be concentrated to slurries containing at least 1% algae (but more often 10% is required) and large volumes must be handled. The final slurry concentration will depend on the extraction methods employed and will impact the required energy input. As the desired percentage of dry biomass increases, energy costs climb steeply.

Different factors influence the downstream necessary processes. The cultivated species, the cell's density, properties and size and the culture conditions all influence the technical choices. The most important factor to consider is the desired final products and the processes subsequently used: some processes require the algae to be completely dewatered, while others may require moderate dewatering (Packer, 2009). Also the desired product quality is one main criterion for selecting a proper harvesting procedure (Richmond, 2004).

Different technologies can be used in series or alternatively. As example, dewatering and heating can occur in the same operation, or can be in series; otherwise with an appropriate dewatering technology or with a two-step dewatering heating can be avoided. Although a universal harvesting method does not exist (Benemann, 2008), this is still an active area for research, being possible to develop an appropriate and economical harvesting system for any algal species. Efforts in the field can enhance the feasibility of biofuels from microalgae. A clear distinction of downstream processes is difficult to be made. There are three major requirements that need to be addressed in the great majority of cases: harvesting, dewatering and/or drying and the extraction of fuel precursors.

1. Harvesting.

The harvest phase is designed to remove the bulk of excess water that the algae are located in. The sedimentation and flotation harvesting techniques mainly apply to open pond cultivation systems because of their low culture concentration, while filtration and centrifugation mainly apply to PBRs or for high-value products. To recover high quality algae such as for food or aquaculture applications, it is often recommended to use continuously operating centrifuges that can process large volumes of biomass.

1.1. Sedimentation and flocculation.

The simple sedimentation system is suitable for microalgae species with naturally high sedimentation rates. Most of microalgae and cyanobacteria remain in suspension in well-managed high growth rate cultures due to their small size (circa 1 to 30 μm). However, naturally flocculation leading to sedimentation occurs in many older cultures. Interrupting the carbon dioxide supply to an algal system can cause algae to flocculate on its own, which is called autoflocculation. Gravity sedimentation is performed in thickeners or clarifiers, standard processes in water treatment plants. Sedimentation tanks or settling ponds are also possible and usually have a retention time around six hours. The capital and operation costs are low.



Fig. 16. Natural sedimentation (Lundquist, 2008).

If the strain has poor sedimentation properties, a flocculation agent can be used. In managed cultures, forms of forced flocculation usually involve chemical additives. Water that is more brackish or saline requires additional chemical flocculants to induce flocculation, causing the algae to settle at the bottom of the tank. Many flocculants are multivalent cations such as aluminum, iron, calcium or magnesium. These positively charged molecules interact with negatively charged particles to reduce the barriers to aggregation. Flocculation is used to increase the size of the algae aggregates and it is often combined with dissolved air floatation (DAF) (DOE, 2010; Packer, 2009). DAF uses tiny bubbles that are injected under high pressure into the water column and as they rise to the surface they drag organic molecules and cells with them. The bubbled air causes the algal clusters to float to the surface. The algae-rich top layer is scraped off to a slurry tank for further processing. These aggregated clusters are more easily filtered and/or settle more rapidly.



Fig. 17. Chemical coagulation plus flotation (Lundquist, 2008).

Flocculation removes roughly 80% of the water from the harvest, leaving behind concentrated algae mass. Compared to sedimentation the flotation process is very fast, it only requires a few minutes instead of hours for sedimentation. Capital and operating costs are also low, but the efficiency may be poor in shallow-depth ponds (Bruton et Al., 2009).

The addition of flocculants increase the efficiency but can also cause problems, depending on how the biomass is to be processed downstream. Electroflocculation and electrocoagulation offer the advantages of no added chemicals.

1.2. Centrifugation.

Centrifugation technologies must consider large initial capital equipment investments; operating costs are relevant, and large quantities of water and algae are treated. The current level of centrifugation technology makes this approach cost-prohibitive for most of the large-scale algae biorefineries. Significant cost and energy savings must be realized before any widespread implementation of this approach can be carried out (DOE, 2010). Albeit at considerable cost, centrifuges are suitable to rapidly concentrate any type of microorganisms, which remain fully contained during recovery. Additionally, these devices can be easily cleaned or sterilized to effectively avoid bacterial contamination or fouling of raw product. The efficiency is dependent on the selected species (as related to size). Fig. 18 shows an example of a commercial centrifuge.

A variation of centrifugation that has not been sufficiently explored for use with algal biomass collection, but is widespread in the petroleum and mining industries, is the use of hydrocyclones (Packer, 2009). In these devices water containing the particles, in this case algal cells, is channeled in a spiral fashion creating centripetal forces causing the denser particles to be spun out of the traversing liquid. Although the technique works for removing dense particles from liquid streams and for separating oil from water, their application to soft algal cells is experimental. However, its simplicity and fewer moving parts avail its potential for large-scale economic application.

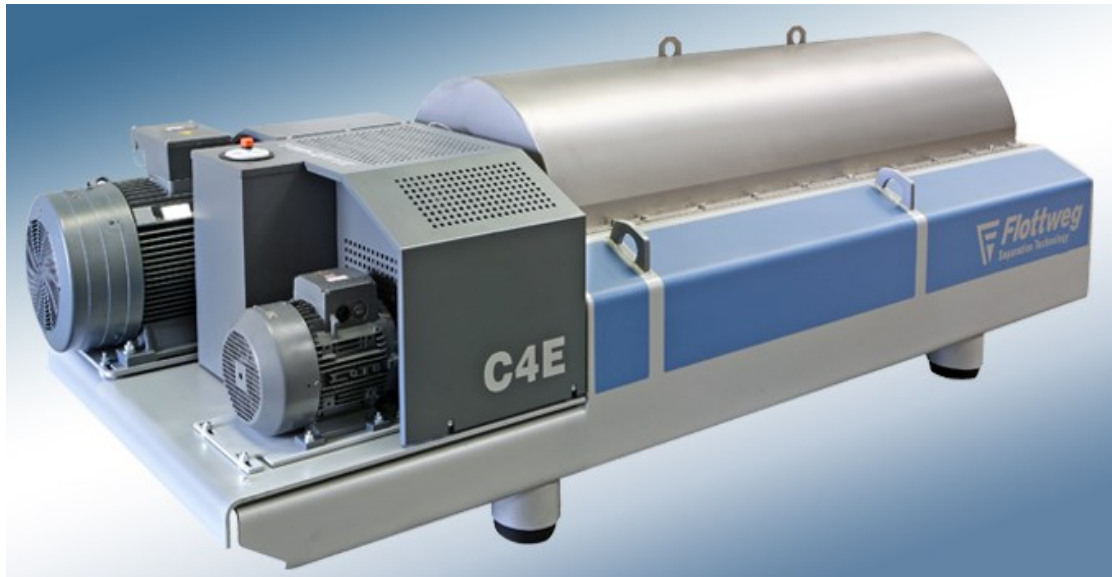


Fig. 18. A centrifuge by Flottweg, used for the thickening and dewatering of a variety of sludges at different stages of wastewater and water treatment (Flottweg, 2011).

2. Dewatering: filtration and drying.

Dewatering processes are strongly related to the harvesting methods used. Different harvesting procedures adjust the density or the acceptable level of moisture in the resulting concentrate right to the optimum subsequent process (Richmond, 2004; Grima et Al., 2003). Gravity sedimented sludge is generally more diluted than centrifugally recovered biomass, which substantially influences the economics of product recovery further downstream. Since costs of thermal drying are much higher than those of mechanical dewatering, in order to reduce the overall production cost, a concentrate with higher solids content is required after harvest to easy biomass dehydration (Mata et Al., 2010). A combination of methods is often used, such as a pre-concentration with a mechanical dewatering step, filtration, or centrifugation and then, a post-concentration by means of a screw centrifuge or a thermal drying.

Microalgae and cyanobacteria present unique filtration challenges because most strains considered for energy feedstocks have cell diameters less than 10 μm . Filtration is conceptually simple but potentially very expensive. Filtration processes can range from simple screening or micro-strainers to dewatering up to complex vacuum or pressure filtration systems. The filter pore size is critically important as it is defined by the size of the algae species and algae aggregation rate. Small algae pass through larger pores decreasing filter efficiency. Decreasing pore size, however, leads to blinding. The main limitation of filtration is plugging that can be solved by vibrating screens or tangential filtrations. Some combined systems use pressing and screening belts, having the advantage of continuous

operation (Bruton et Al., 2009). An ideal filtration design should require minimal or no washing requirements. Power costs will certainly influence design.

Mechanical dewatering (pressing and filtration) can be cheaper than heating (Grima et Al., 2003), but the real key is to have as few steps as possible and simple scalable extraction (Packer, 2009). Filtration is better suited for large microalgae such as *Coelastrum proboscideum* and *S. platensis* but cannot recover organisms with smaller dimensions such as *Scenedesmus*, *Dunaliella*, or *Chlorella* (Grima et Al., 2003).

Drying is required to achieve high biomass concentrations. Drying generally requires heat; sun-drying, spray-drying, freeze-drying, methane drum dryers and other oven-type dryers have been used. However, the costs climb steeply with incremental temperature and/or time increases (DOE, 2010). Air-drying is possible in low-humidity climates, but will require extra space and considerable time. Solutions involving either solar or wind energy are also possible.

After separation from the culture medium algal biomass (5–15% dry weight) must be quickly processed lest it should get spoiled in only a few hours in a hot climate (Mata et Al., 2010).

3. Extraction of oil and other products from algae.

Extraction is dependent on the algal species and growth status and requires the cell disruption of the microalgae cells in order to release the metabolites of interest. Furthermore different harvest process operations could affect extraction processes, as well as the fuel conversion process. Many effective extraction techniques require concentrated substrates, thus a high degree of concentration, with very low water content, may be necessary before some types of extraction can begin. To be successful, any extracting solvent must be able to penetrate through the matrix enclosing the lipid material, physically contact the lipid material, and then solvate the lipid. The tissue structure and cell walls may present formidable barriers to solvent access. (DOE, 2010).

According to Mata et Al. (2010), the possible extraction methods can be either based on mechanical action (e.g. cell homogenizers, bead mills, ultrasounds, autoclave, and spray drying) or non-mechanical action (e.g. freezing, organic solvents and osmotic shock and acid, base and enzyme reactions).

The simplest method is mechanical crushing: the algal oil content can be pressed out with a press. Since different strains of algae vary widely in their physical attributes, various press configurations (screw, expeller, piston, etc.) work better for specific algae types. Often, mechanical crushing is used in conjunction with chemical solvents.

Ultrasonic extraction can greatly accelerate extraction processes. Using an ultrasonic reactor, ultrasonic waves create cavitation bubbles in a solvent material. When these

bubbles collapse near the cell walls, they create shock waves and liquid jets that cause those cells walls to break and release their contents into the solvent.

Hexane benzene and ether have been used as chemical solvents. Hexane extraction is widely used in the food industry and is relatively inexpensive. Working with chemicals involves potential dangers and care must be taken to avoid exposure to vapors and direct contact with the skin. Benzene is classified as a carcinogen. Chemical solvents also present the problem of being an explosion hazard. Hexane in combination with an oil expeller or press is the process typically used. After the oil has been extracted using an expeller, the leftover pulp can be mixed with cyclohexane to extract the remaining oil. The oil dissolves in the cyclohexane, and the pulp is filtered out from the solution. The oil and cyclohexane are separated by means of distillation. These two stages (cold press and hexane solvent) together are able to derive more than 95% of the total oil present in the algae (Packer, 2009). Osmotic shock is a sudden reduction in osmotic pressure, this can cause cells in a solution to rupture. Though requiring low-energy input, osmotic shock is probably the method with the lowest efficiency and creates a further issue for some downstream processes, as water content can be a problem requiring energy input to overcome (Packer, 2009).

Critical point gas/fluid extraction is probably the most efficient method for complete extraction of the oils. CO₂ is the most tested gas for this technology and the CO₂ is recycled during the process. CO₂ is liquefied under pressure and heated to the point that it has the properties of both a liquid and a gas. Then it acts as the solvent in extracting the oil. However this is a very energy-intensive process and it is unlikely to be economic for the extraction of commodity oils for biofuels.

New technologies in extraction methods, such as ultrasound and microwave assisted compared with conventional methods can greatly improve oil extraction with higher efficiency (Mata et Al., 2010).

4. Cost of downstream processes.

The biomass recovery from the culture medium requires difficult and energy-intensive processes that may contribute to 20–30% of the total biomass production cost. The effectiveness of the processes is related with their cost.

A simple example can help to realize harvesting technology's cost. For an Algaelink 1 ton/day dry biomass PBR plant, the algae harvest capacity is around 380 ton/day, with an hourly feed to harvesting equipment of 16 m³/h (Algaelink, 2011). Alfalaval produces centrifuge modules of different sizes. The CLARA separation unit family is specially designed for use in the wine and beverage industries but can be effectively used in algal harvesting (Alfalaval, 2011). CLARA-200 has the correct harvesting capacity for the example above and has a commercial cost around 100 k€. The price is approximately 8-10% of the cost of an

Algaelink's 1 ton/day photobioreactor, including tubes, pumps and control software and sensors.



Fig. 19. Alfalaval CLARA-200 separation centrifuge (Alfalaval, 2011).

Also dewatering processes can be very expensive. A self-discharging centrifuge with the capacity of 60 m³/h can be found in the market for a price around 250 k€. A drum drier with a capacity of 400 kg/h can be found in the market for approximately 300 k€. On the other side, mechanical presses are relatively cheap and several different types are offered in the market. Chemicals solvent techniques are likely to be more expensive. In one project under the ASP, solvent extraction costs were three times higher for algal oil than for soybean oil due to high moisture content of the paste in the experiment (Sheehan et al., 1998).

Relatively low-grade heat (such as waste heat) could be employed to help the separation of solvents from oil, greatly increasing the overall economics.

Powell and Hill (2010) describe an integrated process where microalgae are cultivated in combination with ethanol distillation. The distillation plant produces a huge quantity of steam. Superheated steam at 150 °C is used for both the drying and the extraction processes. In the described design, after dewatering, the resulting thick broth is pumped into a continuous flow, hot hexane extraction/settling tank system. The sudden temperature change and violent mixing causes the microalgae cells to release their oils, which are quickly dissolved in the hexane. These oils are then separated in a flash tank, and all the hexane is recycled while the oil leaves as the desired co-product. The bottoms of the extractor go to a continuous tunnel dryer where superheated, low-pressure steam evaporates the remaining water and generates a 10% moisture content, dried algae biomass co-product. The used steam accounts for 10% of the existing steam already being produced for ethanol distillation in the original plant (Powell and Hill, 2010). Potentially every plant with wasted heat offers great cost saving prospective.

In order to reduce their production costs, some algae-to-biofuels processes attempt to bypass the extraction step by either converting whole algal biomass or by inducing the secretion of the desired product directly.

Algenol is developing a technology that extracts ethanol from microalgae avoiding all the downstream processes described in this chapter. The algae produce internal sugars through

photosynthesis, sugars that are converted into ethanol inside each algal cell. The ethanol diffuses through the cell wall into the culture medium and then evaporates, along with water condensing on the inner surface of the bioreactor. Subsequently it is collected by gravity, concentrated, and then distilled into fuel grade ethanol (Algenol, 2011).

Genetic modifications could offer other solutions. One of the hard parts of dealing with algae is getting the oil out of the cells. At NREL solvents are typically used but the cell wall can resist them. NREL is working to find enzymes that can help degrade the cell wall of algae and allow the solvents access permitting an efficient extraction of the oil. If an enzyme is found that can easily break down the cell wall, it might be possible to isolate the gene for that enzyme and engineer the algae to produce that enzyme just before harvesting processes. In this hypothesis the cells become weakened on their own and it is possible to separate out the oil and return the survived cells into cultivation (NREL, 2010). That would be a real cost savings improvement.

CHAPTER 5

Biofuels conversion.

1. Biofuels obtainable.

A wide range of fuels can be produced from algae. In this chapter several alternatives will be discussed, coupled with the required technologies. Even if possible products range from gaseous compounds like hydrogen and methane, to alcohols and conventional liquid hydrocarbons, to pyrolysis oil and coke, the most attractive targets are liquid transportation fuels (gasoline, diesel, and jet fuel).

Several reasons make them the best-value targets. Nowadays they are the primary products currently created from imported crude oil for the transportation sector. Equivalent biofuels have the potential to be more compatible than other biomass-based fuels with the existing fuel-distribution infrastructure. Moreover, adequate specifications for these fuels already exist. Going deeper in this last aspect, when a fuel meets all customer requirements, it is referred to as “fit for purpose.” The required specifications are numerous. Some of these are energy density, oxidative and biological stability, lubricity, cold-weather performance, elastomer compatibility, corrosivity, emissions (regulated and unregulated), viscosity, distillation curve, ignition quality, flash point, low-temperature heat release, metal content, odor/taste thresholds, water tolerance, specific heat, latent heat, toxicity, environmental fate, sulfur and phosphorus content (DOE, 2010). Posten and Schaub (2009) compared the mass and energy density of petroleum diesel with some biofuels (Table 8). Interestingly values of biodiesel (FAME, Fatty Acid Methyl Ester) and petroleum diesel are quite similar.

Table 8. Energy and mass density for biodiesel, ethanol, synthetic diesel, SNG, hydrogen and petroleum diesel (Posten and Schaub, 2009).

| <i>At 15 C; SNG and H at 200 bar</i> | <i>FAME (Biodiesel)</i> | <i>Ethanol</i> | <i>Synthetic diesel</i> | <i>Substitute Natural Gas</i> | <i>Hydrogen</i> | <i>Petroleum Diesel</i> |
|--|-----------------------------|----------------|-----------------------------|-----------------------------------|-----------------|-----------------------------|
| <i>Density (g/cm³) mass</i> | 0,88 | 0,79 | 0,78 | 0,135 | 0,017 | 0,84 |
| <i>Density (MJ/L) energy</i> | 32,6 | 21,2 | 34,3 | 6,4 | 1,9 | 35 |

There are multiple examples of cars that have used biodiesel from algae successfully (Fig. 20). The U.S. navy successfully tested a 15 meters gunboat fueled by a 50-50 blend of diesel and algae (Reuters, 2010) and great research efforts are made in the development of jet fuels from algae. Algae biodiesel contains no sulfur and performs as well as petroleum diesel, while reducing emissions of particulate matter, CO, hydrocarbons, and SO_x. However emissions of NO_x may be higher in some engine types (Delucchi, 2003).



Fig. 20. First car in the world to run algae biodiesel, December 2005 (Benemann, 2008).

There are some key aspects that have to be considered before approaching in the conversion part. As anticipated in chapter 4, the feedstock, conversion process, and final fuel specifications are highly interdependent and must be considered together if an optimal process is to be identified. Moreover, life cycle analysis of energy and carbon will be a key tool in selecting the preferred fuel conversion technologies. Finally, the greatest challenge in algal fuel conversion is not likely to be how to convert lipids or carbohydrates to fuels most efficiently, but rather how best to use the algal remnants after the lipids or other desirable fuel precursors have been extracted. The reason is that these algal remnants are the key to achieve economics feasibility. Life cycle analysis, energy balance and economic feasibility will be discussed in the next chapter.

A large number of potential pathways exist for the conversion from algal biomass to fuels. These pathways can be classified into the following three general categories (DOE, 2010):

- 1) Those that focus on the direct algal production of recoverable fuel molecules (ethanol, hydrogen, methane, and alkanes) from algae without the need for extraction;
- 2) Those that process whole algal biomass to yield fuel molecules;
- 3) Those that process algal extracts (lipids, carbohydrates) to yield fuel molecules.

2. Conversion technologies.

2.1. Direct production of biofuels from algae.

The direct production of biofuel through heterotrophic fermentation and growth from algal biomass has certain advantages in terms of process cost because it can eliminate several process steps (such as oil extraction) and their associated costs in the overall fuel production process. Heterotrophic growth also allows for maintaining highly controlled conditions, which first could be oriented toward biomass production and then oil production. Furthermore the system is readily scaled up. There are several biofuels that can be produced directly from algae, including alcohols, alkanes, and hydrogen.

Algae are capable of producing ethanol and other alcohols through heterotrophic fermentation of starch (Hon-Nami, 2006). This can be accomplished through the production and storage of starch via photosynthesis within the algae and subsequent anaerobic fermentation of these carbon sources to produce ethanol under dark conditions in closed photobioreactors. The ethanol is secreted into the culture media and is collected in the headspace of the reactor, purified, and stored. Extracting the alcohols directly from the algal culture media requires less capital and energy than competitive algal biofuel processes. The process essentially eliminates the need to separate the biomass from water and extract and process the oils (Fig. 21).



Fig. 21. The direct production of biofuels from algae could potentially eliminates downstream operations such as harvesting, dewatering and extracting the oil, operations described in chapter four and showed above (image from PetroAlgae, 2011).

This technology is estimated to yield 4,000 - 6,000 gallons (15,140 - 22,700 liters) per acre per year, with potential increases up to 10,000 gallons (37,850 liters) per acre per year within

the next 3 to 4 years with significant R&D (DOE, 2010). In addition to ethanol, it is possible to use algae to produce other alcohols, such as methanol and butanol, using a similar process technology.

Also alkanes may be produced directly by heterotrophic metabolic pathways using algae. The algae are fed sugars, the cheap availability of which is a key consideration for cost-effective production of biofuels.

These alkanes can theoretically be secreted and recovered directly without the need for dewatering and extraction. More often are associated with the algae and thus must be recovered through dewatering and extraction with the associated costs, making the advantages of the process (high productivity and high growth rate) less interesting.

Production of biohydrogen by direct photolysis is another interesting option. However, challenges remain before biological hydrogen production can be considered a viable technology. First the biohydrogen from algae is energy-intensive and has a very high cost, far higher than existing industrial processes. One important component of the cost is the high temperature required by the process (Fig. 22). Secondly the social acceptance of hydrogen has still to be achieved; the development of a robust hydrogen infrastructure is necessary.

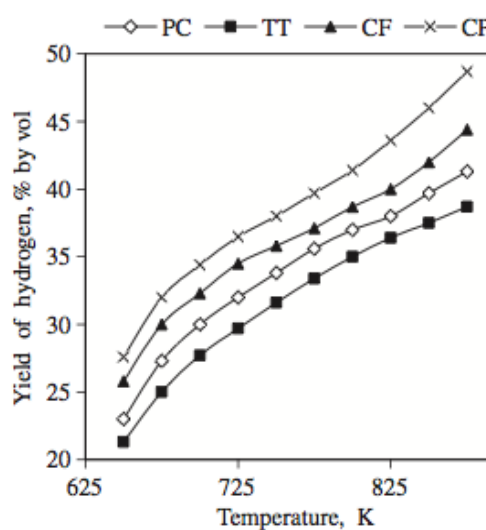


Fig. 22. Plots for yields of hydrogen in gaseous products from the samples by pyrolysis: Polytrichum commune (PC), Thuidium tamarascinum (TT), Cladophora fracta (CF), and Chlorella protothecoides (CP) (Demirbas, 2010).

2.2. Processing the whole algae.

The whole algae can be processed into fuels instead of first extracting oils and post-processing. These methods benefit from reduced costs even though some level of dewatering is still required. Even if macroalgae are not interest of this research, it is important to underline how this technology is probably the best suited for them, because of their low lipid content that makes the conversion to biodiesel economically unfeasible.

According to DOE (2010) there are four major categories of conversion technologies capable of processing whole algae: anaerobic digestion, supercritical processing, pyrolysis and gasification (Fig. 23).

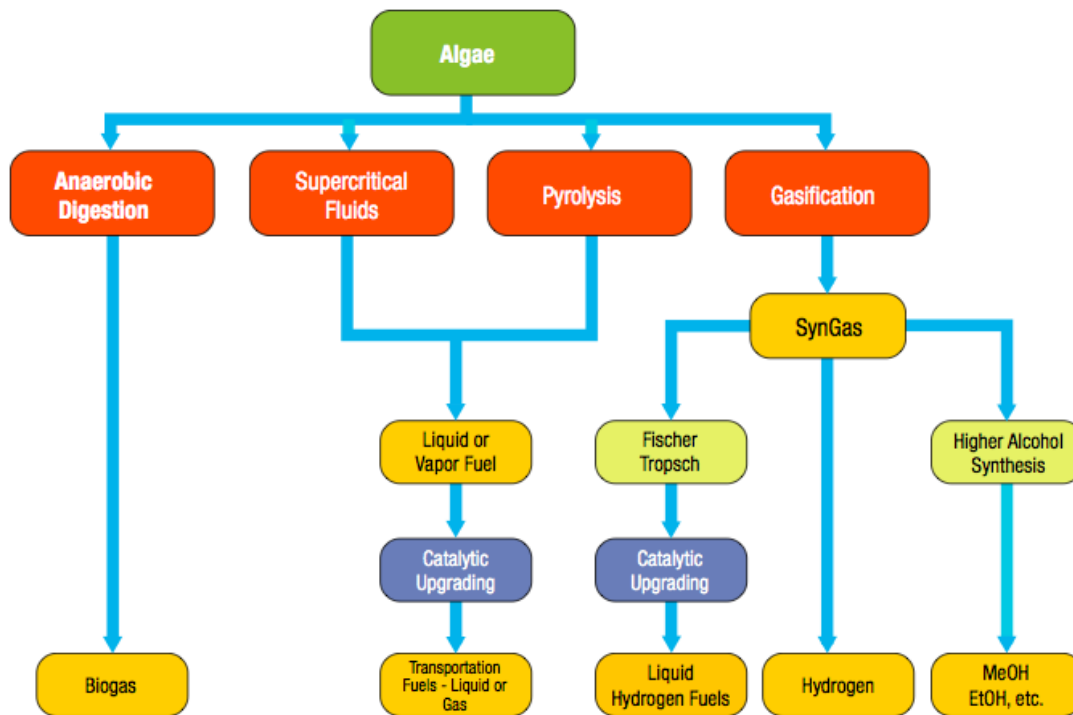


Fig. 23. Schematic of the potential conversion routes for whole algae into biofuels (DOE, 2010).

2.2.1. Anaerobic Digestion of Whole Algae.

The use of this conversion technology eliminates several of the key obstacles that are responsible for the current high costs associated with algal biofuels, including drying, extraction and fuel conversion, and as such may be a cost-effective methodology. The potential of this approach for macroalgae has been demonstrated. The process still needs to be adapted for microalgae; it may be very effective for situations like integrated wastewater treatment, where algae are grown under uncontrolled conditions using strains not optimized for lipid production.

2.2.2. Supercritical Processing.

Supercritical processing is a recent technology capable of simultaneously extracting and converting oils into biofuels. Supercritical fluid extraction of algal oil is far more efficient than traditional solvent separation methods. Supercritical fluids are selective, thus providing high purity and product concentrations. Extraction is efficient at modest operating temperatures (less than 50 °C) ensuring maximum product stability and quality (Demirbas, 2009). Additionally, supercritical fluids can be used on whole algae without dewatering, thereby increasing the efficiency of the process. Although it has been only demonstrated for the simultaneous extraction and transesterification of vegetable oils, it is envisioned as being applicable for the processing of algae. It remains to be seen whether the processing of whole

algae in this fashion is superior, in terms of yield, cost, and efficiency, to the transesterification of the algal oil extracts (DOE, 2010).

The economics of supercritical transesterification process, at least in the case of vegetable oil processing, has been shown to be very favorable for large-scale deployment. One economic analysis has been conducted based on a supercritical process to produce biodiesel from vegetable oils in one step using alcohols (Anitescu et Al., 2008). It was found that the processing cost of the proposed supercritical technology could be near half of that of the actual conventional transesterification methods (i.e., \$0,26/gal vs. \$0,51/gal or \$0,068/l vs. \$0,134/l).

The clear immediate priority is to demonstrate that these supercritical process technologies can be applied in the processing of algae, either whole or its oil extract, with similar yields and efficiencies at a level that can be scaled to commercial production.

2.2.3. Pyrolysis.

Pyrolysis is the chemical decomposition of a condensed substance by heating. The thermochemical treatment of the algae, or other biomass, can result in a wide range of products, depending on the reaction parameters. Pyrolysis has one major advantage over other conversion methods: it is extremely fast, with reaction times of the order of seconds to minutes. However it requires very high temperatures (Fig. 24).

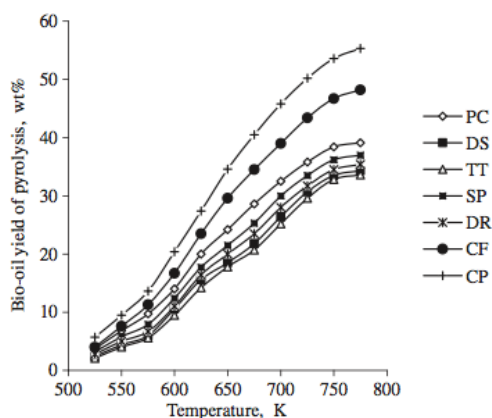


Fig. 24. Curves for yields of bio-oil from pyrolysis of the samples: *Polytrichum commune* (PC), *Dicranum scoparium* (DS), *Thuidium tamarascinum* (TT), *Sphagnum palustre* (SP), *Drepanocladus revolvens* (DR), *Cladophora fracta* (CF), and *Chlorella protothecoides* (CP) (Demirbas, 2010).

Although synthetic diesel fuel cannot yet be produced directly by pyrolysis of algae, a degradable alternative liquid called bio-oil can be produced. The bio-oil has an advantage that it can enter directly into the refinery stream and, with some hydrotreating and hydrocracking, produce a suitable feedstock for generating standard diesel fuel. Achieving an efficient dewatering is the major bottleneck for the technology, even if several pilot plants for fast pyrolysis of biomass have been built in the past years in Germany, Brazil and the United States. The process is very energy-intensive and might produce small amounts of potentially toxic by-products (Packer, 2009).

2.2.4. Gasification.

Gasification of the algal biomass may provide an extremely flexible way to produce different liquid fuels, mainly through Fischer-Tropsch Synthesis (FTS). Gasification of lignocellulose is relatively mature and it is reasonable to expect that once water content is adjusted for, the gasification of algae to these biofuels would be comparatively straightforward.

Conversion of bio-syngas has several advantages over other methods. It is very flexible and it is possible to create a wide variety of fuels with acceptable and known properties. The key roadblocks to using FTS for algae are the required very large-scale production to make the process efficient overall and the cost of clean-up and tar reforming.

2.3. Conversion of algal extracts.

The conversion of extracts derived from algal sources is the typical mode of biofuel production from algae. The most common type of algal extracts under consideration are lipid-based, e.g., triacylglycerides, which can be converted into biodiesel. Chemical, biochemical, and catalytic processes can be employed to convert algal extracts (Fig. 25).

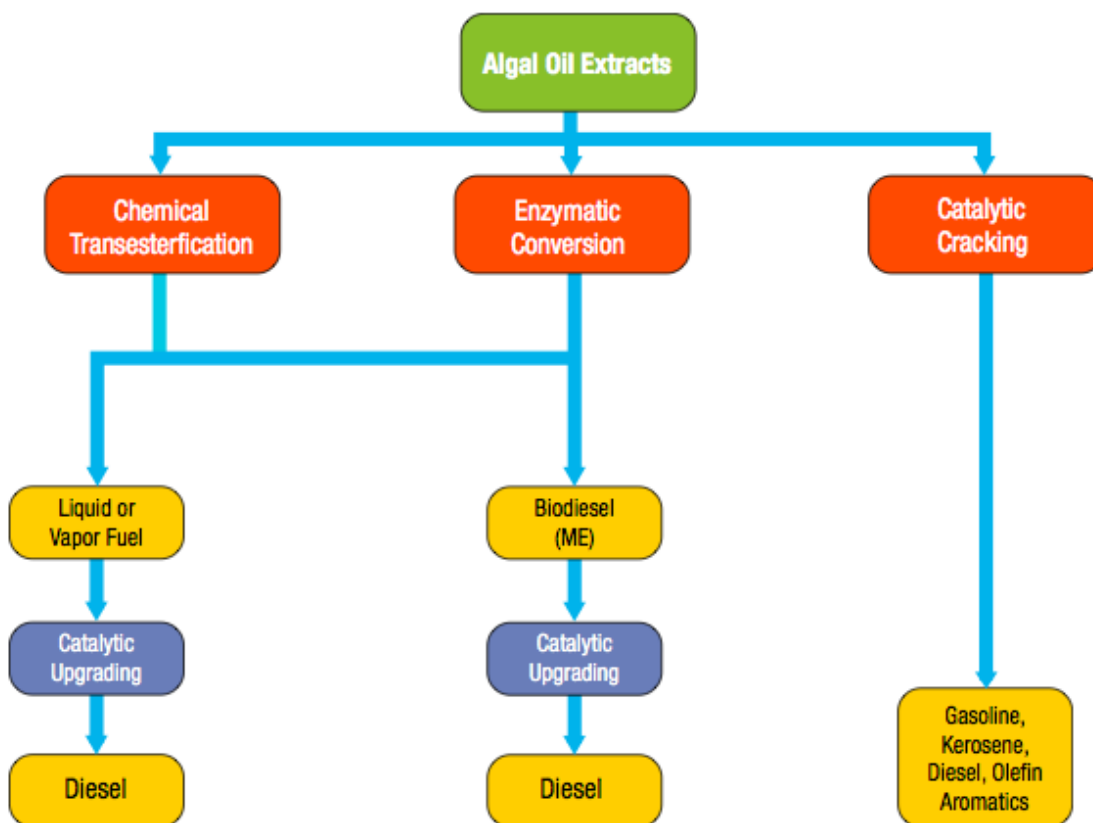


Fig. 25. Schematic of the various conversion strategies of algal extracts into biofuels (DOE, 2010).

The transesterification reaction is employed to convert triacylglycerols extracted from algae to FAMES (fatty acid methyl esters). Transesterification can be performed via catalytic or non-catalytic reaction systems using different heating systems that are required to initiate the reaction. This technology is relatively mature and it is considered the standard technology in

the conversion of vegetable oils into biodiesel. Chemical processes give high conversion of triacylglycerols to their corresponding esters but have drawbacks such as being energy-intensive and glycerol and alkaline catalyst impurities in the product that have to be removed. There are several processing variants that appear promising. An alternative heating system that enhances the kinetics of transesterification involves the use of microwaves, a cost-competitive option with shorter reaction times. Another solution is the ultrasonic reactor method, where ultrasonic waves cause the reaction mixture to produce and collapse bubbles constantly. This cavitation provides simultaneously the mixing and heating required to carry out the transesterification process. Thus using an ultrasonic reactor for biodiesel production drastically reduces the reaction time, reaction temperatures, and energy input (DOE, 2010). The use of biocatalysts (lipases) in transesterification of triacylglycerols for biodiesel production addresses the impurities problems of chemical transesterification and offers an environmentally more attractive option to the conventional processes (Svensson and Adlercreutz, 2008). Although enzymatic approaches have become increasingly attractive, they have not been demonstrated at large scale mainly due to the relatively high price of lipase.

3. Final thoughts.

As noted in the first paragraph, all the biofuels must meet a multitude of performance specifications that include volatility, initial and final boiling point, autoignition characteristics (as measured by octane number or cetane number) etcetera. Although the predominant feedstock for the industry is crude oil, the oil industry has begun to cast a wider net and has spent a great deal of resources developing additional inputs such as oil shale and tar sands (DOE, 2010).

Gasoline, jet fuel, and diesel are generally described as “renewable” or “green” if they are derived from a biological feedstock, such as biomass or plant oil, but have essentially the same performance specifications as the petroleum-based analog. A major characteristic of petroleum-derived

fuels is high energy content which is a function of oxygen content (the lower is the oxygen



Fig. 26. From a refinery's perspective, the ideal biofuel conversion process would make use of those operations already in place (DOE, 2010).

content the higher is the energy). Typical biological molecules have very high oxygen contents as compared to crude oil. Conversion of biological feedstocks to renewable fuels, therefore, is largely a process of eliminating oxygen and maximizing the final energy content. From a refinery's perspective, the ideal conversion process would make use of those operations already in place. In this way, the feedstock is considered fungible with petroleum and can be used for the production of typical fuels without disruptive changes in processes or infrastructure.

CHAPTER 6

Techno-Economic analysis.

1. Studied scenarios.

1.1. Achievable productivity.

The achievable productivity of open ponds and photobioreactors has been discussed in chapter 3.

The critically importance of productivity was underlined for the first time in an analysis by Benemann et Al., 1982. In this analysis sensitivities were run for 13 resources (such as power costs and evaporation), 15 facility design parameters (such as culture depth and mixing), three biological parameters (such as growing season) and eight financial parameters (cost escalations, etc.). The major conclusion was that no single parameter dominated costs sufficiently to achieve the goal of low-cost fuel production. But, as a central conclusion, productivity was the most important parameter: increasing production efficiencies by a factor of about four decreased production costs by almost half.

The prospective of this analysis is that a high productivity (and an high oil yield) is necessary to achieve an economically feasible biofuel production. We take under consideration productivity proposed by scientific researches and claimed by and commercial publications to date. These productivities has been proved in laboratories but it is not certain if they can be effectively reached in large systems; on the contrary we are pretty confident that the system would need an high level of control and optimization, especially during the first period of operation. However, this analysis wants to explore the potential of the technology in the medium period (3-5 years) and it is totally reasonable that, in few years, large systems will be implemented and the emerged problems will have been addressed.

Nowadays industrial secrets abound with very little published numbers available. The productivity assumptions on this paper follow the last achieved and published results.

The achievable microalgae productivity is not endless; on the contrary it is limited by different factors. These factors are well known (Fig. 27)

$$P_a = \frac{\tau \varepsilon_a \dot{E}_s}{E_a}$$

| | | |
|-----------------|----------------------------|---|
| P_a | $\text{kg/m}^2 \text{ yr}$ | <i>Microalgae production rate</i> |
| τ | | <i>Efficiency of light transmission to microalgae</i> |
| ε_a | | <i>Efficiency of conversion of incident sunlight to biomass in microalgae</i> |
| E_s | kW/m^2 | <i>Solar irradiance</i> |
| E_a | kJ/g | <i>Energy content of microalgae</i> |

Fig. 27. Microalgae production rate (Zemke et Al., 2008).

In order to understand the maximum quantity of obtainable biofuels, it is necessary to correct the microalgae production rate. If we take into consideration biodiesel, derived from algal oil, its maximum productivity would be indicated by the useable lipids rate of production. This rate is equal to the microalgae production rate (P_a) corrected by the dry mass microalgae lipid content useable for conversion to biodiesel and by the density of these lipids (Fig. 28). In our analysis a lipid extraction efficiency of 95% is also considered, because of the potential inefficiency of extraction technologies.

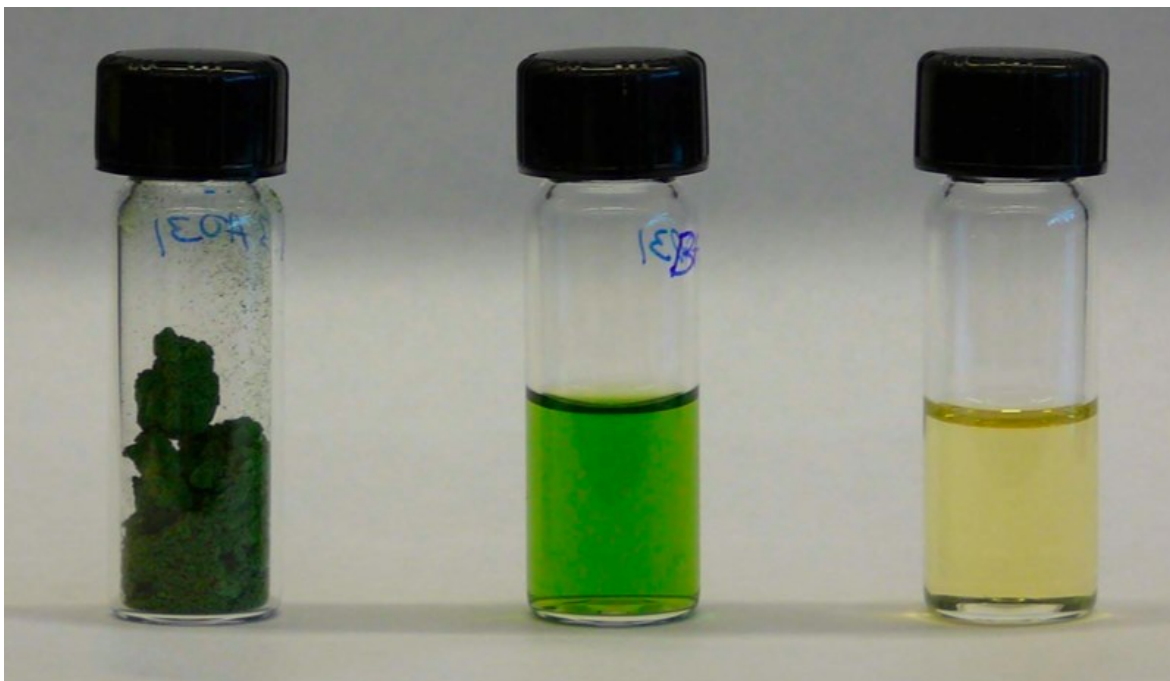


Fig. 28. From left to right, microalgae dry biomass, algal oil and biodiesel. In order to calculate the achievable amount of biodiesel, the microalgae production rate needs to be corrected by the % of useable lipids (Zemke et Al., 2008).

The theoretical maximum production is 43 l/(m² yr) (Zemke et Al., 2008). This value is influenced by solar irradiance and is an average for the United States. However, we take it as reference value.

The oil production of AlgaeLink's photobioreactors equates to 153,3 t/(ha yr) oil produced, (with an oil concentration in algae of 42% and 365 days of work). The density of algae oil is similar to that of soybean oil and is approximately 918 kg/m³ (Oilgae, 2011). So the AlgaeLink oil production is equal to 16,7 l/(m² yr). The technology for oil production from microalgae has reached the 38% of the theoretical maximum production, assuming AlgaeLink's technology as example of the industry state of art.

Concerning open ponds, productivity values close to 100 t/(ha yr) in biomass are reported both by research papers (Richmond (2004) reported obtainable values of 70-90 t/(ha yr) in perfectly managed pond; Richardson et Al. (2009) reported values up to 109,5 t/(ha yr); Hu et Al. (2008) reported an optimistic average productivity of 50 g/(m² day), equals to 182 t/(ha yr)) and by commercial publications (PetroAlgae (2011) claims that his open bioreactors have an overall biomass yield between 80 and 100 t/(ha yr) while AlgaeAtWork (2011) claims a projected 2012 overall biomass yield of 55 g/(m² day), equals to more than 200 t/(ha yr), even if AlgaeAtWork's system uses pond closed by plastic layers and it is not totally comparable with the traditional open pond design). If we assume a productivity of 100 t/(ha yr), with an oil concentration in algae of 42% we obtain 4,6 l/(m² yr), or the 10,3% of the theoretical maximum production. It is important to underline that an oil concentration of 42% is easier to obtain in closed controlled systems (photobioreactors) than in open ponds.

1.2. Analyzed scenarios.

The lack of mass cultivations creates a high uncertain around the achievable microalgae productivity and oil content. Data are mainly derived from laboratory results or small test plants. In order to address this uncertain, our choice is to look at different scenarios, to better understand how different productivities and oil yields can influence the economic results. In details, we consider 3 different productivities (20, 25 and 30 g/(m² day)) and 3 different oil yields (30%, 40% and 50%) for open ponds (9 scenarios, Table 9) and 3 different oil yields for a photobioreactor with high productivity.

Table 9. Different studies scenarios for the techno-economic analysis of biofuels production from microalgae in open pond systems.

| Scenario | A | B | C | D | E | F | G | H | I |
|---|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| Biomass Production [g/(m ² day)] | 20 | 25 | 30 | 20 | 25 | 30 | 20 | 25 | 30 |
| Days of Work | 365 | | | 365 | | | 365 | | |
| Production [t/(ha yr)] | 73 | 91,25 | 109,5 | 73 | 91,25 | 109,5 | 73 | 91,25 | 109,5 |
| Oil Content | 30% | | | 40% | | | 50% | | |
| Biomass Production [t/(ha yr)] | 51,10 | 63,88 | 76,65 | 43,80 | 54,75 | 65,70 | 36,50 | 45,63 | 54,75 |
| Oil Density [t/m ³] | 0,918 | | | 0,918 | | | 0,918 | | |
| Oil extraction Efficiency | 95% | | | 95% | | | 95% | | |
| Oil Production [l/(m ² yr)] | 2,27 | 2,83 | 3,40 | 3,02 | 3,78 | 4,53 | 3,78 | 4,72 | 5,67 |
| Oil Production [barrels/(ha yr)] | 142,55 | 178,18 | 213,82 | 190,06 | 237,58 | 285,09 | 237,58 | 296,97 | 356,37 |

For the photobioreactors, we assumed a biomass productivity of 1 t/(ha day), with oil content of 35%, 45% and 55%.

2. Relevant assumptions.

Our aim is to use microalgae for the production of transportation fuels.

We will analyze open ponds and photobioreactors in two different moments. First we concentrate on open systems and then we will focus on PBRs.

Because the fuel market is enormous and economies of scale are relevant in the used process (especially for downstream treatments, centrifugation and oil extractions technologies above all) we studied an open pond facility of large dimensions. We know that scale-up is challenging and requires important investments. Some images could help to understand the differences between the reached scale-up in laboratories (Fig. 29) and the dimensions that mass cultivation of fuels through microalgae would need (Fig. 30).

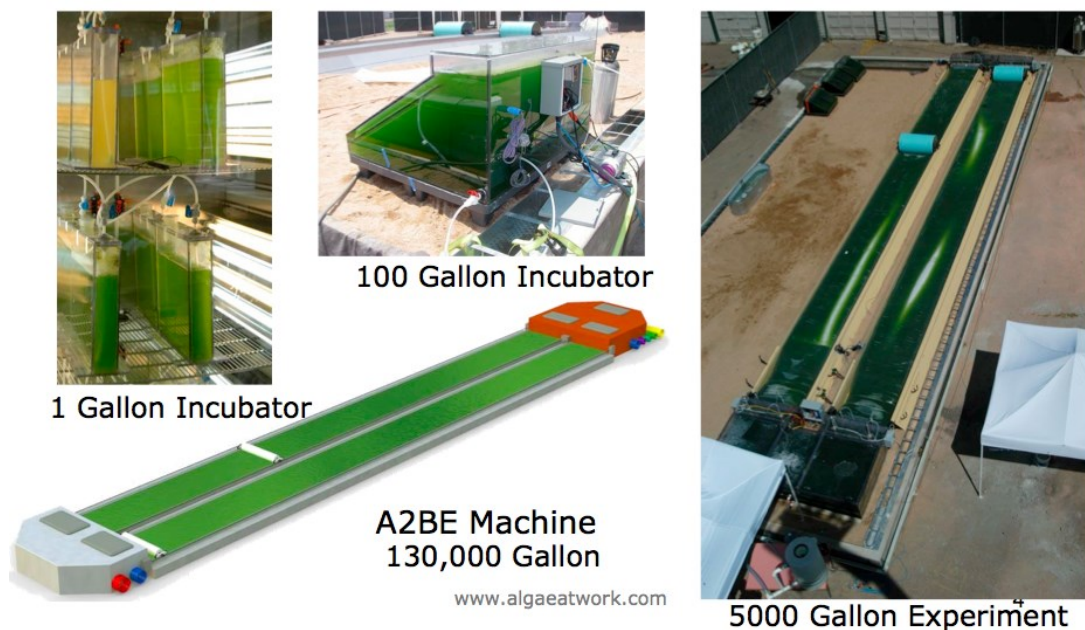


Fig. 29. Scale-up from 1-gallon incubator to a 130000 gallons machine (Algaeatwork, 2011).



Fig. 30. Simulation of a mass cultivation open system (Algaeatwork, 2011).

In this work, raceway ponds equipped with a low-level mixing system (velocity of 15 cm/s) will be considered. Technical and economic aspects of microalgae mass production in such recirculation ponds are largely based on the techno-economic analysis carried out by Benemann and Oswald (1996).

Revenues and costs are considered for a dimension of 1, but costs are estimated for a very large facility (100 ha or more, as seen in Fig. 30). The values for the costs of algae biomass production reported by Benemann and Oswald (1996) were adjusted for this economic analysis, taking into account inflation at an interest rate of 2.5% for 14 years (from 1997 to 2011). It was assumed that other costs considered here have not changed. The currency chosen is Euros (€) and the currency rate exchange applied is US \$1 = €0.7.

We propose the following adjustments:

- Capital costs related with the land are not taken into account since they would be very different, depending on the site location. As alternative approach we consider the European average cost of using low value agricultural land, equal to 100 €/year ha) (Eurostat, 2009). For every hectare we doubled the cost and considered 200 €/year, for two reasons. First it may be difficult to find low value land close to power plants. Secondly, the facility will require extra space in addition to the used for ponds; buildings, roads, harvesting and extraction machineries etc. will all require extra land. However past techno-economic analyses considered the required extra

space as a small fraction of the total facility size. Fig. 31 shows the layout designed by Benemann and Oswald for a 121 ha facility, where over 100 ha were occupied by microalgae cultivation.

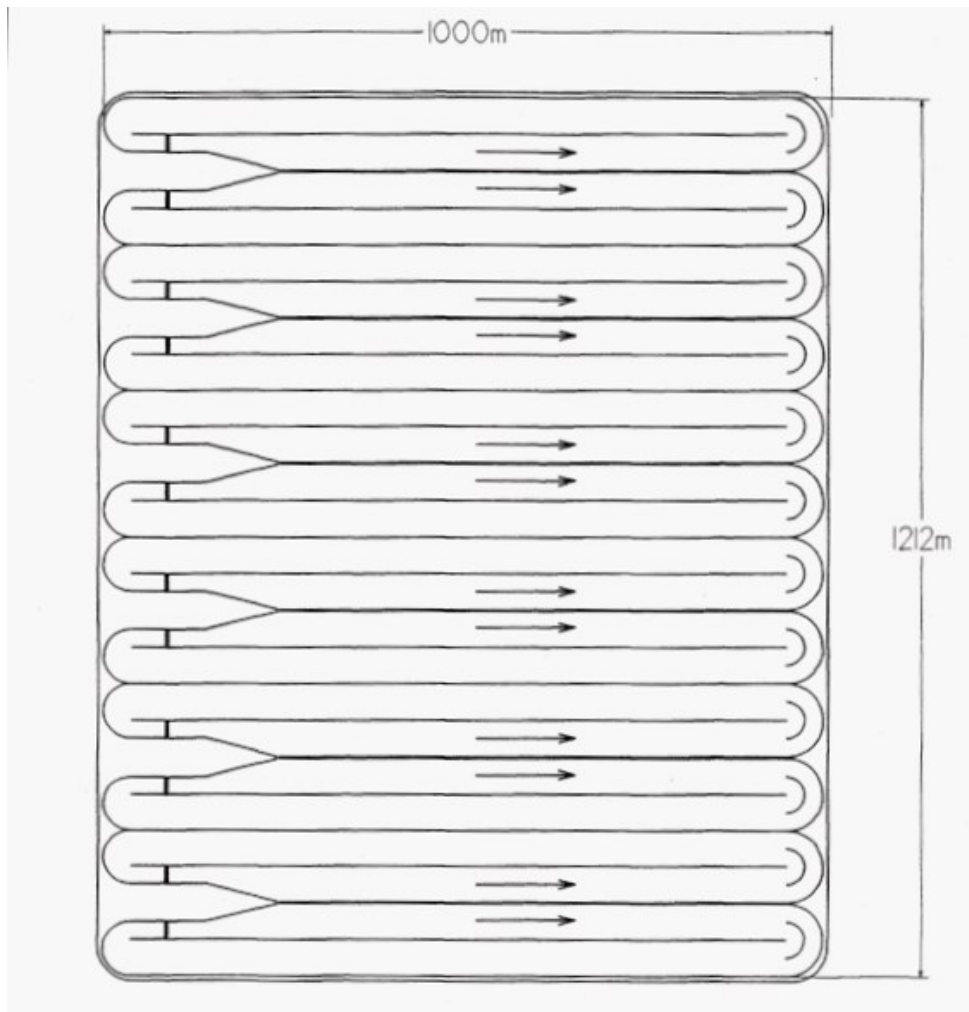


Fig. 31. Open pond large facility design (Benemann and Oswald, 1996)

- For the CO₂ supply, Benemann and Oswald (1996) hypothesized two options; using flue gas from a power plant located 2,5 km from the algae farm or using pure CO₂ supplied by a farm from tank storages. We consider flue gas generated on-site, with no costs for CO₂. Costs for flue gas supply, distribution and pumping for an average of 2 km is calculated. These costs are taken from Benemann and Oswald (1996), even if their described CO₂ piping system is relatively simple and inexpensive. A deeper analysis on these costs may be needed.
- Benemann and Oswald (1996) considered a three stage harvesting process: primary settling with flocculants, dissolved air flotation (DAF) and centrifugation. In our analysis, the cost of these processes is adjusted considering an improved technology that allows a 15% reduction of capital cost and of energy demand. The details of the harvesting first two steps are described in table 10, where volumes are for each ha.

We forecasted a maximum productivity of 45 g/(m² day) (50% bigger than 30 g/(m² day), the biggest productivity in our open system scenarios) because of the variability that productivity could have in different seasons. It is also assumed that 50% of pond's volume is harvested per day (pond's retention time of 2 days); from an initial output of 1000 m³, only 10 m³ are the output of the second harvesting step.

Table 10. Main details for the two first harvesting operations, open ponds.

| HARVESTING OPERATIONS | |
|---|-------|
| Pond depth [m] | 0,2 |
| Total culture volume [m ³] | 2000 |
| Retention time [days] | 2 |
| Maximum productivity [g/(m ² day)] | 45 |
| 1st harvesting system (settling): | |
| Achieved fold concentration | 20 |
| Achieved solid concentration | 0,9% |
| Output volume [m ³] | 50 |
| Output liters | 50000 |
| 2nd harvesting system (DAF): | |
| Achieved fold concentration | 5 |
| Achieved solid concentration | 4,5% |
| Output volume [m ³] | 10 |
| Output liters | 10000 |

- The extraction phase is done by centrifugation. Centrifugation can be used not only to concentrate the biomass but also to extract the algal lipids into an oil phase in a simultaneous operation. This is because of the relatively large difference in the densities of the water, algal lipids and other biomass constituents (Benemann and Oswald, 1996). This concept is based on the commercial process for the extraction of beta-carotene from flocculated algal biomass by a hot oil extraction process. Thus, the harvesting and processing steps overlap, with the flocculation and centrifugation steps being compatible with the oil extraction process. It should be pointed out that this is based on well-known technology, although the present application represents a large scale-up. A large three phase self-cleaning centrifuge that continuously expels three fractions (oil, biomass cake and water) is used. One important issue is the efficiency of extraction. Based on commercial experience with beta-carotene, where an almost quantitative recovery (98% or more) is obtained, 95% extraction efficiency can be a reasonable assumption.
- The energy demand for mixing is taken from Zamalloa et Al. (2010). The design (Fig. 32) and cost of paddle wheels for mixing and of flue gas sumps diffusers is taken from Benemann and Oswald (1996) and adjusted considering an improved technology that allows a 15% cost reduction.

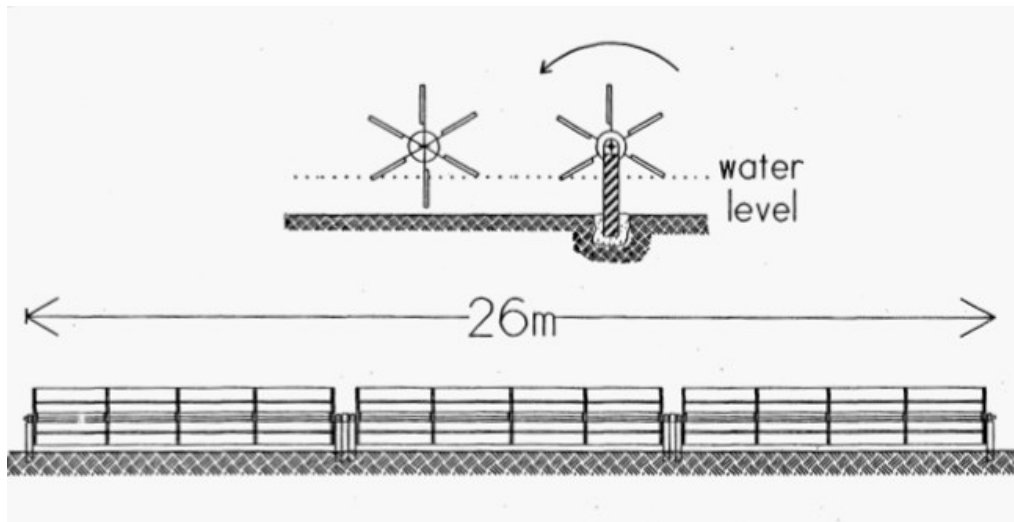


Fig. 32. Paddle wheels design (Benemann and Oswald, 1996)

- It is assumed that the capital cost of the algae pond is not dependent on productivity. In addition we calculate the operational costs for a productivity of 30 g/(m² day). This represents the maximum productivity in our open ponds scenarios; from this point of view we are quite conservative on nutrient supply and flocculants costs.
- Benemann and Oswald (1996) utilized deep brackish groundwater for the algae farms. For our model, it is assumed that the water can be either taken from the sea or that the effluent of a nearby wastewater treatment plant will be used in the ponds, thus making the costs related with water supply lower (they are mainly piping costs).
- The nutrient cost is calculated assuming a 75% recycle of N and 50% recycle of P.
- We assume 365 days of operation, because the cultivation in ponds is a continue process. However this will require optimum weather conditions such as a mild winter and low climate variations between day and night.
- Industrial consumption and price of electricity vary greatly. For this reason it is difficult to estimate the cost of electricity. We classified our plant as a medium industrial consumer, and evaluated the cost of energy from the data reported in Europe's Energy Portal (2011). In detail, Europe's Energy Portal reports two different consumption patterns (small and large customers) with the pricing structure for each member state. Because the location is extremely important for algal cultivation, we took the price of Spain and Italy (probably the two better locations for cultivate algae in Europe) and calculated an average between their energy costs (Table 11).

Table 11. Average of industrial electricity rates in Italy and Spain.

| Industrial electricity rates [€/kWh] | | | |
|--------------------------------------|--------|--------|---------------|
| Consumption | Small | Large | Average |
| Spain | 0,1082 | 0,0808 | 0,0945 |
| Italy | 0,1224 | 0,1009 | 0,1117 |
| Average, Spain and Italy | | | 0,1031 |

3. Costs.

The electricity cost in our analysis is 0,1031 €/kWh (Table 11). The total energy cost will be taken into account in the operational costs. In table 12 the total energy demand (for 1 ha) is calculated, while table 13 shows the total energy costs per hectare.

Table 12. Energy demand for open pond systems, 1 ha.

| ENERGY DEMAND | [kWh/(day ha)] | |
|----------------------------------|----------------|--|
| Mixing (paddle wheel) | 20 | (Zamalloa et Al., 2010) |
| Harvesting (settling, DAF) | 4,1 | (Benemann and Oswald, 1996, reduced by 15%) |
| Centrifugation | 13,3 | (Benemann and Oswald, 1996, reduced by 15%) |
| Water pumping | 13,3 | (Benemann and Oswald, 1996, reduced by 15%) |
| Flue gas pumping | 25 | Distance from a CO ₂ source: 2 km |
| Other | 4 | |
| Total daily energy demand | 79,70 | |

Table 13. Energy costs for open pond systems, 1 ha.

| ENERGY COSTS | |
|--|----------------|
| Total daily energy demand [kWh/(day ha)] | 79,70 |
| Days of operation | 365 |
| Total yearly energy demand [MWh/(ha yr)] | 29,09 |
| Industrial electricity rates [€/kWh] | 0,1031 |
| Daily energy cost [€/ha] | 8,22 |
| Yearly energy cost [€/ha] | 2998,50 |

Capital costs for an open ponds facility are reported in table 14. In addition to the costs, the source of the data is reported. Costs are divided in costs for algae production and costs for downstream processes. The total plant cost is equal to 48578 €/ha.

Table 14. Capital costs for open pond systems.

| CAPITAL COSTS | [€/ha] | |
|--|--------------|---|
| Algae Production | | |
| Pond levees, geotextiles | 3461 | (Benemann and Oswald, 1996) |
| Site preparation, grading, compacting | 2473 | (Benemann and Oswald, 1996) |
| Mixing (paddle wheels) | 4203 | (Benemann and Oswald, 1996, reduced by 15%) |
| Flue gas sumps diffusers | 4203 | (Benemann and Oswald, 1996, reduced by 15%) |
| Flue gas supply, distribution | 297 | (Benemann and Oswald, 1996) |
| Water and nutrient supply, distribution | 3066 | (Benemann and Oswald, 1996) |
| Electrical costs for construction | 1978 | (Benemann and Oswald, 1996) |
| Buildings, roads, drainage | 1978 | (Benemann and Oswald, 1996) |
| Instrumentation and machinery | 495 | (Benemann and Oswald, 1996) |
| Downstream Treatment | | |
| Flocculation and dissolved air flotation | 7567 | (Benemann and Oswald, 1996, reduced by 15%) |
| Centrifugation and oil extraction | 10509 | (Benemann and Oswald, 1996, reduced by 15%) |
| Direct cost of algae production and treatment | | |
| Engineering, design, supervision and management overhead | 6035 | 15% of above |
| Install plant costs | 46265 | |
| Contingency | 2313 | 5% of the Install plant costs |
| Total plant costs | 48578 | |

Operational costs for an open ponds facility are reported in table 15. In addition to the costs, the source of the data is reported. The operational cost is equal to 11462 €/ha yr).

Both capital and operational costs are constant in the different analyzed scenarios.

Table 15. Operational costs for open pond systems.

| OPERATIONAL COSTS | [€/ha yr] | |
|--------------------------------|--------------|--------------------------------------|
| Land cost | 200 | (Eurostat, 2009) |
| Power costs | 2999 | (Benemann and Oswald, 1996) |
| Nutrient supply | 890 | (Benemann and Oswald, 1996) |
| Flocculants | 989 | (Benemann and Oswald, 1996) |
| CO ₂ | 0 | Supposed free, see related paragraph |
| Waste disposal | 989 | (Benemann and Oswald, 1996) |
| Maintenance | 2429 | 5% of total plant costs |
| Labor and overheads | 2967 | (Benemann and Oswald, 1996) |
| Total operational costs | 11462 | |

4. Revenues.

The crude oil price for barrel is the reference for understanding the price that a barrel of algal oil would have. Algal oil has lower energy content than crude oil (Table 8) and the difference is in the order of 7%. The price of algal oil has to take this difference under consideration. The crude oil reference price is 87 \$/barrel (February, 2011). The algal oil price in our analysis is 56,64 €/barrel (Table 16).

The biomass price for an open pond facility is 105 €/t and estimated as showed in table 17. It is important to underline that the biomass prices in table 17 are quite conservative (the source is the DOE, 2010). The aim of our analysis is to verify the economical feasibility of producing biofuels from microalgae, for this reason we assumed conservative prices and do not address the high value co-product market. However, we will analyze the economic potentials of co-products in the next paragraphs, in order to enhance the productivity of the project.

Table 16. Algal oil and biomass prices, open ponds.

| PRICE OF OIL AND BIOMASS FROM MICROALGAE | |
|--|-------|
| Crude oil price [\$/barrel] | 87 |
| Crude oil price [€/barrel] | 60,9 |
| Reduction in energy content | 7% |
| Algal oil price [€/barrel] | 56,64 |
| Biomass price - open pond [€/t] | 105 |

Table 17. Biomass different uses and prices, open ponds.

| Biomass use | Price [€/t] | Use % |
|-----------------------|-------------|-------|
| Anaerobic digestion | 75 | 60% |
| High protein food | 150 | 40% |
| High value co-product | 500 | 0% |

Table 18 presents the revenues for the different studied scenarios. Most of the revenues come from the algal oil. The higher are the productivity and the oil content, the higher are the revenues.

Table 18. Total revenues for the different examined scenarios.

| Scenario | A | B | C | D | E | F | G | H | I |
|---|----------|----------|----------|----------|----------|----------|----------|----------|----------|
| Biomass Production [g/(m ² day)] | 20 | 25 | 30 | 20 | 25 | 30 | 20 | 25 | 30 |
| Oil Content | 30% | | | 40% | | | 50% | | |
| Revenue from oil | € 8.073 | € 10.092 | € 12.110 | € 10.765 | € 13.456 | € 16.147 | € 13.456 | € 16.820 | € 20.183 |
| Revenue from biomass | € 5.366 | € 6.707 | € 8.048 | € 4.599 | € 5.749 | € 6.899 | € 3.833 | € 4.791 | € 5.749 |
| Total revenues | € 13.439 | € 16.799 | € 20.158 | € 15.364 | € 19.204 | € 23.045 | € 17.288 | € 21.610 | € 25.932 |

5. Economic analysis – feasibility of scenarios.

In this first part of the economic analysis, we studied all the 9 different scenarios using a quite simplified method. We calculate an interest to the debit (initial investment) and we subtract all the operational profits from the debit of the year.

The main purpose was to estimate when the investment reach the breakeven (calculate the payback period) with a timeline of ten years. We did not take into account a longer period because, in most of the circumstances, investors do not found attractive projects with a long payback period.

To generate insight in the profitability of oil (biofuels) production through the cultivation of algae, not only the payback period of the project was calculated, but also the net present value (NPV) in tandem with the internal rate of return (IRR). The use of (NPV) and (IRR) will be more insightful in the next part of the economic analysis, net profit and loss account.

5.1. Economic indicators.

- The NPV indicates whether the project is profitable, taking into account the time value of the cash flows such as revenue streams, capital investments and operational costs. NPV compares the value of an amount today to the value of that same amount in the future, taking inflation and returns into account. In our economic analysis the inflation rate is equal to 2,5%.
- The IRR is the discount rate that produces a zero NPV. The higher a project's internal rate of return, the more desirable it is to undertake the project. As such, IRR can be used to rank several prospective projects a firm is considering. Assuming all other factors are equal among the various projects, the project with the highest IRR would probably be considered the best and undertaken first. The IRR could also be viewed as indicator of the rate of growth a project is expected to generate.
- The payback period refers to the number of years it takes to generate enough revenues to pay the investment back, without taking into account the time value of money. All other things being equal, the better investment is the one with the shorter payback period. However, it ignores any benefits that occur after the payback period and, therefore, does not measure profitability.

5.2. Interest rate.

The interest rate used is 6%, compounded annually. The interest rate is an indicator of the weighted average cost of capital, not taking into concern the source of the used capital. In this first analysis the rate could be both the interest applied in a bank loan or the Minimum Acceptable Rate of Return (MARR) of the equity (the amount of the funds contributed by the owners) that financed the project. In the next analysis the interest rate will be linked to a bank loan.

There are different interest rate options for investors. The rate is chosen equal to 6% because this value is a good approximation of a variable interest rate related to the EURIBOR Rate (EURO Inter Bank Offered Rate) plus the bank's spread. Furthermore the 6%

rate takes into account a financial insurance of the maximum future amount that the EURIBOR could reach. From this point of view, it is acceptable to assume the rate fixed and compounded annually, because the insurance avoids the rate to be higher than a fixed maximum cap (in this case the cap is close to 6%). The rate is estimated for a large investment of 10 M€ (approximately 200 ha of open ponds), with equity of 20% and a financial plan of 10 years.

5.3. Results of the preliminary economic analysis.

Table 19. Results of the preliminary economic analysis, open pond system, 9 scenarios. The color indicates the feasibility.

| Scenario | A | B | C | D | E | F | G | H | I |
|---------------------------------|----------|----------|----------|----------|----------|----------|----------|----------|----------|
| Biomass Production [g/(m2 day)] | 20 | 25 | 30 | 20 | 25 | 30 | 20 | 25 | 30 |
| Oil Content | | 30% | | | 40% | | | 50% | |
| Revenue from oil | € 8.073 | € 10.092 | € 12.110 | € 10.765 | € 13.456 | € 16.147 | € 13.456 | € 16.820 | € 20.183 |
| Revenue from biomass | € 5.366 | € 6.707 | € 8.048 | € 4.599 | € 5.749 | € 6.899 | € 3.833 | € 4.791 | € 5.749 |
| Total revenues | € 13.439 | € 16.799 | € 20.158 | € 15.364 | € 19.204 | € 23.045 | € 17.288 | € 21.610 | € 25.932 |
| Operational profits | € 1.976 | € 5.336 | € 8.696 | € 3.901 | € 7.742 | € 11.583 | € 5.826 | € 10.148 | € 14.470 |
| Feasibility | | | | | | | | | |
| Net present value | - | - | € 20.849 | - | € 11.569 | € 47.815 | - | € 34.536 | € 73.992 |
| Payback period | - | - | 8 years | - | 9 years | 5 years | - | 6 years | 4 years |
| IRR | - | -6,5% | 8,0% | -6,5% | 4,7% | 16,5% | -3,8% | 12,4% | 23,9% |

In table 19 the results of the analysis are reported, with indication about the feasibility (color) and economic results (NPV, payback period and IRR). In the scenarios A, B, D and G, the operational profits were not sufficient to pay back the initial investment (capital costs), the interest accumulated and the operational costs within ten years. The scenario G will have a deeper discussion in the next part of the analysis, because with some assumption could have a positive NPV. The first scenario (A) is the worst one, the debit increase year after year due to the profits, which are lower than the interests (Table 20).

The other scenarios are all positive. However, scenarios C and E have a rather long payback period and this could really influence the approach of potential investors. We will exclude scenarios A, B and D from the second part of the analysis.

Next we propose a summary of the analysis for the different cases. Revenues, operational costs, EBITDA (Earnings Before Interest, Taxes, Depreciation and Amortization), interests at a 6% rate, net result before taxes, accumulated cash flow, NPV, payback period and IRR are presented for each scenario (Table 20 – Table 28).

Table 20. Results for the scenario A, with a production of 20 g/(m² day) and oil content of 30%.

| Scenario A | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------------|------|-----------|-------------------|----------|-----------|-----------|----------------|-------------------|-----------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | - |
| Biomass Production | 1 | € 13.439 | -€ 11.462 | € 1.976 | -€ 2.915 | -€ 49.516 | € - | -€ 49.516 | |
| 20 [g/(m² day)] | 2 | € 13.439 | -€ 11.462 | € 1.976 | -€ 2.971 | -€ 50.510 | € - | -€ 50.510 | Payback period |
| | 3 | € 13.439 | -€ 11.462 | € 1.976 | -€ 3.031 | -€ 51.564 | € - | -€ 51.564 | - |
| Oil Content 30% | 4 | € 13.439 | -€ 11.462 | € 1.976 | -€ 3.094 | -€ 52.682 | € - | -€ 52.682 | |
| | 5 | € 13.439 | -€ 11.462 | € 1.976 | -€ 3.161 | -€ 53.866 | € - | -€ 53.866 | IRR |
| | 6 | € 13.439 | -€ 11.462 | € 1.976 | -€ 3.232 | -€ 55.122 | € - | -€ 55.122 | - |
| | 7 | € 13.439 | -€ 11.462 | € 1.976 | -€ 3.307 | -€ 56.453 | € - | -€ 56.453 | |
| | 8 | € 13.439 | -€ 11.462 | € 1.976 | -€ 3.387 | -€ 57.863 | € - | -€ 57.863 | |
| | 9 | € 13.439 | -€ 11.462 | € 1.976 | -€ 3.472 | -€ 59.359 | € - | -€ 59.359 | |
| | 10 | € 13.439 | -€ 11.462 | € 1.976 | -€ 3.562 | -€ 60.944 | € - | -€ 60.944 | |
| | | € 134.389 | -€ 114.624 | € 19.765 | -€ 32.131 | | € - | -€ 60.944 | |

Table 21. Results for the scenario B, with a production of 25 g/(m² day) and oil content of 30%.

| Scenario B | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------------|------|-----------|-------------------|----------|-----------|-----------|----------------|-------------------|-----------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | € - |
| Biomass Production | 1 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.915 | -€ 46.156 | € - | -€ 46.156 | |
| 25 [g/(m² day)] | 2 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.769 | -€ 43.589 | € - | -€ 43.589 | Payback period |
| | 3 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.615 | -€ 40.868 | € - | -€ 40.868 | - |
| Oil Content 30% | 4 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.452 | -€ 37.984 | € - | -€ 37.984 | |
| | 5 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.279 | -€ 34.927 | € - | -€ 34.927 | IRR |
| | 6 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.096 | -€ 31.687 | € - | -€ 31.687 | -6,5% |
| | 7 | € 16.799 | -€ 11.462 | € 5.336 | -€ 1.901 | -€ 28.252 | € - | -€ 28.252 | |
| | 8 | € 16.799 | -€ 11.462 | € 5.336 | -€ 1.695 | -€ 24.611 | € - | -€ 24.611 | |
| | 9 | € 16.799 | -€ 11.462 | € 5.336 | -€ 1.477 | -€ 20.751 | € - | -€ 20.751 | |
| | 10 | € 16.799 | -€ 11.462 | € 5.336 | -€ 1.245 | -€ 16.660 | € - | -€ 16.660 | |
| | | € 167.986 | -€ 114.624 | € 53.362 | -€ 21.444 | | € - | -€ 16.660 | |

Table 22. Results for the scenario C, with a production of 30 g/(m² day) and oil content of 30%.

| Scenario C | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------------|------|-----------|-------------------|----------|-----------|-----------|----------------|-------------------|-----------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | € 20.849 |
| Biomass Production | 1 | € 20.158 | -€ 11.462 | € 8.696 | -€ 2.915 | -€ 42.796 | € - | -€ 42.796 | |
| 30 [g/(m² day)] | 2 | € 20.158 | -€ 11.462 | € 8.696 | -€ 2.568 | -€ 36.668 | € - | -€ 36.668 | Payback period |
| | 3 | € 20.158 | -€ 11.462 | € 8.696 | -€ 2.200 | -€ 30.172 | € - | -€ 30.172 | 8 years |
| Oil Content 30% | 4 | € 20.158 | -€ 11.462 | € 8.696 | -€ 1.810 | -€ 23.287 | € - | -€ 23.287 | |
| | 5 | € 20.158 | -€ 11.462 | € 8.696 | -€ 1.397 | -€ 15.988 | € - | -€ 15.988 | IRR |
| | 6 | € 20.158 | -€ 11.462 | € 8.696 | -€ 959 | € 8.252 | € - | € 8.252 | 8,0% |
| | 7 | € 20.158 | -€ 11.462 | € 8.696 | -€ 495 | € 51 | € - | € 51 | |
| | 8 | € 20.158 | -€ 11.462 | € 8.696 | € 3 | € - | € 8.642 | € 8.642 | |
| | 9 | € 20.158 | -€ 11.462 | € 8.696 | € - | € - | € 8.696 | € 17.338 | |
| | 10 | € 20.158 | -€ 11.462 | € 8.696 | € - | € - | € 8.696 | € 26.034 | |
| | | € 201.583 | -€ 114.624 | € 86.959 | -€ 12.348 | | € 26.034 | € 26.034 | |

Table 23. Results for the scenario D, with a production of 20 g/(m² day) and oil content of 40%.

| Scenario D | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------------|------|-----------|-------------------|----------|-----------|-----------|----------------|-------------------|-----------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | € - |
| Biomass Production | 1 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.915 | -€ 46.156 | € - | -€ 46.156 | |
| 20 [g/(m² day)] | 2 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.769 | -€ 43.589 | € - | -€ 43.589 | Payback period |
| | 3 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.615 | -€ 40.868 | € - | -€ 40.868 | - |
| Oil Content 40% | 4 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.452 | -€ 37.984 | € - | -€ 37.984 | |
| | 5 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.279 | -€ 34.927 | € - | -€ 34.927 | IRR |
| | 6 | € 16.799 | -€ 11.462 | € 5.336 | -€ 2.096 | -€ 31.687 | € - | -€ 31.687 | -6,5% |
| | 7 | € 16.799 | -€ 11.462 | € 5.336 | -€ 1.901 | -€ 28.252 | € - | -€ 28.252 | |
| | 8 | € 16.799 | -€ 11.462 | € 5.336 | -€ 1.695 | -€ 24.611 | € - | -€ 24.611 | |
| | 9 | € 16.799 | -€ 11.462 | € 5.336 | -€ 1.477 | -€ 20.751 | € - | -€ 20.751 | |
| | 10 | € 16.799 | -€ 11.462 | € 5.336 | -€ 1.245 | -€ 16.660 | € - | -€ 16.660 | |
| | | € 167.986 | -€ 114.624 | € 53.362 | -€ 21.444 | | € - | -€ 16.660 | |

Table 24. Results for the scenario E, with a production of 25 g/(m² day) and oil content of 40%.

| Scenario E | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------------|------|-----------|-------------------|----------|-----------|-----------|----------------|-------------------|-----------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | € 11.569 |
| Biomass Production | 1 | € 19.204 | -€ 11.462 | € 7.742 | -€ 2.915 | -€ 43.750 | € - | -€ 43.750 | Payback period |
| 25 [g/(m² day)] | 2 | € 19.204 | -€ 11.462 | € 7.742 | -€ 2.625 | -€ 38.633 | € - | -€ 38.633 | |
| | 3 | € 19.204 | -€ 11.462 | € 7.742 | -€ 2.318 | -€ 33.209 | € - | -€ 33.209 | |
| Oil Content 40% | 4 | € 19.204 | -€ 11.462 | € 7.742 | -€ 1.993 | -€ 27.460 | € - | -€ 27.460 | IRR |
| | 5 | € 19.204 | -€ 11.462 | € 7.742 | -€ 1.648 | -€ 21.366 | € - | -€ 21.366 | |
| | 6 | € 19.204 | -€ 11.462 | € 7.742 | -€ 1.282 | -€ 14.906 | € - | -€ 14.906 | 4,7% |
| | 7 | € 19.204 | -€ 11.462 | € 7.742 | -€ 894 | -€ 8.058 | € - | -€ 8.058 | |
| | 8 | € 19.204 | -€ 11.462 | € 7.742 | -€ 483 | -€ 799 | € - | -€ 799 | |
| | 9 | € 19.204 | -€ 11.462 | € 7.742 | -€ 48 | € - | € 6.895 | € 6.895 | |
| | 10 | € 19.204 | -€ 11.462 | € 7.742 | € - | € - | € 7.742 | € 14.637 | |
| | | € 192.044 | -€ 114.624 | € 77.420 | -€ 14.206 | | € 14.637 | € 14.637 | |

Table 25. Results for the scenario F, with a production of 30 g/(m² day) and oil content of 40%.

| Scenario F | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------------|------|-----------|-------------------|-----------|-----------|-----------|----------------|-------------------|-----------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | € 47.815 |
| Biomass Production | 1 | € 23.045 | -€ 11.462 | € 11.583 | -€ 2.915 | -€ 39.910 | € - | -€ 39.910 | Payback period |
| 30 [g/(m² day)] | 2 | € 23.045 | -€ 11.462 | € 11.583 | -€ 2.395 | -€ 30.721 | € - | -€ 30.721 | |
| | 3 | € 23.045 | -€ 11.462 | € 11.583 | -€ 1.843 | -€ 20.982 | € - | -€ 20.982 | |
| Oil Content 40% | 4 | € 23.045 | -€ 11.462 | € 11.583 | -€ 1.259 | -€ 10.658 | € - | -€ 10.658 | IRR |
| | 5 | € 23.045 | -€ 11.462 | € 11.583 | -€ 639 | € - | € 286 | € 286 | |
| | 6 | € 23.045 | -€ 11.462 | € 11.583 | € - | € - | € 11.583 | € 11.869 | 16,5% |
| | 7 | € 23.045 | -€ 11.462 | € 11.583 | € - | € - | € 11.583 | € 23.452 | |
| | 8 | € 23.045 | -€ 11.462 | € 11.583 | € - | € - | € 11.583 | € 35.034 | |
| | 9 | € 23.045 | -€ 11.462 | € 11.583 | € - | € - | € 11.583 | € 46.617 | |
| | 10 | € 23.045 | -€ 11.462 | € 11.583 | € - | € - | € 11.583 | € 58.200 | |
| | | € 230.453 | -€ 114.624 | € 115.829 | -€ 9.051 | | € 58.200 | € 58.200 | |

Table 26. Results for the scenario G, with a production of 20 g/(m² day) and oil content of 50%.

| Scenario G | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------------|------|-----------|-------------------|----------|-----------|-----------|----------------|-------------------|-----------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | € - |
| Biomass Production | 1 | € 17.288 | -€ 11.462 | € 5.826 | -€ 2.915 | -€ 45.667 | € - | -€ 45.667 | Payback period |
| 20 [g/(m² day)] | 2 | € 17.288 | -€ 11.462 | € 5.826 | -€ 2.740 | -€ 42.581 | € - | -€ 42.581 | |
| | 3 | € 17.288 | -€ 11.462 | € 5.826 | -€ 2.555 | -€ 39.310 | € - | -€ 39.310 | |
| Oil Content 50% | 4 | € 17.288 | -€ 11.462 | € 5.826 | -€ 2.359 | -€ 35.843 | € - | -€ 35.843 | IRR |
| | 5 | € 17.288 | -€ 11.462 | € 5.826 | -€ 2.151 | -€ 32.168 | € - | -€ 32.168 | |
| | 6 | € 17.288 | -€ 11.462 | € 5.826 | -€ 1.930 | -€ 28.272 | € - | -€ 28.272 | -3,8% |
| | 7 | € 17.288 | -€ 11.462 | € 5.826 | -€ 1.696 | -€ 24.143 | € - | -€ 24.143 | |
| | 8 | € 17.288 | -€ 11.462 | € 5.826 | -€ 1.449 | -€ 19.765 | € - | -€ 19.765 | |
| | 9 | € 17.288 | -€ 11.462 | € 5.826 | -€ 1.186 | -€ 15.126 | € - | -€ 15.126 | |
| | 10 | € 17.288 | -€ 11.462 | € 5.826 | -€ 908 | -€ 10.207 | € - | -€ 10.207 | |
| | | € 172.881 | -€ 114.624 | € 58.257 | -€ 19.887 | | € - | -€ 10.207 | |

Table 27. Results for the scenario H, with a production of 25 g/(m² day) and oil content of 50%.

| Scenario H | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------------|------|-----------|-------------------|-----------|-----------|-----------|----------------|-------------------|-----------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | € 34.536 |
| Biomass Production | 1 | € 21.610 | -€ 11.462 | € 10.148 | -€ 2.915 | -€ 41.345 | € - | -€ 41.345 | Payback period |
| 25 [g/(m² day)] | 2 | € 21.610 | -€ 11.462 | € 10.148 | -€ 2.481 | -€ 33.678 | € - | -€ 33.678 | |
| | 3 | € 21.610 | -€ 11.462 | € 10.148 | -€ 2.021 | -€ 25.550 | € - | -€ 25.550 | |
| Oil Content 50% | 4 | € 21.610 | -€ 11.462 | € 10.148 | -€ 1.533 | -€ 16.936 | € - | -€ 16.936 | IRR |
| | 5 | € 21.610 | -€ 11.462 | € 10.148 | -€ 1.016 | -€ 7.804 | € - | € - | |
| | 6 | € 21.610 | -€ 11.462 | € 10.148 | -€ 468 | € - | € 1.876 | € 1.876 | 12,4% |
| | 7 | € 21.610 | -€ 11.462 | € 10.148 | € - | € - | € 10.148 | € 12.023 | |
| | 8 | € 21.610 | -€ 11.462 | € 10.148 | € - | € - | € 10.148 | € 22.171 | |
| | 9 | € 21.610 | -€ 11.462 | € 10.148 | € - | € - | € 10.148 | € 32.319 | |
| | 10 | € 21.610 | -€ 11.462 | € 10.148 | € - | € - | € 10.148 | € 42.467 | |
| | | € 216.102 | -€ 114.624 | € 101.478 | -€ 10.433 | | € 42.467 | € 42.467 | |

Table 28. Results for the scenario I, with a production of 30 g/(m² day) and oil content of 50%.

| Scenario I | Year | Revenues | Operational Costs | EBITDA | Interests | Debit | Net Result | Cumuled Cash Flow | Net present Value |
|-----------------------------|------|-----------|-------------------|-----------|-----------|-----------|----------------|-------------------|-------------------|
| | 0 | | | | | -€ 48.578 | (before taxes) | -€ 48.578 | € 73.992 |
| Biomass Production | 1 | € 25.932 | -€ 11.462 | € 14.470 | -€ 2.915 | -€ 37.023 | € - | -€ 37.023 | |
| 30 [g/(m ² day)] | 2 | € 25.932 | -€ 11.462 | € 14.470 | -€ 2.221 | -€ 24.774 | € - | -€ 24.774 | Payback period |
| | 3 | € 25.932 | -€ 11.462 | € 14.470 | -€ 1.486 | -€ 11.791 | € - | -€ 11.791 | 4 years |
| Oil Content 50% | 4 | € 25.932 | -€ 11.462 | € 14.470 | -€ 707 | € - | € 1.972 | € 1.972 | |
| | 5 | € 25.932 | -€ 11.462 | € 14.470 | € - | € - | € 14.470 | € 16.441 | IRR |
| | 6 | € 25.932 | -€ 11.462 | € 14.470 | € - | € - | € 14.470 | € 30.911 | 23,9% |
| | 7 | € 25.932 | -€ 11.462 | € 14.470 | € - | € - | € 14.470 | € 45.381 | |
| | 8 | € 25.932 | -€ 11.462 | € 14.470 | € - | € - | € 14.470 | € 59.851 | |
| | 9 | € 25.932 | -€ 11.462 | € 14.470 | € - | € - | € 14.470 | € 74.321 | |
| | 10 | € 25.932 | -€ 11.462 | € 14.470 | € - | € - | € 14.470 | € 88.791 | |
| | | € 259.322 | -€ 114.624 | € 144.698 | -€ 7.330 | | € 88.791 | € 88.791 | |

6. Economic analysis – net profit and loss account.

6.1. Assumptions.

This second part of the economic analysis is more complete; we take into account several additional aspects.

- Source of the used capital: it is common to finance investments with a bank loan. In order to obtain bank loans, the investors need to contribute with a part of the amount (equity). Furthermore “green” projects are eligible for government “green” grants. These grants depend on the location and on the state’s government. However the studied project has all the prerequisites (production of biofuels, CO₂ capture) for being considered “green”. Our assumption is to finance the project mainly with a bank loan (80% of the capital costs, equal to 38862 €). We do not make distinctions between the sources of the other 20%. If a 20% green grant contributes to the project, any NPV over zero are positive. Otherwise, with 20% equity, the NPV should be bigger than the 20% of the investment costs (9716 €).
- Taxation: taxation is calculated over the EBIT (Earning Before Interest and Taxes). It is directly associated to the plant location, as different countries have different taxes. In our analysis we consider Italian taxation, calculated as an IRAP taxation of 3,9% plus an IRES taxation of 27,5% (total taxation 31,4%). However, because Italian taxation is rather high, we also included in our analysis a lower taxation of 20%, as reference.
- Amortization: for financial reasons, the economy of the project can be enhanced when the initial capital costs are divided in several years, lowering the EBIT, and thus the paid taxes for those years. We considered an amortization period of 8 years.

6.2. Results of the net profit and loss account analysis.

In table 29 the results of the analysis are reported, with indication about the feasibility (color) and economic results (NPV, payback period and IRR). The scenarios A, B and D are not taken into concern in this part of the analysis, due to their not sufficient profitability, as

demonstrated earlier. The following discussion is related to the high taxation (31,4%) situation.

Table 29. Results of the net profit and loss account analysis, open pond system, 9 scenarios.

| Scenario | A | B | C | D | E | F | G | H | I |
|---------------------------------|----------|----------|-----------|----------|----------|----------|----------|----------|----------|
| Biomass Production [g/(m2 day)] | 20 | 25 | 30 | 20 | 25 | 30 | 20 | 25 | 30 |
| Oil Content | 30% | | | 40% | | | 50% | | |
| Revenue from oil | € 8.073 | € 10.092 | € 12.110 | € 10.765 | € 13.456 | € 16.147 | € 13.456 | € 16.820 | € 20.183 |
| Revenue from biomass | € 5.366 | € 6.707 | € 8.048 | € 4.599 | € 5.749 | € 6.899 | € 3.833 | € 4.791 | € 5.749 |
| Total revenues | € 13.439 | € 16.799 | € 20.158 | € 15.364 | € 19.204 | € 23.045 | € 17.288 | € 21.610 | € 25.932 |
| Operational profits | € 1.976 | € 5.336 | € 8.696 | € 3.901 | € 7.742 | € 11.583 | € 5.826 | € 10.148 | € 14.470 |
| Feasibility | Red | | Green | Red | Yellow | Green | Orange | Green | |
| NPV, tax 31,4% | - | - | € 20.456 | - | € 14.190 | € 38.805 | € 800 | € 29.756 | € 56.693 |
| Payback period, tax 31,4% | - | - | 6/7 years | - | 7 years | 5 years | 10 years | 6 years | 4 years |
| IRR, tax 31,4% | - | - | 9,9% | - | 7,1% | 17,2% | 0,5% | 13,7% | 23,6% |
| NPV, tax 20% | - | - | € 24.979 | - | € 17.793 | € 46.217 | € 2.527 | € 35.708 | € 67.008 |
| Payback period, tax 20% | - | - | 6 years | - | 7 years | 5 years | 10 years | 6 years | 4 years |
| IRR, tax 20% | - | - | 11,6% | - | 8,6% | 19,7% | 1,4% | 15,8% | 26,9% |

The scenario G has a positive NPV; nevertheless with the use of equity it is not an interesting scenario, due to its low NPV (800 €). With the assumption of a government grant it could be considered positive, even if it has long payback period and low IRR. We would not consider this option as feasible, due to future uncertain and low margin, even if we have already taken contingency risks into account in the project costs.

The two scenarios C and E had a rather long payback period in the feasibility study. Now, with 80% bank loan, the payback period is lower and the 2 options appear more interesting. The scenario E with 20% equity (almost 10 k€) might be not profitable enough for the investors, even if the NPV is positive and bigger than 14 k€. The same consideration could be valid for the scenario C, even if it is rather positive (NPV of over 20 k€).

The three last scenarios, F, H and I are very positive. They have rather short payback periods, NPV of over 3 times the equity (almost 6 times for the case I) and IRR of over 15%. We would definitely recommend these investments to any investor.

Next we propose the results of the analysis for the different cases. Revenues, operational costs, EBITDA, amortization, EBIT, interests at a 6% rate, taxes at 31,4%, taxes at 20%, net result for both the taxations, accumulated cash flow for both the taxations, NPV, payback period and IRR are presented for each scenario (Table 30 – Table 35).

Table 30. Results for the scenario C, with a production of 30 g/(m² day) and oil content of 30%.

| Scenario C | Year | Revenues | Op. Costs | EBITDA | Amortization | EBIT | Interests | Taxes 31,4% | Taxes 20% | Net Result (Taxes 31,4%) | Net Result (Taxes 20%) | Cum. Cash Flow (Taxes 31,4%) | Cum. Cash Flow (Taxes 20%) | NPV 31,4% |
|-----------------------------|------|-----------|------------|----------|--------------|----------|-----------|-------------|-----------|-----------------------------|---------------------------|---------------------------------|-------------------------------|----------------|
| | 0 | | | | | | | | | | | | | € 20.456 |
| Biomass Production | 1 | € 20.158 | -€ 11.462 | € 8.696 | -€ 4.858 | € 3.838 | -€ 2.332 | -€ 1.205 | -€ 768 | € - | € - | -€ 33.703 | -€ 33.266 | NPV 20% |
| 30 [g/(m ² day)] | 2 | € 20.158 | -€ 11.462 | € 8.696 | -€ 4.858 | € 3.838 | -€ 1.950 | -€ 1.205 | -€ 768 | € - | € - | -€ 28.162 | -€ 27.287 | € 24.979 |
| | 3 | € 20.158 | -€ 11.462 | € 8.696 | -€ 4.858 | € 3.838 | -€ 1.545 | -€ 1.205 | -€ 768 | € - | € - | -€ 22.217 | -€ 20.904 | |
| Oil Content 30% | 4 | € 20.158 | -€ 11.462 | € 8.696 | -€ 4.858 | € 3.838 | -€ 1.116 | -€ 1.205 | -€ 768 | € - | € - | -€ 15.842 | -€ 14.092 | Payback Period |
| | 5 | € 20.158 | -€ 11.462 | € 8.696 | -€ 4.858 | € 3.838 | -€ 661 | -€ 1.205 | -€ 768 | € - | € - | -€ 9.012 | -€ 6.825 | 6/7 years |
| | 6 | € 20.158 | -€ 11.462 | € 8.696 | -€ 4.858 | € 3.838 | -€ 179 | -€ 1.205 | -€ 768 | € - | € 924 | -€ 1.701 | € 924 | |
| | 7 | € 20.158 | -€ 11.462 | € 8.696 | -€ 4.858 | € 3.838 | € - | -€ 1.205 | -€ 768 | € 5.790 | € 7.928 | € 5.790 | € 8.853 | IRR 31,4% |
| | 8 | € 20.158 | -€ 11.462 | € 8.696 | -€ 4.858 | € 3.838 | € - | -€ 1.205 | -€ 768 | € 7.491 | € 7.928 | € 13.281 | € 16.781 | |
| | 9 | € 20.158 | -€ 11.462 | € 8.696 | € - | € 8.696 | € - | -€ 2.731 | -€ 1.739 | € 5.965 | € 6.957 | € 19.246 | € 23.738 | IRR 20% |
| | 10 | € 20.158 | -€ 11.462 | € 8.696 | € - | € 8.696 | € - | -€ 2.731 | -€ 1.739 | € 5.965 | € 6.957 | € 25.211 | € 30.694 | 11,6% |
| | | € 201.583 | -€ 114.624 | € 86.959 | -€ 38.862 | € 48.097 | -€ 7.783 | -€ 15.103 | -€ 9.619 | € 25.211 | € 30.694 | € 25.211 | € 30.694 | |

Table 31. Results for the scenario E, with a production of 25 g/(m² day) and oil content of 40%.

| Scenario E | Year | Revenues | Op. Costs | EBITDA | Amortization | EBIT | Interests | Taxes 31,4% | Taxes 20% | Net Result (Taxes 31,4%) | Net Result (Taxes 20%) | Cum. Cash Flow (Taxes 31,4%) | Cum. Cash Flow (Taxes 20%) | NPV 31,4% |
|-----------------------------|------|-----------|------------|----------|--------------|----------|-----------|-------------|-----------|-----------------------------|---------------------------|---------------------------------|-------------------------------|----------------|
| | 0 | | | | | | | | | | | | | € 14.190 |
| Biomass Production | 1 | € 19.204 | -€ 11.462 | € 7.742 | -€ 4.858 | € 2.884 | -€ 2.332 | -€ 906 | -€ 577 | € - | € - | -€ 34.358 | -€ 34.029 | NPV 20% |
| 25 [g/(m ² day)] | 2 | € 19.204 | -€ 11.462 | € 7.742 | -€ 4.858 | € 2.884 | -€ 2.007 | -€ 906 | -€ 577 | € - | € - | -€ 29.528 | -€ 28.871 | € 17.793 |
| | 3 | € 19.204 | -€ 11.462 | € 7.742 | -€ 4.858 | € 2.884 | -€ 1.663 | -€ 906 | -€ 577 | € - | € - | -€ 24.355 | -€ 23.369 | |
| Oil Content 40% | 4 | € 19.204 | -€ 11.462 | € 7.742 | -€ 4.858 | € 2.884 | -€ 1.298 | -€ 906 | -€ 577 | € - | € - | -€ 18.817 | -€ 17.502 | Payback Period |
| | 5 | € 19.204 | -€ 11.462 | € 7.742 | -€ 4.858 | € 2.884 | -€ 912 | -€ 906 | -€ 577 | € - | € - | -€ 12.892 | -€ 11.248 | 7 years |
| | 6 | € 19.204 | -€ 11.462 | € 7.742 | -€ 4.858 | € 2.884 | -€ 502 | -€ 906 | -€ 577 | € - | € - | -€ 6.558 | -€ 4.585 | |
| | 7 | € 19.204 | -€ 11.462 | € 7.742 | -€ 4.858 | € 2.884 | € 67 | -€ 906 | -€ 577 | € 211 | € 2.513 | € 211 | € 2.513 | IRR 31,4% |
| | 8 | € 19.204 | -€ 11.462 | € 7.742 | -€ 4.858 | € 2.884 | € - | -€ 906 | -€ 577 | € 6.836 | € 7.165 | € 7.048 | € 9.678 | 7,1% |
| | 9 | € 19.204 | -€ 11.462 | € 7.742 | € - | € 7.742 | € - | -€ 2.431 | -€ 1.548 | € 5.311 | € 6.194 | € 12.359 | € 15.872 | IRR 20% |
| | 10 | € 19.204 | -€ 11.462 | € 7.742 | € - | € 7.742 | € - | -€ 2.431 | -€ 1.548 | € 5.311 | € 6.194 | € 17.670 | € 22.065 | 8,6% |
| | | € 192.044 | -€ 114.624 | € 77.420 | -€ 38.862 | € 38.558 | -€ 8.781 | -€ 12.107 | -€ 7.712 | € 17.670 | € 22.065 | € 17.670 | € 22.065 | |

Table 32. Results for the scenario F, with a production of 30 g/(m² day) and oil content of 40%.

| Scenario F | Year | Revenues | Op. Costs | EBITDA | Amortization | EBIT | Interests | Taxes 31,4% | Taxes 20% | Net Result (Taxes 31,4%) | Net Result (Taxes 20%) | Cum. Cash Flow (Taxes 31,4%) | Cum. Cash Flow (Taxes 20%) | NPV 31,4% |
|-----------------------------|------|-----------|------------|-----------|--------------|----------|-----------|-------------|-----------|-----------------------------|---------------------------|---------------------------------|-------------------------------|----------------|
| | 0 | | | | | | | | | | | | | € 31.805 |
| Biomass Production | 1 | € 23.045 | -€ 11.462 | € 11.583 | -€ 4.858 | € 6.725 | -€ 2.332 | -€ 2.112 | -€ 1.345 | € - | € - | -€ 31.723 | -€ 30.956 | NPV 20% |
| 30 [g/(m ² day)] | 2 | € 23.045 | -€ 11.462 | € 11.583 | -€ 4.858 | € 6.725 | -€ 1.777 | -€ 2.112 | -€ 1.345 | € - | € - | -€ 24.028 | -€ 22.495 | € 46.217 |
| | 3 | € 23.045 | -€ 11.462 | € 11.583 | -€ 4.858 | € 6.725 | -€ 1.188 | -€ 2.112 | -€ 1.345 | € - | € - | -€ 15.745 | -€ 13.445 | |
| Oil Content 40% | 4 | € 23.045 | -€ 11.462 | € 11.583 | -€ 4.858 | € 6.725 | € 565 | -€ 2.112 | -€ 1.345 | € - | € - | -€ 6.839 | -€ 3.772 | Payback Period |
| | 5 | € 23.045 | -€ 11.462 | € 11.583 | -€ 4.858 | € 6.725 | € - | -€ 2.112 | -€ 1.345 | € 2.632 | € 6.466 | € 2.632 | € 6.466 | 5 years |
| | 6 | € 23.045 | -€ 11.462 | € 11.583 | -€ 4.858 | € 6.725 | € - | -€ 2.112 | -€ 1.345 | € 9.471 | € 10.238 | € 12.104 | € 16.704 | |
| | 7 | € 23.045 | -€ 11.462 | € 11.583 | -€ 4.858 | € 6.725 | € - | -€ 2.112 | -€ 1.345 | € 9.471 | € 10.238 | € 21.575 | € 26.941 | IRR 31,4% |
| | 8 | € 23.045 | -€ 11.462 | € 11.583 | -€ 4.858 | € 6.725 | € - | -€ 2.112 | -€ 1.345 | € 9.471 | € 10.238 | € 31.046 | € 37.179 | 17,2% |
| | 9 | € 23.045 | -€ 11.462 | € 11.583 | € - | € 11.583 | € - | -€ 3.637 | -€ 2.317 | € 7.946 | € 9.266 | € 38.992 | € 46.446 | IRR 20% |
| | 10 | € 23.045 | -€ 11.462 | € 11.583 | € - | € 11.583 | € - | -€ 3.637 | -€ 2.317 | € 7.946 | € 9.266 | € 46.938 | € 55.712 | 19,7% |
| | | € 230.453 | -€ 114.624 | € 115.829 | -€ 38.862 | € 76.967 | -€ 5.861 | -€ 24.167 | -€ 15.393 | € 46.938 | € 55.712 | € 46.938 | € 55.712 | |

Table 33. Results for the scenario G, with a production of 20 g/(m² day) and oil content of 50%.

| Scenario G | Year | Revenues | Op. Costs | EBITDA | Amortization | EBIT | Interests | Taxes 31,4% | Taxes 20% | Net Result (Taxes 31,4%) | Net Result (Taxes 20%) | Cum. Cash Flow (Taxes 31,4%) | Cum. Cash Flow (Taxes 20%) | NPV 31,4% |
|-----------------------------|------|-----------|------------|----------|--------------|----------|-----------|-------------|-----------|-----------------------------|---------------------------|---------------------------------|-------------------------------|----------------|
| | 0 | | | | | | | | | | | | | € 800 |
| Biomass Production | 1 | € 17.288 | -€ 11.462 | € 5.826 | -€ 4.858 | € 968 | -€ 2.332 | -€ 304 | -€ 194 | € - | € - | -€ 35.672 | -€ 35.562 | NPV 20% |
| 20 [g/(m ² day)] | 2 | € 17.288 | -€ 11.462 | € 5.826 | -€ 4.858 | € 968 | -€ 2.122 | -€ 304 | -€ 194 | € - | € - | -€ 32.272 | -€ 32.052 | € 2.527 |
| | 3 | € 17.288 | -€ 11.462 | € 5.826 | -€ 4.858 | € 968 | -€ 1.900 | -€ 304 | -€ 194 | € - | € - | -€ 28.650 | -€ 28.319 | |
| Oil Content 50% | 4 | € 17.288 | -€ 11.462 | € 5.826 | -€ 4.858 | € 968 | -€ 1.664 | -€ 304 | -€ 194 | € - | € - | -€ 24.793 | -€ 24.352 | Payback Period |
| | 5 | € 17.288 | -€ 11.462 | € 5.826 | -€ 4.858 | € 968 | -€ 1.415 | -€ 304 | -€ 194 | € - | € - | -€ 20.686 | -€ 20.134 | 10 years |
| | 6 | € 17.288 | -€ 11.462 | € 5.826 | -€ 4.858 | € 968 | -€ 1.150 | -€ 304 | -€ 194 | € - | € - | -€ 16.314 | -€ 15.652 | |
| | 7 | € 17.288 | -€ 11.462 | € 5.826 | -€ 4.858 | € 968 | -€ 869 | -€ 304 | -€ 194 | € - | € - | -€ 11.662 | -€ 10.889 | IRR 31,4% |
| | 8 | € 17.288 | -€ 11.462 | € 5.826 | -€ 4.858 | € 968 | -€ 572 | -€ 304 | -€ 194 | € - | € - | -€ 6.712 | -€ 5.829 | 0,5% |
| | 9 | € 17.288 | -€ 11.462 | € 5.826 | € - | € 5.826 | -€ 257 | -€ 1.829 | -€ 1.165 | € - | € - | -€ 2.972 | -€ 1.425 | IRR 20% |
| | 10 | € 17.288 | -€ 11.462 | € 5.826 | € - | € 5.826 | € - | -€ 1.829 | -€ 1.165 | € 1.024 | € 3.235 | € 1.024 | € 3.235 | 1,4% |
| | | € 172.881 | -€ 114.624 | € 58.257 | -€ 38.862 | € 19.395 | -€ 12.281 | -€ 6.090 | -€ 3.879 | € 1.024 | € 3.235 | € 1.024 | € 3.235 | |

Table 34. Results for the scenario H, with a production of 25 g/(m² day) and oil content of 50%.

| Scenario H | Year | Revenues | Op. Costs | EBITDA | Amortization | EBIT | Interests | Taxes 31,4% | Taxes 20% | Net Result (Taxes 31,4%) | Net Result (Taxes 20%) | Cum. Cash Flow (Taxes 31,4%) | Cum. Cash Flow (Taxes 20%) | NPV 31,4% |
|---|------|-----------|------------|-----------|--------------|----------|-----------|-------------|-----------|-----------------------------|---------------------------|---------------------------------|-------------------------------|----------------|
| | 0 | | | | | | | | | | | | | € 29.756 |
| Biomass Production 25 [g/(m ² day)] | 1 | € 21.610 | -€ 11.462 | € 10.148 | -€ 4.858 | € 5.290 | -€ 2.332 | -€ 1.661 | -€ 1.058 | € - | € - | € 32.707 | -€ 32.104 | NPV 20% |
| | 2 | € 21.610 | -€ 11.462 | € 10.148 | -€ 4.858 | € 5.290 | -€ 1.863 | -€ 1.661 | -€ 1.058 | € - | € - | -€ 26.083 | -€ 24.877 | € 35.708 |
| | 3 | € 21.610 | -€ 11.462 | € 10.148 | -€ 4.858 | € 5.290 | -€ 1.366 | -€ 1.661 | -€ 1.058 | € - | € - | -€ 18.962 | -€ 17.153 | |
| Oil Content 50% | 4 | € 21.610 | -€ 11.462 | € 10.148 | -€ 4.858 | € 5.290 | -€ 839 | -€ 1.661 | -€ 1.058 | € - | € - | -€ 11.314 | -€ 8.902 | Payback Period |
| | 5 | € 21.610 | -€ 11.462 | € 10.148 | -€ 4.858 | € 5.290 | -€ 280 | -€ 1.661 | -€ 1.058 | € - | € - | -€ 3.108 | -€ 92 | 6 years |
| | 6 | € 21.610 | -€ 11.462 | € 10.148 | -€ 4.858 | € 5.290 | € - | -€ 1.661 | -€ 1.058 | € 5.379 | € 8.997 | € 5.379 | € 8.997 | |
| | 7 | € 21.610 | -€ 11.462 | € 10.148 | -€ 4.858 | € 5.290 | € - | -€ 1.661 | -€ 1.058 | € 8.487 | € 9.090 | € 13.866 | € 18.087 | IRR 31,4% |
| | 8 | € 21.610 | -€ 11.462 | € 10.148 | -€ 4.858 | € 5.290 | € - | -€ 1.661 | -€ 1.058 | € 8.487 | € 9.090 | € 22.352 | € 27.177 | 13,7% |
| | 9 | € 21.610 | -€ 11.462 | € 10.148 | € - | € 10.148 | € - | -€ 3.186 | -€ 2.030 | € 6.961 | € 8.118 | € 29.314 | € 35.295 | IRR 20% |
| | 10 | € 21.610 | -€ 11.462 | € 10.148 | € - | € 10.148 | € - | -€ 3.186 | -€ 2.030 | € 6.961 | € 8.118 | € 36.275 | € 43.413 | 15,8% |
| | | € 216.102 | -€ 114.624 | € 101.478 | -€ 38.862 | € 62.616 | -€ 6.679 | -€ 19.661 | -€ 12.523 | € 36.275 | € 43.413 | € 36.275 | € 43.413 | |

Table 35. Results for the scenario I, with a production of 30 g/(m² day) and oil content of 50%.

| Scenario I | Year | Revenues | Op. Costs | EBITDA | Amortization | EBIT | Interests | Taxes 31,4% | Taxes 20% | Net Result (Taxes 31,4%) | Net Result (Taxes 20%) | Cum. Cash Flow (Taxes 31,4%) | Cum. Cash Flow (Taxes 20%) | NPV 31,4% |
|---|------|-----------|------------|-----------|--------------|-----------|-----------|-------------|-----------|-----------------------------|---------------------------|---------------------------------|-------------------------------|----------------|
| | 0 | | | | | | | | | | | | | € 56.893 |
| Biomass Production 30 [g/(m ² day)] | 1 | € 25.932 | -€ 11.462 | € 14.470 | -€ 4.858 | € 9.612 | -€ 2.332 | -€ 3.018 | -€ 1.922 | € - | € - | € 29.742 | -€ 28.647 | NPV 20% |
| | 2 | € 25.932 | -€ 11.462 | € 14.470 | -€ 4.858 | € 9.612 | -€ 1.603 | -€ 3.018 | -€ 1.922 | € - | € - | -€ 19.894 | -€ 17.703 | € 67.008 |
| | 3 | € 25.932 | -€ 11.462 | € 14.470 | -€ 4.858 | € 9.612 | -€ 831 | -€ 3.018 | -€ 1.922 | € - | € - | -€ 9.274 | -€ 5.987 | |
| Oil Content 50% | 4 | € 25.932 | -€ 11.462 | € 14.470 | -€ 4.858 | € 9.612 | -€ 13 | -€ 3.018 | -€ 1.922 | € 2.165 | € 6.548 | € 2.165 | € 6.548 | Payback Period |
| | 5 | € 25.932 | -€ 11.462 | € 14.470 | -€ 4.858 | € 9.612 | € - | -€ 3.018 | -€ 1.922 | € 11.452 | € 12.547 | € 13.616 | € 19.095 | 6 years |
| | 6 | € 25.932 | -€ 11.462 | € 14.470 | -€ 4.858 | € 9.612 | € - | -€ 3.018 | -€ 1.922 | € 11.452 | € 12.547 | € 25.068 | € 31.642 | |
| | 7 | € 25.932 | -€ 11.462 | € 14.470 | -€ 4.858 | € 9.612 | € - | -€ 3.018 | -€ 1.922 | € 11.452 | € 12.547 | € 36.519 | € 44.190 | IRR 31,4% |
| | 8 | € 25.932 | -€ 11.462 | € 14.470 | -€ 4.858 | € 9.612 | € - | -€ 3.018 | -€ 1.922 | € 11.452 | € 12.547 | € 47.971 | € 56.737 | 23,6% |
| | 9 | € 25.932 | -€ 11.462 | € 14.470 | € - | € 14.470 | € - | -€ 4.544 | -€ 2.894 | € 9.926 | € 11.576 | € 57.897 | € 68.313 | IRR 20% |
| | 10 | € 25.932 | -€ 11.462 | € 14.470 | € - | € 14.470 | € - | -€ 4.544 | -€ 2.894 | € 9.926 | € 11.576 | € 67.824 | € 79.889 | 26,9% |
| | | € 259.322 | -€ 114.624 | € 144.698 | -€ 38.862 | € 105.836 | -€ 4.780 | -€ 33.232 | -€ 21.167 | € 67.824 | € 79.889 | € 67.824 | € 79.889 | |

7. Economic analysis for photobioreactors.

7.1. Revenues.

This paragraph examined microalgae production with photobioreactors. Our aim is to study the economic feasibility of biofuels from microalgae and this is the target that drives our evaluation. For this reason, even if the products of photobioreactor could be extremely valuable due to the high growth control attained, we want the production to be focused on oil and biofuels. There are two aspects that must be recalled: first, when producing algae with high oil percentage, it is difficult to match the biofuels production with the one of other high-value products, which have their own biological characteristics and thus, different nutrition and growth needs. Secondly, if the revenues from the biomass remained after the oil extraction are bigger than the revenues from oil, biofuels production makes no sense and it would be better to produce just high valued products. In table 36 and in table 37 our assumptions for the biomass use and prices are presented, with the resulting crude oil and biomass prices.

Table 36. Biomass different uses and prices, PBRs.

| Biomass use | Price [€/t] | Use % |
|-----------------------|-------------|-------|
| Anaerobic digestion | 75 | 0% |
| High protein food | 150 | 40% |
| High value co-product | 500 | 60% |

Table 37. Algal oil and biomass prices, PBRs.

| PRICE OF OIL AND BIOMASS FROM MICROALGAE | |
|--|-------|
| Crude oil price [\$/barrel] | 87 |
| Crude oil price [€/barrel] | 60,9 |
| Reduction in energy content | 7% |
| Algal oil price [€/barrel] | 56,64 |
| Biomass price - PBRs [€/t] | 360 |

The resulting biomass price is 360 €/t. This price is selected for one reason: it makes the revenues from oil and biomass very similar, so it is the maximum price that justifies the oil production. As it will be underlined in the next paragraphs, co-products could be priced over 10 k€/kg, so a higher biomass price could be assumed.

In the photobioreactors analysis, we assumed the highest published productivity (Algaelink's 365 t/(ha yr)) and three different scenarios, with 35%, 45% and 55% oil content. As it could be easily noticed, both productivity and oil content are higher than in the open systems scenarios. Furthermore, the revenues between different scenarios are similar, because of the assumed biomass price (Table 38).

Table 38. Different scenarios and revenues, PBRs.

| Scenario | J | K | L |
|---|-----------|-----------|-----------|
| Biomass Production [g/(m2 day)] | 100 | 100 | 100 |
| Days of Work | 365 | | |
| Production [t/(ha yr)] | 365 | 365 | 365 |
| Oil Content | 35% | 45% | 55% |
| Biomass Production [t/(ha yr)] | 237,25 | 200,75 | 164,25 |
| Oil Density [t/m3] | 0,918 | | |
| Oil extraction Efficiency | 95% | | |
| Oil Production [l/(m2 yr)] | 13,22 | 17,00 | 20,77 |
| Oil Production [barrels/(ha yr)] | 831,52 | 1.069,10 | 1.306,67 |
| Revenue from oil | € 47.095 | € 60.550 | € 74.006 |
| Revenue from biomass | € 85.410 | € 72.270 | € 59.130 |
| Total revenues | € 132.505 | € 132.820 | € 133.136 |

7.2. Costs.

Costs of PBRs are way higher than for open ponds systems. Here we use the cost of an Algaelink photobioreactor. Several assumptions could be made, different producer could have a lower price, a substantial cost reduction could be obtained with bigger systems because of economies of scale or the cost that we have could not be the best deal or it could have been decreased in the last months (it is a 2008 cost). We assumed a cost reduction of 15% from the original cost of 1,2 M€ for the system (Table 39). Because the harvesting needs are similar to the open ponds one (Table 40) and the same volume of medium need to be treated, we assumed that the harvesting operations discussed by Benemann and Oswald (1996) are appropriate for PBRs. However, because of the smaller scale of Algaelink's

system (if compared to the assumed large open system facility) we estimated a cost increase of 50% for the harvesting operations and of 100% for the centrifugation. Benemann and Oswald assumed very large systems in order to obtain a significant costs reduction of downstream machineries. The resulting plant is very costly (Table 39). The main cost component is the photobioreactor.

Table 39. Capital costs for a photobioreactor system.

| CAPITAL COSTS | [€/ha] | |
|--|----------------|--|
| Algae Production | | |
| Photobioreactor | 1020000 | (Algaelink, 2008, reduced by 15%) |
| Control instruments | 0 | Included in the PBR cost |
| Polyethylene tubes | 0 | Included in the PBR cost |
| Software | 0 | Included in the PBR cost |
| Sensors | 0 | Included in the PBR cost |
| Pumps | 0 | Included in the PBR cost |
| Buildings, roads, drainage | 1978 | (Benemann and Oswald, 1996) |
| Downstream Treatment | | |
| Flocculation and dissolved air flotation | 13053 | (Benemann and Oswald, 1996, increased by 50%) |
| Centrifugation and oil extraction | 24171 | (Benemann and Oswald, 1996, increased by 100%) |
| Direct cost of algae production and treatment | | |
| Engineering and design | 0 | Included in the PBR cost |
| Install plant costs | 1059202 | |
| Contingency, supervision and management overhead | 52960 | 5% of the Install plant costs |
| Total plant costs | 1112162 | |

Table 40. Main details for the two first harvesting operations, photobioreactor.

| HARVESTING OPERATIONS | |
|--|-------|
| Tube diameter [m] | 0,32 |
| Tube length [m] | 24868 |
| Total culture volume [m3] | 2000 |
| Retention time [days] | 2 |
| Maximum productivity [g/(m2 day)] | 60 |
| 1st harvesting system (settling): | |
| Achieved fold concentration | 20 |
| Achieved solid concentration | 1,2% |
| Output volume [m3] | 50 |
| Output liters | 50000 |
| 2nd harvesting system (DAF): | |
| Achieved fold concentration | 5 |
| Achieved solid concentration | 6,0% |
| Output volume [m3] | 10 |
| Output liters | 10000 |

Generally speaking, the operating costs of PBRs are higher than in open systems, because of the higher productivity (major needs of nutrients and flocculants, here an increase of 25% is assumed) and because of higher maintenance costs (related to plant costs). Operating costs for PBRs are reported in table 41.

Table 41. Operating costs for a photobioreactor system.

| OPERATIONAL COSTS | [€/(ha yr)] | |
|--------------------------------|--------------|---|
| Land cost | 200 | (Eurostat, 2009) |
| Power costs | 2999 | (Benemann and Oswald, 1996) |
| Nutrient supply | 1112 | (Benemann and Oswald, 1996, increased by 25%) |
| Flocculants | 1236 | (Benemann and Oswald, 1996, increased by 25%) |
| CO ₂ | 0 | Supposed free, see related paragraph |
| Waste disposal | 989 | (Benemann and Oswald, 1996) |
| Maintenance | 55608 | 5% of total plant costs |
| Labor and overheads | 2967 | (Benemann and Oswald, 1996) |
| Total operational costs | 65111 | |

7.3. Economic Analysis.

Table 42 shows the operational profits for the three photobioreactor scenarios. The investment cost (of over 1,1 M€) needs operational profits of over 110 k€/yr, in order to be paid back in ten year. Several considerations are possible: our cost assumptions could be wrong, or a longer payback period could be considered. However, we are quite confident in concluding this simple economic analysis of microalgae cultivation in photobioreactors for the production of oil. The oil and biofuels production would be feasible only when photobioreactors will become less expensive (a cost reduction of circa 80-100% could be supposed). Algal cultivation in PBRs is a promising technology for high-value products, but nowadays a large scaled facility for a mass production of oil through PBRs is not economically feasible.

Table 42. Operational profits for a photobioreactor system.

| Scenario | J | K | L |
|---|-----------|-----------|-----------|
| Biomass Production [g/(m² day)] | 100 | 100 | 100 |
| Oil Content | 35% | 45% | 55% |
| Revenue from oil | € 47.095 | € 60.550 | € 74.006 |
| Revenue from biomass | € 85.410 | € 72.270 | € 59.130 |
| Total revenues | € 132.505 | € 132.820 | € 133.136 |
| Operational profits | € 67.394 | € 67.709 | € 68.025 |

CHAPTER 7

System optimization.

1. Combined processes.

Production of biodiesel and other bio-products from microalgae can be more environmentally sustainable, cost-effective and profitable, if combined with processes such as wastewater and flue gas treatments. In fact various studies demonstrated the use of microalgae for production combined with environmental applications (Mata et Al., 2010). Here our goal is to identify the synergies that microalgae production has with wastewater treatments and with CO₂ capture. Several of these synergies have been assumed in our economic analysis (chapter 6).

1.1. Integration with water treatment facilities.

Supply and cost of nutrients (nitrogen, phosphorus, and potassium) is a key issue for achieving affordable and sustainable scale-up of algae biofuels production. Wastewaters are excellent algal growth media (Lundquist, 2008). High rates of algae production lead to high rates of nutrient removal and wastewater treatment. Thus, the objectives of biofuel feedstock production and wastewater treatment are aligned, at least in terms of maximizing biomass production. Fig. 33 shows the integration of algae production with wastewater treatment for nutrient removal and biomass production.

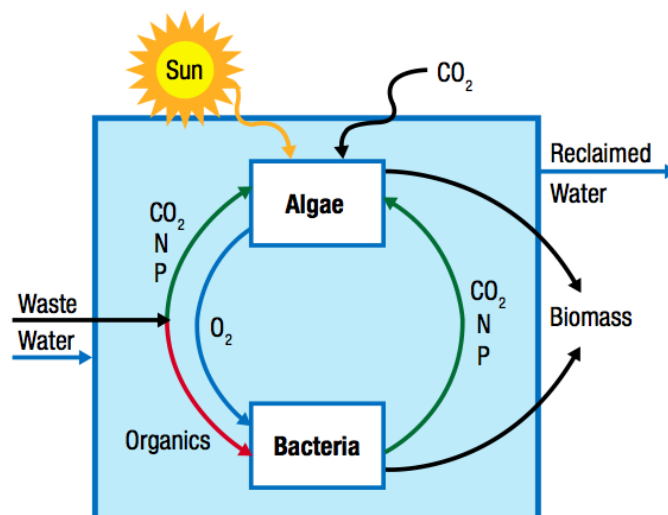


Fig. 33. Algae production coupled with wastewater treatment, basic principles of operation (DOE, 2010)

The main connections of algae production and wastewater treatment are the following (DOE, 2010):

- Treatment technology is needed to recycle nutrients and water from algae biofuel processing residuals for use in algae production.
- Imported wastewater provides nutrients and water to make-up inevitable losses. The imported wastewater would be treated as part of the algae production.
- Algae-based wastewater treatment provides a needed service.
- Early opportunity to develop large-scale algae production infrastructure and development of skilled algae production workforce.
- Wastewater treatment revenue that offsets algae production costs (there are also potentials for wastewater credits, depending on the country).
- Lower capital and operational costs than conventional wastewater treatment.
- Lower energy intensity than conventional wastewater treatment (a greenhouse gas benefit).

The last two points deserve an in-depth examination: algae-based treatment facilities are typically less expensive to build and to operate than conventional mechanical treatment facilities. For example, high-productivity algae ponds have a total cost that is about 70% less than activated sludge, which is the leading water treatment technology used in the United States (DOE, 2010).

This cost savings, coupled with the tremendous need for expanded and improved wastewater treatment in the United States (DOE, 2010) and throughout the world, provide a practical opportunity to install algae production facilities in conjunction with wastewater treatment. The major classes of wastewaters to be treated are municipal, organic industrial (e.g., food processing) and organic agricultural (e.g., confined animal facilities). In current technology, with very few exceptions (e.g., the City of Sunnyvale, California) the algal biomass is not harvested (Sheenan et Al., 1998).

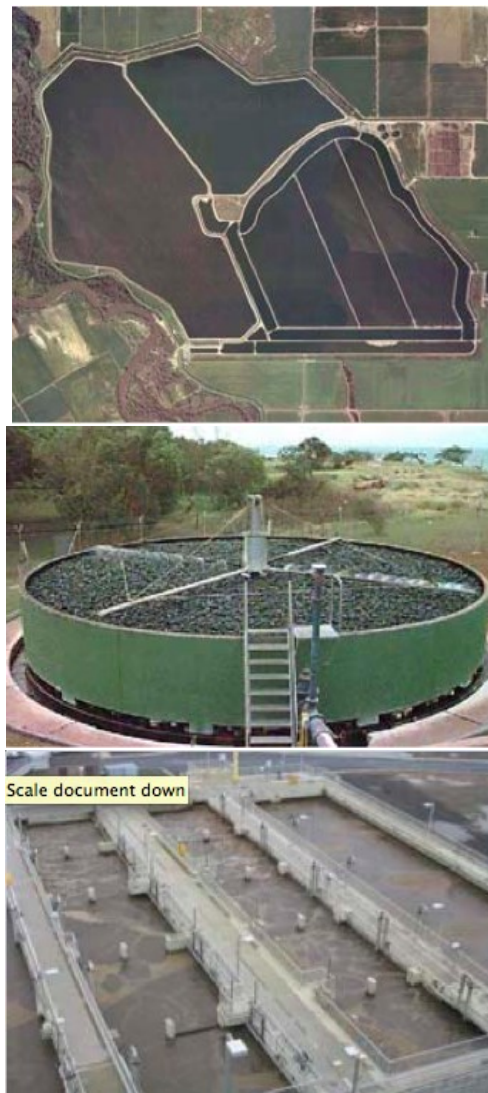


Fig. 34 a, b, c. Traditional wastewater technologies. From the top: Conventional ponds, mechanical systems and activated sludge (Lundquist, 2008).

Fig. 34 shows the traditional wastewater technologies.

Closed photobioreactors are not emphasized in this discussion since they are likely to be economical only when also producing high-value products (>100 \$/kg biomass), which is unlikely when wastewater contaminants are present.

1.2. Integration with carbon-emitting industries.

In our project of a large-scale algal-cultivation system we assumed the CO₂ to be supplied by an existing power plant or another industrial carbon producer. This approach not only provides the raw materials for the system, but also converts wastes into resources. Production of algae at a power plant is a great opportunity, in order to take advantage of waste CO₂ and possibly also to utilize the waste heat from the power plant. The largest and most concentrated individual sources of CO₂ are coal fired power stations, oil sands projects, gas processing, refining and upgrading, petrochemicals, pulp and paper, cement/lime production and pipeline compressors.

Resulting costs for CO₂ will be site-specific and dependent on methods of capture, conditioning, and distance of transport to algae cultivation sites. Costs are expected to be way lower than for pure commercially supplied CO₂, but economic viability must be determined case by case (DOE, 2010). Fig 35 shows the production of algae combined to a power plant.



Fig. 35. Algae cultivation in a greenhouse, using photobioreactors, connected with a power plant CO₂ supplier in Arizona (Sun and Hobbs, 2008).

Here we list the main advantages of co-locating algae production with stationary industrial CO₂ sources:

- Abundant quantities of concentrated CO₂ available from stationary industrial sources can supplement low concentration CO₂ from the atmosphere.
- Excess heat or power may be available to provide heating or cooling for improved thermal management of algae cultivation systems; this will allow developing algal cultivation facilities under a broader range of geographic and climate conditions on or near a year-round basis.
- Excess wastewater or cooling water may be available, found often in proximity of power plants, thus overcoming a primary resource challenge for algae cultivation at scale, while providing beneficial re-use of cooling water and wastewater.
- Potential carbon credit for utilities. This factor will need an established policy on carbon absorption and re-use as transportation fuel in lieu of permanent sequestration. To take into account the environmental impact of an energy technology, the IEA puts a price on the emitted CO₂. The basic assumption of the IEA (2010) was a carbon price of 30 €/ton of CO₂. Thus, CO₂-intensive technologies are penalized as they carry a cost of €30 per ton CO₂ emitted. However this is just an indication from an international agency and there is still great uncertain about regulations of single states.

Not only advantages but also barriers to co-location of algae production with stationary industrial CO₂ sources have to be considered:

- Power plants could operate as peaking plants and have intermittency in the supply of CO₂ for algae growth. The impact on algae production would depend on the phasing of the intermittency with respect to the daylight hours when photosynthesis is active.
- Unclear regulatory framework for carbon-capture credits: until there are regulations in place that quantify carbon credits from algal growth facilities, the uncertainty may pose a barrier for wide commercial adoption of the technology.
- Although here it is considered a “free” resource, the capture and delivery of CO₂ from stationary industrial sources as a supplement to enhance and optimize algae production may not be “free”.

2. Co-products.

When biofuel production is considered to be the primary goal, the generation of other co-products must be correspondingly low since their generation will inevitably compete for carbon, reductant, and energy from photosynthesis. Indeed, the concept of a biorefinery for utilization of every component of the biomass raw material must be considered as a means to enhance the economics of the process.

Using appropriate technologies, all primary components of algal biomass (carbohydrates, fats (oils), proteins and a variety of inorganic and complex organic molecules) can be converted into different products. The nature of the end products and of the technologies to be employed will be determined, primarily by the economics of the system. Co-products from algal refineries should address one of these three criteria to be commercially viable and acceptable:

1. Being identical to an existing chemical, fuel, or other product. In this instance, the only issue is price.
2. Being identical in functional performance to an existing chemical, fuel or other product. Here price is a major factor, but the source of the material can often provide some advantage. Price becomes less of an issue if the product can be labeled “organic” and thus saleable at a premium.
3. Being a new material with unique and useful functional performance characteristics.

Fig. 36 shows the market size for possible co-products from algae.

| COMMERCIAL PRODUCT | MARKET SIZE (TONS/YR) | SALES VOLUME (MILLION \$US/YR) | REFERENCE |
|--|---|--------------------------------|--|
| BIOMASS | | | |
| Health Food | 7,000 | 2,500 | Pulz&Gross (2004) |
| Aquaculture | 1,000 | 700 | Pulz&Gross (2004) Spolaore et al., (2006) |
| Animal Feed Additive | No available information | 300 | Pulz&Gross (2004) |
| POLY-UNSATURATED FATTY ACIDS (PUFAs) | | | |
| ARA | No available information | 20 | Pulz&Gross (2004) |
| DHA | <300 | 1,500 | Pulz&Gross (2004) Spolaore et al., (2006) |
| PUFA Extracts | No available information | 10 | Pulz&Gross (2004) |
| GLA | Potential product, no current commercial market | | Spolaore et al., (2006) |
| EPA | Potential product, no current commercial market | | Spolaore et al., (2006) |
| ANTI-OXIDANTS | | | |
| Beta-Carotene | 1,200 | >280 | Pulz&Gross (2004) Spolaore et al., (2006) |
| Tocopherol CO ₂ Extract | No available information | 100-150 | Pulz&Gross (2004) |
| COLORING SUBSTANCES | | | |
| Astaxanthin | < 300 (biomass) | < 150 | Pulz&Gross (2004) Spolaore et al., (2006) |
| Phycocyanin | No available information | >10 | Pulz&Gross (2004) |
| Phycocerythrin | No available information | >2 | Pulz&Gross (2004) |
| FERTILIZERS/SOIL CONDITIONERS | | | |
| Fertilizers, growth promoters, soil conditioners | No available information | 5,000 | Pulz&Gross (2004) Metting&Pyne (1986) |

Fig. 36. Commercial products from algae: market size and sales volume (DOE, 2010).

2.1. Recovering economic value from biomass.

According to DOE (2010), there are at least five different options for recovering economic value from the lipid-extracted microalgal biomass (Fig. 37). These are:

- Option 1 – Maximum energy recovery from the lipid extracted biomass, with potential use of residuals as soil amendments.
- Option 2 – Recovery of protein from the lipid- extracted biomass for use in food and feed.
- Option 3 – Recovery and utilization of non-fuel lipids.
- Option 4 – Recovery and utilization of carbohydrates from lipid-extracted biomass, and the glycerol from the transesterification of lipids to biodiesel.
- Option 5 – Recovery/extraction of fuel lipids only, with use of the residual biomass as soil fertilizer and conditioner.

Here we briefly discuss the first two and the last options and associated technologies, because are the easier to implement, referring to DOE, 2010 for a wider discussion.

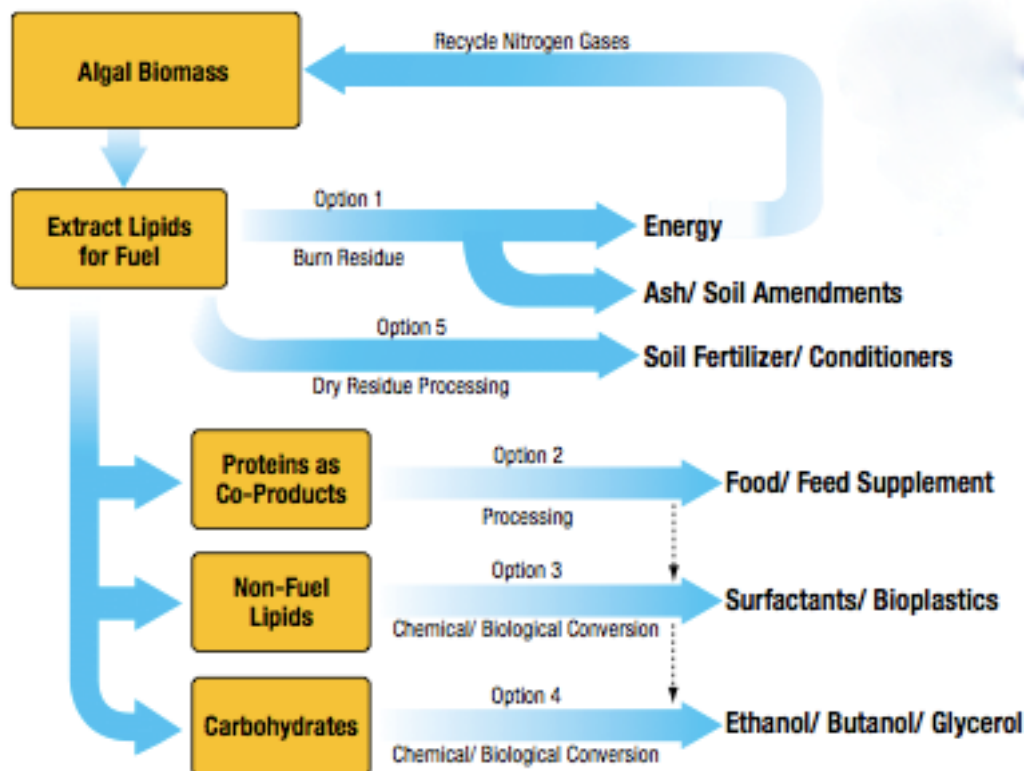


Fig. 37. Different options for recovering economic value from the lipid-extracted microalgal biomass (DOE, 2010).

Option 1: Given the large amounts of lipid-extracted biomass residues that will likely be generated in future microalgal biofuels production systems, it may be difficult to identify large enough markets for potential co-products. Therefore, one option would be to convert as much of the lipid-extracted biomass into energy, which could then be either sold on the open market or used on-site in the various biorefinery operations.

The most promising energy recovery technology, both from a practical and economic perspective, is the anaerobic digestion of lipid-extracted biomass. In our economic analysis we considered the economic value of the produced methane equivalent to about 75 € per ton of digested biomass (DOE, 2010 reported 100 \$/ton), which is significant in terms of reducing the overall cost of liquid biofuels production. The residuals remaining after anaerobic digestion could either be recycled as nutrients for algal cultivation or could be sold as soil fertilizers and conditioners.

In addition to anaerobic digestion, thermochemical conversion technologies, such as pyrolysis, gasification, and combustion, could also be potentially considered for the recovery of energy from the lipid-extracted biomass (see Chapter 5).

Option 2: Following the extraction of lipids from the microalgal biomass for liquid biofuel production, the protein fraction from the residual biomass could be extracted and used as a food and feed supplement. As was pointed out above, the market for animal feed (cattle, pigs, poultry, fish, and pets) is already very large and growing (estimated to rise to approximately 60 million tons per year for distillers dry grains plus soluble) (DOE, 2010). In our economic analysis we considered a biomass price for this use of 150 € per ton.

Option 5: In case none of the other four options are cost-effective, it is possible to revert to the simplest option, which involves the extraction of only fuel lipids and the subsequent use of the biomass residues rich in nitrogen and organic matter as soil fertilizer and conditioners. The market for organic fertilizer is large and potentially growing.

3. Important economical aspects.

3.1. Distribution and utilization.

Distribution and utilization are challenges associated with virtually all biofuels. Although the biofuel products from algal biomass would ideally be energy-dense and completely compatible with the existing liquid transportation fuel infrastructure, few studies exist that address outstanding issues of storing, transporting, pipelining, blending, combusting, and dispensing algal biomass, fuels intermediates, biofuels, and bioproducts (DOE, 2010).

Lowering costs associated with the delivery of raw biomass, fuel intermediates, and final fuels from the feedstock production center to the ultimate consumer are common goals for all biofuels. In all cases, biofuels infrastructure costs can be lowered in four ways:

1. Minimizing transport distance between process units.
2. Maximizing the substrate energy-density and stability.
3. Maximizing compatibility with existing infrastructure (e.g. storage tanks, high capacity; delivery vehicles, pipelines, dispensing equipment, and end-use vehicles).

4. Optimizing the scale of operations to the parameters stated above.

3.2. Intangibles.

This paragraph analyzes the intangible advantages that algae related products could have. As example, in the paragraph about co-products we underlined how they could be sold at a premium if they can be labeled “organic”. We identified two main intangible values that have to be considered: security of fuel supply and eco-friendliness of bioproducts.

3.2.1. Security of supply.

Security of supply is one of the aspects of oil dependency. It is defined as reliability of energy supply at affordable prices. In the first chapter we have seen as oil reserves are unequally distributed through the world, the Middle East has 63% of the global reserves and is the dominant supplier of petroleum (Hacisalihoglu et Al. 2009). The lack of fossil fuels supply could have massive macroeconomic consequences over nations that import most of their used fuels. For this reason, here we suggest accounting for the costs that are not reflected in the market oil prices, but are expected to emerge as a response to incremental supply changes. It is difficult, however, to find a proper way to estimate its monetary value, even if energy security is a policy priority for several nations.

3.2.2. Eco-friendliness of bioproducts.

A survey about biorefineries has been made in six European countries by Kurka (2009). The resulting assessment of biorefineries was very positive (Fig. 38). In particular, no strong public opposition towards biorefinery plants in the 6 surveyed countries must be expected, and we can assume a survey global validity. Concerning biorefineries, their “eco-friendliness” and their “positive economic effects” should be highlighted in public communication activities.

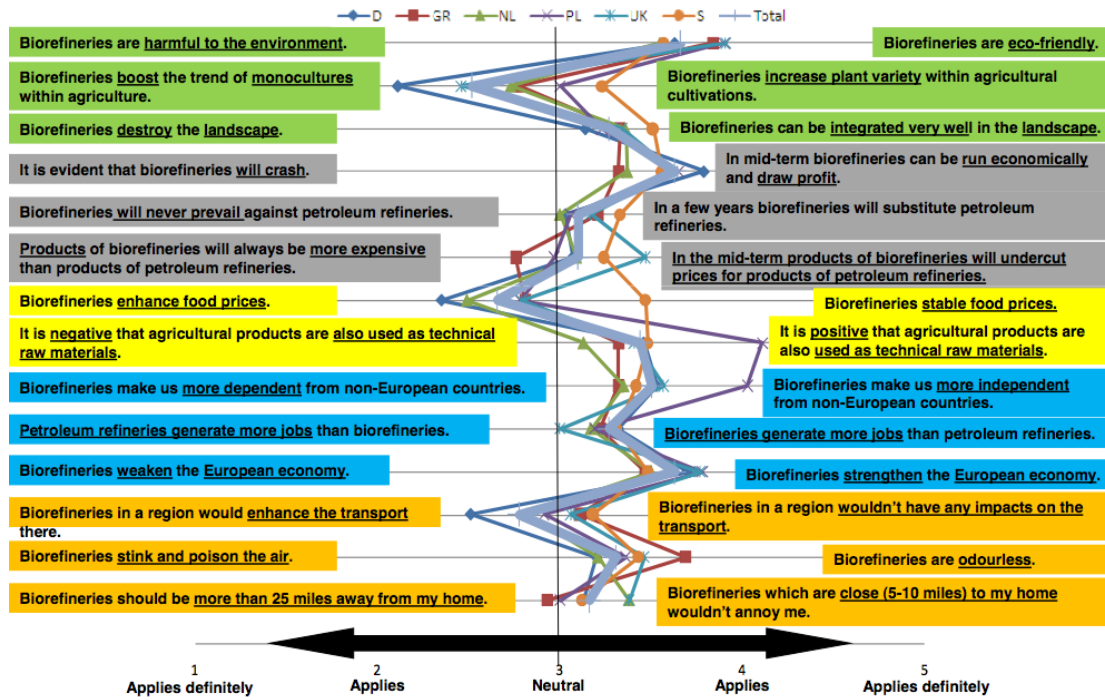


Fig. 38. Survey about attitude towards biorefineries (Kurka, 2009).

Several products could be obtained from biorefineries in addition to biofuels. The survey investigated the important factors for consumers buying bioplastic products. Searched product characteristics were being ecological, sustainable and safe for health. When these characteristics were satisfied, the consumers were available to pay a (limited) extra price for the surveyed products (biobased shampoo, washing-up liquid and other commodity bioproducts) (Fig. 39).

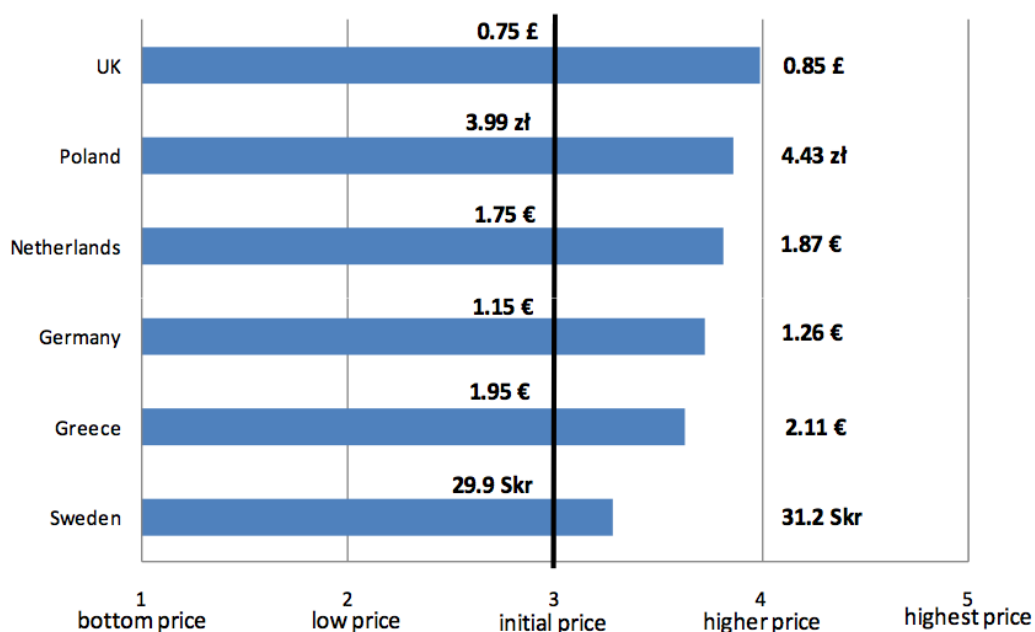


Fig. 39. Consumers willingness to pay more for bioproducts (Kurka, 2009).

The survey suggested intensive marketing activities to convince consumers about biobased products; furthermore the interested buyer segments should be addressed first. Consumers could be available to pay more for a “green” fuel; this is the obvious connection between the discussion above and fuels. Thus, algal biodiesel could be sold at a premium. Several gas stations already offer biofuels, at a higher price. This is an extra value that could be considered in the economical feasibility of algae biofuels.

3.3. Investments in the algae industry.

There is always uncertainty about the success of new products and investors have to consider carefully the proper energy sources in which to invest. A drop in fossil fuel oil prices might make consumers and therefore investors lose interest in renewable energy. Investors have different expectations about returns and length of investments and thus, our economic analysis provides just general indications.

To develop algae biofuel as a viable commercial fuel, a large investment of starting capital and several efforts in research and development will be required. Nevertheless, algal biofuels are gaining a wider attention through investors. In 2006, about \$15 million were invested in this industry by venture capital companies. In 2007, investment rose to \$32 million. In 2008, until end of Oct, about \$180 million in venture capital money has been raised for algae research. In July 2009, Exxon Mobil signed a \$600 million dollar multi-year partnership with a biotechnology company, Synthetic Genomes, which is the largest and most serious investment that has been made thus far in the development of commercial algae biofuel (Oilgae, 2011).

In Italy the most important project concerning algal biodiesel is probably the one in the Venetian Laguna. The project has the double goal of producing biofuels and solving the problem of polluting algae in Venice, which leads to unpleasant issues for local resident and tourists mainly by clogging the motors of boats and ferries. Enalg runs the project, with the aim of powering the city and its port. Even if the project addresses macroalgae, it is an important step towards acquiring the acceptance of algal fuels. It is predicted to produce 40 MW of energy and to power half of Venice (Enalg, 2011).

4. Combined heat and power cogeneration system.

The main aim of this paper is to determine the feasibility of producing fuels from microalgae. As underlined in the previous chapters, algal oil can be transformed in transportation fuels quite straightforward in refineries. In the economic analysis we made a comparison between algal oil and petrol oil, and determined the revenues stream that algae cultivation could have if the oil is sold as crude oil. However, an interesting alternative exists and deserves

coverage. Oil can be used in a combined heat and power cogeneration system, a highly efficient approach to produce electricity and heat in a single thermodynamic process from different sources of fuels. Here we consider a system made by Siatem Technology. The system has 485 kW of electric maximum power and a thermal power of 450 kW_t when the maximum electrical energy is produced. The used oil is 220 gr/kWh (+/- 5%). Typical efficiencies for combined heat and power cogeneration systems of similar dimensions are around 40% electric energy conversion and 45% thermal energy conversion (Zamalloa et Al., 2010). Therefore, the energy losses are around 15%.

A great advantage of combining these systems with the cultivation of algae is that the produced heat can be re-used within the processes, enhancing the productivity (water temperature control) and reducing downstream costs. Downstream operations such as dewatering and oil extraction often require heat and thus the system can be more economical.

4.1. Feed-in tariff.

There is a huge diversity in existing feed-in tariff between producing renewable energies and bio-fuels. Renewable technologies are in almost all cases more expensive than fossil fuel-based technologies. To stimulate investments in new renewable technologies, some governments subsidize the production of renewable energy by introducing feed-in-tariffs. The feed-in or minimum price systems guarantee fixed tariffs for the feed-in of green electricity into the grid. Usually biofuels are incentivized only when they are used for producing electrical (renewable) energy. Normally biofuels use in transportation is not incentivized. Even if we are confident that the situation will probably change, at the moment in some nations it is way more economical to burn biofuels in energy plants instead of using them for transportation.

In Italy, GSE (Gestore Servizi Energetici) plays a central role in promotion, support and development of renewable energy. Producers may apply for green certificates or for the all-inclusive feed-in tariff, which includes the support and the revenue from the sale of electricity (only for plants with a capacity of less than 1 MW) (GSE, 2011). Because of the dimension of the cogeneration plant in consideration, we analyze the all-inclusive feed-in tariff, applied to the net electricity generated and concurrently injected into the grid.

The all-inclusive feed-in tariff is applied at the request of the operator, over a period of 15 years and may be revised every three years by a Decree of the Minister of Economic Development. For algae oil the tariff is 0,28 €/kWh (Fig. 40).

| Source | All-inclusive feed-in rate (€cent/kWh) |
|--|---|
| Wind (P < 200 kW) | 30 |
| Geothermal | 20 |
| Waves and tides | 34 |
| Hydro (other than the one indicated in the previous point) | 22 |
| Biomass, biogases and bioliquids when complying with EU Regulation 73/2009 | 28 |
| Landfill gas, sewage treatment plant gas, biogases and bioliquids | 18 |

Fig. 40. All-inclusive feed in rate in Italy (GSE, 2011).

4.2. Algae oil costs.

We made a simple estimation of the produced oil cost for the different scenarios, assuming a payback of capital costs in ten years. Oil costs are reported in table 43.

Table 43. Algae oil costs for the different scenarios.

| Scenario | A | B | C | D | E | F | G | H | I |
|--------------------------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| Capital costs [total/10 years] | -€ 4.858 | -€ 4.858 | -€ 4.858 | -€ 4.858 | -€ 4.858 | -€ 4.858 | -€ 4.858 | -€ 4.858 | -€ 4.858 |
| Operational costs | -€ 11.462 | -€ 11.462 | -€ 11.462 | -€ 11.462 | -€ 11.462 | -€ 11.462 | -€ 11.462 | -€ 11.462 | -€ 11.462 |
| Revenue from biomass | € 5.366 | € 6.707 | € 8.048 | € 4.599 | € 5.749 | € 6.899 | € 3.833 | € 4.791 | € 5.749 |
| Cost of oil production | -€ 10.955 | -€ 9.613 | -€ 8.272 | -€ 11.721 | -€ 10.571 | -€ 9.422 | -€ 12.488 | -€ 11.530 | -€ 10.571 |
| Oil production [t/(ha yr)] | 21,9 | 27,375 | 32,85 | 29,2 | 36,5 | 43,8 | 36,5 | 45,625 | 54,75 |
| Cost [€/t] | € 500 | € 351 | € 252 | € 401 | € 290 | € 215 | € 342 | € 253 | € 193 |
| Cost [€/kg] | € 0,50 | € 0,35 | € 0,25 | € 0,40 | € 0,29 | € 0,22 | € 0,34 | € 0,25 | € 0,19 |

In algae oil cost estimation, we did not take into account interests and taxes. For this reason we consider the highest resulting price (0,50 €/kg) in determining the economical convenience of the combined heat and power cogeneration system.

4.3. Economic analysis.

We took into account maintenance and insurance costs as proportional to the produced energy; a cost of 1,8 c€/kWh_e has been considered as inclusive of both. The system is used in order to produce the maximum amount of electrical energy. Thermal energy is utilized in the algae production processes and does not provide revenues. Typically cogeneration

systems are able to work for over 8000 hours per year. Table 44 shows the budget for the proposed Siatem's 485 kW system, which has a plant cost of € 700.000.

Table 44. Budget for the cogeneration system.

| Cogeneration Budget | | |
|--------------------------------------|--|--------------------|
| Electric Energy Produced [kW] | | 485 |
| Hours of working [per year] | | 8.000 |
| Energy Production [kWh/year] | | 3.880.000 |
| Vegetal oil cost [€/kg] | | 0,50 |
| Oil use [kg/h] | | 107 |
| Vegetal oil cost [€/h] | | 54 |
| Plant cost | | € 700.000 |
| Oil costs per year | | € 428.000 |
| Maintenance and insurance costs | | € 69.840 |
| Yearly total costs | | € 497.840 |
| Feed-in tariff [€/kWh _e] | | € 0,28 |
| Heat revenues [€/kWh _t] | | € - |
| Yearly total revenues | | € 1.086.400 |

In table 45 we present the pre-tax profit and loss account. The considered scenario has a lifespan of ten years. The economic indicators of the cogeneration investment are very positive. The project has a payback period of 2 years, a net present value of over 7 times the initial investment and a very high IRR of 83,9%. Furthermore, feed-in incentives are applied over a period of 15 years, so the investment can have better economic results when a longer period than 10 years is considered.

Table 45. Cogeneration pre-tax profit and loss account.

| Year | Revenues | Operative Costs | Operative Profits | Cash Flow | Acc. Cash Flow | NPV |
|------|---------------------|---------------------|--------------------|--------------------|----------------|-----------------------|
| 0 | | | | -€ 700.000 | -€ 700.000 | € 4.342.551 |
| 1 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | -€ 111.440 | |
| 2 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 477.120 | Payback period |
| 3 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 1.065.680 | 2 years |
| 4 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 1.654.240 | |
| 5 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 2.242.800 | IRR |
| 6 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 2.831.360 | 83,9% |
| 7 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 3.419.920 | |
| 8 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 4.008.480 | |
| 9 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 4.597.040 | |
| 10 | € 1.086.400 | -€ 497.840 | € 588.560 | € 588.560 | € 5.185.600 | |
| | € 10.864.000 | -€ 4.978.400 | € 5.885.600 | € 5.185.600 | | |

CHAPTER 8

Conclusions.

The goals of this thesis were to analyze the production of biofuels from microalgae, to discuss the different technologies and processes and to determine if a best solution exists and whether the different technologies are cost-effective or not. In this final part we will first discuss the achieved results, and then the future perspectives and technological developments.

1. Achieved results.

Our economic analysis has showed that a mass production of algal biofuels in open ponds is possible and economically sustainable. Assuming a productivity of 30 g/(m² day) and oil content of 50 %, a net present value of over six times the equity is possible, with 80% of the investment financed by a bank loan, an analyzed timeline of ten years, inflation rate of 2,5% and interest rate of 6%.

Nowadays commercial mass production plants do not exist, and only few economic analyses have been published. Conversely industrial secrets and commercial (not demonstrated) claims abound. Our assumed productivities and oil yields are on the line with the latest researches and laboratory results.

Several additional assumptions have been made in order to enhance the feasibility of the project. The most relevant are: free CO₂, low water and nutrient costs, not purchasing the used land and a system of huge dimensions.

Starting from the latter point, our goal was the analysis of mass production of biofuels. From this point of view, great dimension is a justified supposition. As discussed in chapter 7, free CO₂, water medium and nutrients are possible when the system is combined with a power plant and a wastewater facility. However, CO₂ in large quantities may be not free. Richardson et Al. (2009) discussed the assumption of free CO₂ and analyzed an innovative algae test farm where also an alternative solution is used. In this test facility aggressively bubbling in atmospheric CO₂ produces a healthy algae population that is capable of high levels of oil production, without the need of industrial CO₂. Though, we are confident in the possibility of coupling the algae facility to industrial plants, because of the future emission credits that probably will give a further economic spin to the already existing synergies. Renewable fuels can be considered a policy driven industry. Assuming the rent of land is really positive to the investment's budget. An open ponds farm needs an adaptation of the land, not a major

change. Thus if the algae facility is dismissed, the land can be reconverted quite easily. Furthermore if the investors buy the land, its value most likely will not decrease over time.

The economics of the process could be improved when high-value co-products are produced but this will require additional treatment processes. High valued products may be difficult to produce in open ponds, where a low level of control is obtainable. Moreover the connection with wastewater facility and power plants can bring contaminants in the cultivation.

Production of biofuels from photobioreactors is not economically feasible. Capital costs are way higher than for open ponds and revenues from biofuels are relatively low. It is not possible to produce cheap biofuels in very expensive bioreactors; a photobioreactors cost reduction is necessary. Nowadays photobioreactors, which allow great control over the culture, are used to produce high priced co-products.

Also for open facilities biodiesel may not be the primary source of income; not only other products from microalgae but also electricity generation from the produced biomass and oil could be a positive addition to the profits. For example in some nations, such as Italy, feed-in tariffs incentivize the production of electrical energy, while incentives for the production of transportation fuels do not exist at the moment.

Algae production for transportation fuels still has a mild attention from investors, but recently the consideration of stockholders towards the process is increasing quickly. There is much interest with lots of developers searching for investors.

Besides all the treated arguments, we are confident that biofuels from algae will have a bright future because, as stated in chapter 1, they address a market where the demand is increasing and the offer is limited. Higher crude oil costs will enhance the feasibility of the project. In particular, in the last few days (late February 2011) the crisis in northern Africa created a peak of crude oil prices, which reached over 105 \$/barrel in the European market (BRENTD), when the assumed price in the economic analysis in chapter 6 is \$ 87.

2. Future perspectives and technological developments.

Our work demonstrated that producing algal biofuels is feasible. Nevertheless there are still several uncertainties.

Despite the very real promise in algal technology for biofuel production and carbon recycling there is much basic science to be done to allow its deployment in large scale. This includes integration of various steps on the algal value chain for overall economic success, such as coupling the process with nutrients from wastewater and using waste heat, and designing fuel production processes where less dewatering is necessary.

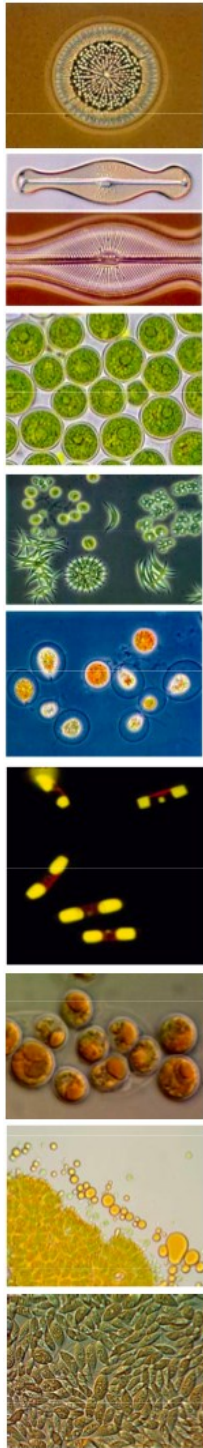


Fig. 41. Different algal strains (Hu and Sommerfeld, 2008)

Even if the production of fuels from microalgae is feasible, there is limited technical expertise and biofuels from algae has been made only in laboratories.

Producing algal biofuels requires large-scale cultivation and harvesting systems, with the challenging of reducing the cost per unit area. At a large scale, the algal growth conditions need to be carefully controlled and optimum cultivating environment have to be provided. The first investors will face risks and uncertainties in demonstrating large-scale low cost algal cultivation.

The main difficulty in efficient biodiesel production from algae lie in finding a strain with high lipid content and fast growth rate that is easy to growth, resistant to contaminant and not too difficult to harvest, all in a large scale system. To isolate and select an algal strain for mass culture is the first target for future research and development. Fig. 41 shows the high diversity in algal strains.

Mutagenesis and genetic modification of algae has received so far little attention despite molecular biology procedures appear essential in order to obtain domestication of algal strains for cultivation in mass culture. In particular, researches will need to focus in finding the ‘lipid trigger’ or channeling the algal metabolism toward oil accumulation for biodiesel production.

From a different side, strategic planning and political and economic support are important for a successful development of the technology. The most evident example is the doubt regarding incentives; a clear policy will drive the industry and the investments. A biorefinery based production strategy, can reduce the cost of producing microalgal biodiesel; like a petroleum refinery, a biorefinery uses every component of the biomass raw material to produce useable products.

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