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Greenhouse gas emissions and energy balance of biodiesel production from microalgae cultivated in photobioreactors in Denmark: a life-cycle modeling

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Abstract

The current use of fossil fuels is problematic for both environmental and economic reasons and biofuels are regarded as a potential solution to current energy issues. This study analyzes the energy balances and greenhouse gas emissions of 24 different technology scenarios for the production of algal biodiesel from *Nannochloropsis* cultivated at industrial scale in photobioreactors in Denmark. Both consolidated and pioneering technologies are analyzed focusing on strengths and weaknesses which influence the performance. Based on literature data, energy balance and greenhouse gas emissions are determined in a comparative 'well-to-tank' Life Cycle Assessment against fossil diesel. Use of by-products from biodiesel production such as glycerol obtained from transesterification and anaerobic digestion of residual biomass are included. Different technologies and methods are considered in cultivation stage (freshwater vs. wastewater; synthetic CO_2 vs. waste CO_2), harvesting stage (flocculation vs. centrifugation) and oil extraction stage (hexane extraction vs. supercritical CO_2 extraction). The choices affecting environmental performance of the scenarios are evaluated. Results show that algal biodiesel produced through current conventional technologies has higher energy demand and greenhouse gas emissions than fossil diesel. However, greenhouse gas emissions of algal biodiesel can be significantly reduced through the use of 'waste' flows (nutrients and CO_2) but there are still technical difficulties with both microalgae cultivation in wastewater as well as transportation and injection of waste CO_2. In any way, a positive energy balance is still far from being achieved. Considerable improvements must be made to develop an environmentally beneficial microalgae biodiesel production on an industrial scale. In particular, different aspects of cultivation need to be enhanced, such as the use of wastewater and CO_2–rich flue gas from industrial power plants.

HIGHLIGHTS

- The best existing technologies for algal biodiesel production via PBRs have been compared.
- Fossil diesel has been taken as reference product.
- Energy balance and greenhouse gas emissions have been evaluated.
- Algal biodiesel has higher impacts compared to fossil diesel.
- Great improvements must be achieved to develop algal biodiesel on industrial scale

KEYWORDS

Biofuel; Renewable fuels; Biorefinery; Bioenergy; Biogas; *Nannochloropsis*

1 Introduction

The use of fossil fuels is increasingly problematic from both an economic and an environmental point of view. It has been necessary to identify compatible mitigation strategies to avoid the exhaustion of fossil fuels and minimize the excess of CO_2 emissions related to energy production (Ribeiro et al., 2007). In recent times, the European Commission has presented the EU Directive 2009/28/CE aiming to establish a target of 20% share of renewable energy sources in energy consumption by 2020. In this context at least 10% of the energy for transportation must be based on renewable energy sources (European Commission, 2009). As a renewable energy source, biofuels are an attractive alternative to current petroleum based fuels (Festel et al., 2014). Biofuels refer to liquid, gas and solid fuels derived from biomass, including a.o. dedicated energy crops, residues from agriculture, and algae. Biofuels are classified as first (from crop based feedstock), second (from non-food feedstock), third (from algae) and fourth (from genetically engineered crops) generation fuels on the basis of the biomass origin and production technology (Demirbas, 2011; Lü et al., 2011; Liew et al., 2014).

Due to several features, algae are regarded as a promising source of biofuels and are considered an interesting alternative to current biofuel crops (Singh et al., 2011; Aitken et al., 2014). The production of fuel from algae provides many advantages: algae do not compete with land use and crop production since they are aquatic organisms; their growth rate is higher than that of terrestrial plants from which the first-generation biofuels derive (Scott et al., 2010); they do not need chemicals, herbicides, pesticides for growth (Kumar et al., 2010; Yang and Chen, 2012); they can remove nitrogen and phosphorus from wastewater (Clarens et al., 2010); and, under certain conditions, such as nitrogen stress, algae are characterized by high lipid accumulation, a feature that increases biofuel production (Rodolfi et al., 2009).

On the other hand, there are several difficulties associated with the production of the third-generation biofuels and, until now, their commercial production has not been achieved on industrial scale in a cost-efficient manner (Biofuel.org.uk, 2010). Currently, only a few pilot plant projects have been developed (e.g. BFS Bio Fuel Systems, 2015; All-gas, 2012). At present, microalgae have been commercially cultivated only to obtain valuable products like carotenoids (β-carotene and astaxanthin) and long-chain poly-unsatured fatty acids (Hannon et al., 2010). The main challenge that the algae biofuels sector is facing is to reduce capital and operating costs and so far only few studies have suggested the development of biodiesel production from microalgae on a commercial scale (Brentner et al., 2011; Sevigné Itoiz et al., 2012).

Cultivation of microalgae can be done in open systems (lakes, ponds) or in controlled closed systems called photobioreactors (PBRs). Open ponds and lagoons have lower costs but also suffer from low productivity and contamination problems. PBRs enhance productivity, avoid cultivation contamination and are more reliable but they have high capital construction and operating costs (Demirbas, 2010; Benson et al., 2014). In both open and closed systems, there is a high energy requirement for mixing water with nutrients and CO_2 during the cultivation stage (Rodolfi et al., 2009). Moreover, harvesting and dewatering of biomass lead to high costs for production facilities as well as a high energy use (Brennan and Owende, 2013).

As part of the increasing research activities on algal biofuels, several Life Cycle Assessment (LCA) studies on biodiesel production from algae have been performed in order to assess their environmental performances. The results of these LCAs are conflicting, showing that only under specific conditions and assumptions the third-generation biofuels could be energetically and environmentally sustainable (Lardon et al., 2009; Khoo et al., 2011; Holma et al., 2013).

This study takes origin in the encouraging results obtained by Brentner et al., 2011 on flat panel PBRs hypothetically located in Phoenix, AZ. The location of PBRs has been moved to Denmark, and a variety of technologies and implementation strategies to produce biodiesel from microalgae has been analyzed. Some of these technologies have already been developed on an industrial scale to produce valuable algal compounds while others are still on an experimental laboratory scale. Combing through different technologies in the different production stages, a total of 24 scenarios have been created. The energy demand and GHG emissions of the 24 scenarios and of the fossil diesel have been benchmarked and compared using a 'well-to-tank' life cycle approach. The sensitivity of some parameters that could affect biodiesel production have been evaluated.

2 Material and methods

This study applies Life Cycle Assessment (LCA) to evaluate the environmental performance of the different scenarios. LCA quantifies the environmental impacts of a product system considering its entire life cycle and is standardized by ISO 14040/14044 (ISO, 2006a and 2006b). The method has four phases: goal and scope definition, life cycle inventory (LCI), life cycle impact assessment (LCIA), and interpretation of results. Below, data used and approaches applied in each of these phases are described.

2.1 Goal and scope definition

The goal of this LCA study was to assess algal biodiesel production on a hypothetical commercial scale by analyzing and comparing both consolidated (from algae-based industry) and pioneering technologies, focusing on strengths and weaknesses which influence the performance.

Assuming 39.35 MJ/kg as high heating value (HHV) (Brentner et al., 2011), the functional unit was 1 MJ of biodiesel and the system boundaries were 'well-to-tank' (i.e. from cultivation to biodiesel storage). The stages included were (Fig. 1): cultivation, harvesting, drying, oil extraction, transesterification, anaerobic digestion of residual biomass with subsequent biogas combustion to generate energy (Zhang et al., 2013), and use of the by-product glycerol for the synthesis of propylene glycol. Substitution by system expansion was considered for biogas production and glycerol use. Substitution of glycerol in the production of propylene glycol has been chosen since this use is claimed to be the most economically attractive within the chemical industry (Pagliaro and Rossi, 2010). The algae selected was *Nannochloropsis* cultivated in flat panel PBRs and the production was assumed to be located in Denmark. Manufacturing, facilities maintenance, and use of infrastructures were not taken into account, except for the materials used for PBRs. The PBRs manufacturing is included since Sevigné-Itoiz et al. (2012) state that construction of PBRs contributes significantly to energy use and environmental impacts. On the other hand, Brentner et al. (2011), who included also construction materials in the assessment, find that those materials contribute less than 1% to the cumulative energy demand (CED). The biodiesel combustion is not included by system boundaries.

2.2 Life cycle inventory (LCI)

All main inventory data are shown in Table 1. As indicated most of the data were compiled from previous works and were adapted to a Danish scenario (Table 2). The databases used for obtaining the additional process data were Gabi Professional 2006 (PE International, 2007) and Ecoinvent 2.2 (Ecoinvent Centre, 2007).

2.2.1 Scenarios

A summary of the cultivation system and technologies assumed for each of the 24 scenarios are reported in Table 3. As shown, cultivation in either freshwater (scenarios from 1 to 6) or wastewater (scenarios from 7 to 12) were considered. The algae require an injection of CO_2 into the growth medium for optimal growth and each scenario alternatively assumed the use of either pure CO_2 (where the carbon dioxide is delivered in tanks) or waste CO_2 (named wCO_2, with flue gas pumped from a nearby cement production plant into the PBRs). In the harvesting stage, three techniques were assessed: flocculation with aluminum sulfate (scenarios 1, 4, 7, 10), flocculation with lime (scenarios 2, 5, 8, 11), and centrifugation (scenarios 3, 6, 9, 12). Finally, both hexane extraction (scenarios 1, 2, 3, 7, 8, 9) and sCO_2 (supercritical CO_2) extraction (scenarios 4, 5, 6, 10, 11, 12) were assessed in the oil extraction stage.

Consolidated technologies of the current market (i.e. flocculation, centrifugation, extraction with hexane, algal cultivation in freshwater and with pure CO_2) have thus been compared with advanced technologies not implemented on large scale (i.e. use of wastewater and waste CO_2, and extraction with sCO_2). The next sections, describe each stage in details.

2.3 Algal biomass cultivation and harvesting

Inventory data for cultivation and harvesting are showed in Table 1 and parameters used for modeling the *Nannochloropsis* cultivation in PBRs are illustrated in Table 4.

The wastewater scenarios did not involve synthetic nutrients since wastewater is supposed to contain an adequate amount of nutrients to serve as a suitable growth medium for microalgae (Pittman et al., 2011). The CO_2 taken up during algal growth was subtracted from the total amount of CO_2 emissions in both 'pure CO_2' and 'waste CO_2' scenarios, whereas the CO_2 emissions from the production process of pure CO_2 are accounted for.

The water content of wet algal biomass after harvesting is assumed to be about 70% (Singh et al., 2012).

2.4 Drying and algal oil extraction

Inventory data for drying and algal oil extraction are showed in Table 1. Drying stage was only assumed to be a requirement for hexane oil extraction since sCO_2 extraction is carried out directly from wet biomass (Xu et al., 2011; Mendes et al., 1995).

We assumed the use of thermal dryers with an energy consumption around 3.3 MJ per kilogram of evaporated water (Xu et al., 2011).

A dry biomass content in *Nannochloropsis* of 29% lipid, 10% carbohydrates and 30% proteins is hypothesized (Rodolfi et al., 2009; Razon and Tan, 2011). According to Brentner et al. (2011), the extraction efficiency with hexane is assumed to be 0.91.

Supercritical CO_2 for algal lipids extraction has been applied in laboratory on a number of algal species: *Skeletonema costatum* and *Ochromonas danica* (Polak et al., 1989), *Chlorella vulgaris* (Mendes et al., 1995), *Botryococcus braunii*, *Dunaliella salina*, *Arthrospira maxima* (Mendes et al., 2003), *Haematococcus pluvialis* (Thana et al., 2008). Recently, experiments have also been started on *Nannochloropsis sp*. (Andrich et al., 2005; Douglas, 2011; Crampon et al., 2013) but little information is reported on extraction efficiency even if the authors analyze the effects of operating conditions on the kinetics of the supercritical fluid extraction (Andrich et al., 2005; Crampon et al., 2013; Baskette, 2015). In scenarios assuming extraction with supercritical CO_2, 27.5 MPa and 47.5 °C were chosen as operating conditions (Mendes et al., 1995) and the extraction efficiency is assumed to be equal to the one with hexane (0.91). Neither hexane nor CO_2 recycling were considered in the LCI analysis.

2.5 Transesterification and use of glycerol

The amount of electricity and heat used in transesterification stage are shown in Table 1. The conversion efficiency was hypothesized 98% (Brentner et al., 2011) and the catalyst used was methanol. The avoided production of propylene oxide has been calculated on the basis of the stoichiometric ratio and the process yields of the involved reactions. Data for propylene oxide to propylene glycol were from Ecoinvent 2.2 (Ecoinvent Centre, 2007), data for glycerol to propylene glycol were from Pagliaro et al. (2007); the yields were 95% and 73%, respectively.

2.6 Life cycle impact assessment (LCIA)

The LCIA method applied was IMPACT 2002+ which proposes a feasible implementation of a combined midpoint/damage approach (Humbert et al., 2012). The chosen impact categories have been: global warming potential (GWP) and non-renewable energy consumption. For each scenario, the performances of algal biodiesel were compared with those of fossil diesel (from Ecoinvent 2.2; Ecoinvent Centre, 2007).

2.7 Sensitivity analysis

The sensitivity analysis estimates the influence of assumptions, i.e. changes in input parameters, on the model outcome (ISO, 2006a; ISO, 2006b). Among all the possible parameters to be considered for the sensitivity analysis, we have selected two. The first parameter is the extraction efficiency ranging from 0.91 in the Base case (extraction efficiency with hexane, Brentner et al., 2011) to 0.95 in the Case 1 (extraction efficiency with supercritical CO_2, Brentner et al., 2011).

The second parameter considered is the lipid content in the algal biomass which can vary dramatically as a result of the nitrogen supply (Jorquera et al., 2010; Khoo et al., 2011; Razon and Tan, 2011). The considered range of lipid content varies from 29% (lipid content experimentally observed in standard conditions by Rodolfi et al., 2009) to 60% (lipid content experimentally observed under nitrogen deprivation conditions by Rodolfi et al., 2009).

3 Results and discussion

The results generally show that 'pure' CO_2 (grey columns, Fig.2 and Fig.3) causes GHG emissions and energy consumption at least 25%-30% higher than waste CO_2 (white columns, Fig.2 and Fig.3). This agrees well with the results obtained by Borkowski et al. (2012) which demonstrated that the use of waste CO_2 for algae cultivation in PBR from a nearby power plant decreased GHG emissions by about 50% compared to the use of ‘pure’ CO_2.

In general, GWP of biodiesel scenarios is one order of magnitude higher than GWP of fossil diesels (black column, Fig.2). Only the last three scenarios (Sc10-wCO_2, Sc11-wCO_2 and Sc12-wCO_2) show GHG emissions similar to or lower than those of fossil diesel. The last three scenarios achieve the best performances also considering non-renewable energy consumption (Fig. 3), even if this is considerably higher compared to fossil diesel. This indicates that the coupling of the ‘waste flows’ for algal cultivation with the use of sCO_2 for algal oil extraction – that avoids the drying stage – could be an interesting production system. The best scenario is Sc10-wCO_2 (flocculation with aluminum sulphate) which shows a negative GWP indicating a GHG sequestration and the lowest energy consumption. The result is in accordance with the studies by Lardon et al. (2009) which observed that only wet extraction can save GHG emissions in algal biodiesel production and by Vasudevan et al. (2012) which calculated very low GHG emissions (0.053 kg of CO_2 eq/MJ) when wet extraction was applied. Also Xu et al. (2011) observed that wet extraction dramatically decreases energy consumption.

Interesting information is provided by the ‘non-renewable energy investment in energy delivered’ (NEIED) (Yang and Chen, 2012). NEIED is expressed as the ratio between the non-renewable energy used directly and indirectly in the production process and the energy content in the biofuel. In this study the NEIED is >1 in all 24 scenarios. In particular, in our simulations algal biodiesel production requires from 20 MJ (Sc10-wCO_2) to 90 MJ (Sc3-CO_2) for producing 1 MJ of biodiesel. These values are very high but comparable with results obtained by other authors. Jorquera et al. (2010) find a consumption of about 14 MJ/MJ for tubular PBRs including only cultivation stage and Sevigné Itoiz et al. (2012) report a consumption of 901 MJ/kg of DW biomass for indoor PBRs. In fact, cultivation in PBRs has a large energy demand due to the CO_2 pumping and nutrients mixing (Weinberg et al., 2012; Borkowski et al., 2012; Khoo et al., 2011).

Below we evaluate the relative contributions to GWP and non-renewable energy consumption of each stage in the worst (Sc3-CO_2) and the best scenarios (Sc10-wCO_2). The stages analyzed are: 1) algae cultivation; 2) harvesting; 3) (drying and) oil extraction; 4) transesterification; 5) anaerobic digestion (of residual biomass) and 6) use of glycerol.

Figure 4 illustrates the relative contributions in the worst scenario. As shown, the cultivation stage has the highest contribution to GWP and non-renewable energy consumption (62% and 66%, respectively), followed by drying and oil extraction (23% and 24%, respectively) and harvesting through centrifugation (15% and 13%, respectively). Anaerobic digestion contributes by avoiding GHG emissions and non-renewable energy consumption (both about -2%) while transesterification and use of glycerol in the propylene oxide industry do not give a relevant contribution. These results completely agree with previous studies. Many authors observed that cultivation (Batan et al., 2010; Borkowski et al., 2012; Weinberg et al., 2012), drying (Razon and Tan, 2011; Xu et al., 2011) and lipid extraction (Khoo et al., 2011) were the most impacting stages for biodiesel production both in terms of GHG emissions and energy requirements.

Figure 5 illustrates the best scenario. As far as GWP concerns, cultivation (-40%) and anaerobic digestion (-25%) contribute by avoiding GHG emissions while the most impacting stages are harvesting (15%) and sCO_2 extraction (15%). Transesterification and the glycerol use are negligible. Regarding non-renewable energy consumption, the most significant process is algae cultivation (92%) while the other stages have a very low contribution. The negative contribution of cultivation on GHG emissions is due to the sequestration of CO_2 in the algal cells and to the use of wastewater which eliminate the need of fertilizer production. However, these improvements do not eliminate the need of electric power during the cultivation stage. As a final result, in the best-case scenario we have an increment of the cultivation stage contribution to the energy consumption. This is due to the fact that the energy demand of the algal harvesting and lipid extraction stages decreases in comparison to the worst case.

The percentage contribution analysis has identified three stages as the bottlenecks of algal biodiesel production: cultivation, drying and oil extraction, and harvesting. Regarding the cultivation stage, the contribution of the different processes to the environmental impact are detailed in the Supplementary Data, figures 2.1, 2.2, 2.3 and 2.4. Electricity is always a significant contributor and when 'pure' CO_2 and/or nutrients are required these contribute significantly as well. The contributions of nutrients, CO_2, and electricity vary for the different scenarios. Contributions from construction materials, low density polyethylene (LDPE) sheets and reinforcing steel, are always negligible. Considering the performances of Sc10-wCO_2, Sc11-wCO_2 and Sc12-wCO_2 scenarios, it is evident that the capability to cultivate algae using waste flows (aqueous and gaseous) plays a fundamental role for an environmental beneficial development of large scale biodiesel production from microalgae. Anyway, these technologies need to be improved further to become efficient, affordable and accessible. Currently, the cultivation of algae in wastewater has not been developed on commercial scale yet but only on pilot plants. Several challenges exists, e.g. the high turbidity of wastewater restricting the light penetration and making the algal cultivation inefficient (Pedroni et al., 2001). Therefore, a water clarification pre-treatment could be necessary in order to reduce the presence of suspended matter and organic load (Pedroni et al., 2001). Also the use of waste CO_2 is still experimental on a pilot scale. The main issues to be solved are the transfer of waste flue gas from an industrial plant to PBRs and the CO_2 losses during this transfer. In fact, the energy demand for pumping the flue gas and the distance from the plant to PBRs limit this transfer (Pedroni et al., 2001). Moreover, it is challenging to control the O_2-concentration and the temperature which has to be reduced from above 100℃ to app. 25℃ (Dorminey, 20 13). Additionally, flue gases contain pollutants such as NOx and SO_2 which may have adverse effects on the algal species. However, first findings from studies reveal that the presence of pollutants in the flue gas in today's industrial emissions seems to be less of a problem in relation to the growth of the algae (Mortensen and Gislerød, 2014).

The drying and oil extraction stage is the second relevant bottleneck. Oil extraction with the sCO_2–process decreases the impact contribution because it does not require drying of the algal biomass. Also in this case, the 'key' technology is very innovative and must be further enhanced. Mendes et al. (1995) observed that higher pressures and temperatures led to higher efficiencies in the extraction of lipids but Santana et al. (2012) found a correlation between the pressure and the presence of unsaturated compounds, i.e. high pressure leads to high amounts of unsaturated compounds in the algal oil thus reducing the biodiesel quality.

In terms of energy consumption and GHG emissions, the harvesting stage also played a significant role. In general, flocculation requires less energy than centrifugation; in particular, *Nannochloropsis* centrifugation has a large energy demand due to the small size of the cells (Rodolfi et al., 2009). This in line with Sander and Murthy (2010) who also identified a high energy demand of centrifugation (50% higher than flocculation) in comparison to other algal harvesting technologies such as separation or filtration. Flocculation with aluminum sulphate (scenarios 1, 4, 7, 10) and with lime (scenarios 2, 5, 8, 11) show similar GHG emissions and energy performances, see Fig. 2 and Fig. 3. However, although flocculation requires less energy than centrifugation, both flocculants present some disadvantages. The main product of flocculation with aluminum sulphate is aluminum hydroxide which forms aggregates with algal biomass rendering it toxic for methanogens during anaerobic digestion (Demirbas, 2010). Even if lime is less toxic than aluminum sulphate, it is less used for flocculation due to the precipitate formation, i.e. $CaCO_3$, in the water (Pedroni et al., 2001).

A possible improvement with respect to both flocculation and centrifugation could be the development of bio-flocculation (Pedroni et al., 2001). Bio-flocculation is biologically induced by bacteria (Lee et al., 2009). Recently, a naturally flocculating diatom *Skeletonema* was used to form flocs of *Nannochloropsis* (Schenk et al., 2008). Bio-flocculation is not toxic for microalgae, it requires low operating costs, and has a low energy demand. However, bio-flocculation is affected by environmental conditions which are the most relevant aspects to improve (Schenk et al., 2008).

4 Sensitivity analysis

Tables 5A and 5B present the results of the sensitivity analysis. Increasing extraction efficiency from 0.91 to 0.95, results in lower values for GWP and non-renewable energy consumption (about 5% less than Basic Case).

Likewise, increasing the lipid content from 29% to 60% reduces both GHG emissions and energy consumption by app. 50%. Therefore, lipid content was confirmed as an important parameter for biodiesel production. Nonetheless, even with high lipid content the energy and GHG emissions performances of algal biodiesel are still inferior to those of diesel from fossil sources. These results are in agreement with the observation of Khoo et al. (2011) and Razon and Tan (2011). In particular, Khoo et al. (2011) demonstrated that increasing the lipid content by about 10% and 20% decreased the energy consumption by about 4% and 6%, respectively.

5 Conclusion

Algal biodiesel produced through current conventional technologies shows higher energy demand and GHG emissions than those of fossil diesel. 'Wastewater scenarios' coupled with waste CO_2 have the lowest impact in GHG-emissions and non-renewable energy consumption, in some cases even better than fossil diesel in terms of GHG-emissions. However, a positive energy balance is still far from being achieved by algal biodiesel. Thus, further improvements are required in order to achieve a beneficial development of biodiesel production on an industrial scale. In particular, different aspects of cultivation need to be enhanced, such as the use of wastewater as source of nutrient and CO_2–rich flue gas from industrial power plants as source of carbon. The research has been addressed towards algae cultivation with ‘waste flows’, that seems to be the key to reduce both the demand of energy and the GHG-emissions of biodiesel from microalgae. Additionally, the energy demand for mixing, pumping, etc. of the cultivation stage should be dramatically decreased. Considering the extraction, supercritical CO_2 extraction appears to be an interesting technology. However, further studies are needed to address the main limitations; how to achieve high temperatures and high pressures and still avoiding the formation of unsaturated compounds.

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Table 1

Summary of the inventory data for producing 1 MJ of algal biodiesel (HHV=39.35 MJ/kg of biodiesel).

FRESHWATER CULTIVATION	AMOUNT	UNIT	NOTES
Carbon dioxide	0.61	kg	Calculated from 1
Tap water	0.47	m^3	Calculated from 2
Electricity consumption	0.78	kWh	Calculated from 2
Ammonium nitrate	0.08	kg	Calculated from 3
Monocalcium phosphate	0.03	kg	Calculated from 3
WASTEWATER CULTIVATION			
Carbon dioxide	0.61	kg	Calculated from 1
Wastewater	0.47	m^3	Calculated from 2
Electricity consumption	0.78	kWh	Calculated from 2
FLOCCULATION			
Electricity consumption	0.05	kWh	Calculated from 2
Aluminium sulphate	0.04	kg	Calculated from 4
Lime	0.15	kg	Calculated from 5
CENTRIFUGATION			
Electricity consumption	0.11	kWh	Calculated from 6
DRYING			
Heat	1.12	MJ	Calculated from 7
EXTRACTION WITH HEXANE			
Electricity consumption	0.01	kWh	Calculated from 2
Heat	0.10	MJ	Calculated from 2
Hexane	0.39	g	Calculated from 5
SUPERCRITICAL CO_2 EXTRACTION			
CO_2 liquid	3.7	g	Calculated from 8
Electricity consumption	0.18	kWh	Calculated from 9
TRANSESTERIFICATION			
Electricity consumption	0.001	kWh	Calculated from 2
Heat	0.02	MJ	Calculated from 2
Methanol	2.9	g	Calculated from 5

1: Wijffels and Barbosa, 2010
2: Brentner et al., 2011
3: Grobbelaar, 2004
4: Grima et al., 2003
5: Lardon et al., 2009
6: Foley et al., 2011
7: Xu et al., 2011
8: Mendes et al., 1995
9: Singh and Olsen, 2012

Table 2

Parameters and processes used in the study adapted to the Danish situation.

PARAMETERS	AMOUNT	UNIT	REFERENCES
Denmark's electricity mix	-	-	Ecoinvent 2.2
Reference year of electricity mix	2004		Ecoinvent 2.2
Denmark's carbon intensity	0.2	kg CO_2/2005 US $	US EIA, 2015
Average solar irradiation in Denmark	3730	MJ/m2/y	Danish Meteorological Institute, 2013
Productivity days	200	n°	Danish Meteorological Institute, 2013
CO_2 emission from Danish cement industry	1420067	t/y	Singh and Olsen, 2012

Table 3

Summary of cultivation systems and technologies used for each analysed scenario.

CODE	CO_2 SOURCE	WATER SOURCE	HARVESTING MODE	EXTRACTION MODE
Sc1-CO_2	Pure CO_2	Tap water	Aluminum sulfate	With hexane
Sc1-wCO_2	Waste CO_2	Tap water	Aluminum sulfate	With hexane
Sc2-CO_2	Pure CO_2	Tap water	Lime	With hexane
Sc2-wCO_2	Waste CO_2	Tap water	Lime	With hexane
Sc3-CO_2	Pure CO_2	Tap water	Centrifugation	With hexane
Sc3-wCO_2	Waste CO_2	Tap water	Centrifugation	With hexane
Sc4-CO_2	Pure CO_2	Tap water	Aluminum sulfate	Supercritical CO_2
Sc4-wCO_2	Waste CO_2	Tap water	Aluminum sulfate	Supercritical CO_2
Sc5-CO_2	Pure CO_2	Tap water	Lime	Supercritical CO_2
Sc5-wCO_2	Waste CO_2	Tap water	Lime	Supercritical CO_2
Sc6-CO_2	Pure CO_2	Tap water	Centrifugation	Supercritical CO_2
Sc6-wCO_2	Waste CO_2	Tap water	Centrifugation	Supercritical CO_2
Sc7-CO_2	Pure CO_2	Wastewater	Aluminum sulfate	With hexane
Sc7-wCO_2	Waste CO_2	Wastewater	Aluminum sulfate	With hexane
Sc8-CO_2	Pure CO_2	Wastewater	Lime	With hexane
Sc8-wCO_2	Waste CO_2	Wastewater	Lime	With hexane
Sc9-CO_2	Pure CO_2	Wastewater	Centrifugation	With hexane
Sc9-wCO_2	Waste CO_2	Wastewater	Centrifugation	With hexane
Sc10-CO_2	Pure CO_2	Wastewater	Aluminum sulfate	Supercritical CO_2
Sc10-wCO_2	Waste CO_2	Wastewater	Aluminum sulfate	Supercritical CO_2
Sc11-CO_2	Pure CO_2	Wastewater	Lime	Supercritical CO_2
Sc11-wCO_2	Waste CO_2	Wastewater	Lime	Supercritical CO_2
Sc12-CO_2	Pure CO_2	Wastewater	Centrifugation	Supercritical CO_2
Sc12-wCO_2	Waste CO_2	Wastewater	Centrifugation	Supercritical CO_2

Table 4

Parameters used for modelling the *Nannochloropsis* cultivation in PBRs.

PARAMETERS	AMOUNT	UNIT	REFERENCES
Nannochloropsis productivity	0.27	kg/m^3/day	Jorquera et al., 2010
Biomass productivity	37.8	t/ha/year	Singh and Olsen, 2012
Number of PBR	2667	per hectare	Brentner et al., 2011
PBR lenght	2.5	m	Brentner et al., 2011
PBR height	1.5	m	Brentner et al., 2011
PBR thick	0.070	m	Brentner et al., 2011
PBR volume	0.263	m^3	Brentner et al., 2011
Residence time	2.6	days	Brentner et al., 2011
Area	3.75	m^2	Brentner et al., 2011
LDPE sheet	0.011	kg/kg biomass	Brentner et al., 2011
Life time	50	years	Brentner et al., 2011
Steel	0.00085	kg/kg biomass	Brentner et al., 2011
Life time	50	years	Brentner et al., 2011

Table 5A

Results of the sensitivity analysis for GWP (kg CO2-eq). Basic case (91% extraction efficiency and 29% lipid content) compared to the increase of extraction efficiency (95%) and lipid content (60%). The functional unit is 1 MJ of biodiesel.

CODE	BASIC CASE	EXTRACTION EFFICIENCY 95%	LIPID CONTENT 60%
Sc1-CO_2	5.95E+00	5.70E+00	2.90E+00
Sc1-wCO_2	3.11E+00	2.98E+00	1.53E+00
Sc2-CO_2	6.23E+00	5.97E+00	3.04E+00
Sc2-wCO_2	3.39E+00	3.25E+00	1.67E+00
Sc3-CO_2	6.71E+00	6.43E+00	3.28E+00
Sc3-wCO_2	3.88E+00	3.72E+00	1.90E+00
Sc4-CO_2	4.60E+00	4.41E+00	2.25E+00
Sc4-wCO_2	1.77E+00	1.69E+00	8.83E-01
Sc5-CO_2	4.88E+00	4.68E+00	2.39E+00
Sc5-wCO_2	2.05E+00	1.96E+00	1.02E+00
Sc6-CO_2	5.37E+00	5.14E+00	2.62E+00
Sc6-wCO_2	2.54E+00	2.43E+00	1.25E+00
Sc7-CO_2	4.01E+00	3.84E+00	1.97E+00
Sc7-wCO_2	1.18E+00	1.13E+00	5.96E-01
Sc8-CO_2	4.29E+00	4.11E+00	2.10E+00
Sc8-wCO_2	1.46E+00	1.40E+00	7.32E-01
Sc9-CO_2	4.78E+00	4.58E+00	2.34E+00
Sc9-wCO_2	1.94E+00	1.86E+00	9.67E-01
Sc10-CO_2	2.66E+00	2.55E+00	1.32E+00
Sc10-wCO_2	-1.67E-01	-1.60E-01	-8.32E-02
Sc11-CO_2	2.94E+00	2.82E+00	1.45E+00
Sc11-wCO_2	1.13E-01	1.08E-01	5.61E-02
Sc12-CO_2	3.43E+00	3.29E+00	1.69E+00
Sc12-wCO_2	5.99E-01	5.74E-01	3.02E-01

Table 5B

Results of the sensitivity analysis for non-renewable energy consumption (MJ). Basic case (91% extraction efficiency and 29% lipid content) compared to the increase of extraction efficiency (95%) and lipid content (60%). The functional unit is 1 MJ of biodiesel.

CODE	BASIC CASE	EXTRACTION EFFICIENCY 95%	LIPID CONTENT 60%
Sc1-CO_2	8.27E+01	7.92E+01	4.03E+01
Sc1-wCO_2	6.51E+01	6.24E+01	3.18E+01
Sc2-CO_2	8.36E+01	8.01E+01	4.08E+01
Sc2-wCO_2	6.60E+01	6.33E+01	3.23E+01
Sc3-CO_2	9.27E+01	8.88E+01	4.52E+01
Sc3-wCO_2	7.51E+01	7.20E+01	3.67E+01
Sc4-CO_2	6.26E+01	6.00E+01	3.06E+01
Sc4-wCO_2	4.50E+01	4.32E+01	2.21E+01
Sc5-CO_2	6.36E+01	6.09E+01	3.11E+01
Sc5-wCO_2	4.60E+01	4.41E+01	2.26E+01
Sc6-CO_2	7.26E+01	6.96E+01	3.55E+01
Sc6-wCO_2	5.51E+01	5.28E+01	2.70E+01
Sc7-CO_2	5.93E+01	5.69E+01	2.91E+01
Sc7-wCO_2	4.18E+01	4.00E+01	2.06E+01
Sc8-CO_2	6.03E+01	5.78E+01	2.95E+01
Sc8-wCO_2	4.27E+01	4.09E+01	2.10E+01
Sc9-CO_2	6.94E+01	6.65E+01	3.39E+01
Sc9-wCO_2	5.18E+01	4.96E+01	2.54E+01
Sc10-CO_2	3.93E+01	3.77E+01	1.94E+01
Sc10-wCO_2	2.17E+01	2.08E+01	1.09E+01
Sc11-CO_2	4.02E+01	3.86E+01	1.98E+01
Sc11-wCO_2	2.26E+01	2.17E+01	1.13E+01
Sc12-CO_2	4.83E+01	4.73E+01	2.42E+01
Sc12-wCO_2	3.17E+01	3.04E+01	1.57E+01

Figure captions

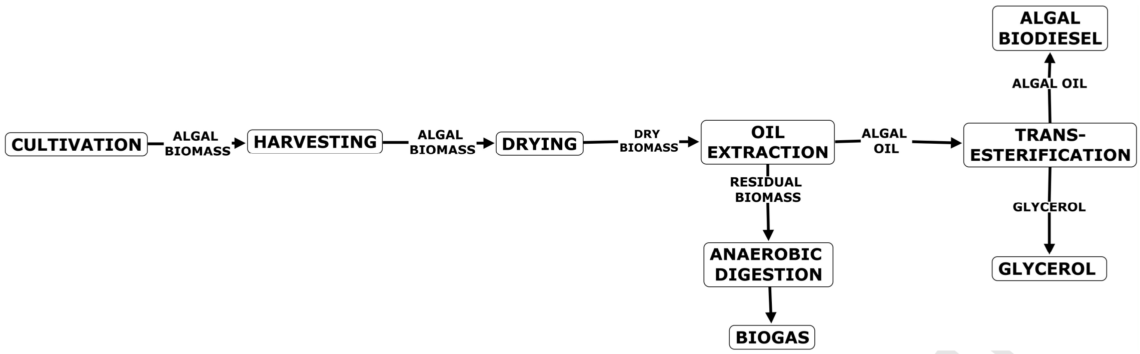
Fig. 1 System boundaries of biodiesel production and the most important flows used for each stage.

Fig. 2 GWP (kg CO_2-eq) of all 24 scenarios. CO_2 indicates the use of ‘pure’ CO_2 (grey column) for algae cultivation whereas wCO_2 specifies the use of waste CO_2 (white column) in microalgae cultivation stage. All scenarios have been compared to fossil diesel (black column, Ecoinvent Centre, 2007).

Fig. 3 Non-renewable energy consumption (MJ) of all 24 scenarios. CO_2 indicates the use of industrial CO_2 (grey column) for algae cultivation whereas wCO_2 specifies the use of waste CO_2 (white column) in microalgae cultivation stage. All scenarios have been compared to fossil diesel (black column, Ecoinvent Centre, 2007).

Fig. 4 Relative contribution of each stage of the worst scenario, which assumed the use of freshwater and ‘pure’ CO_2 for algae cultivation, centrifugation for algal harvesting and algal oil extraction with hexane. (Read the legend from top to bottom)

Fig. 5 Relative contributions of each stage of the best scenario, which assumed the use of wastewater and waste CO_2 for algae cultivation, flocculation with aluminium sulphate for algal harvesting and sCO_2 extraction in algal oil extraction. (Read the legend from top to bottom)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CULTIVATION
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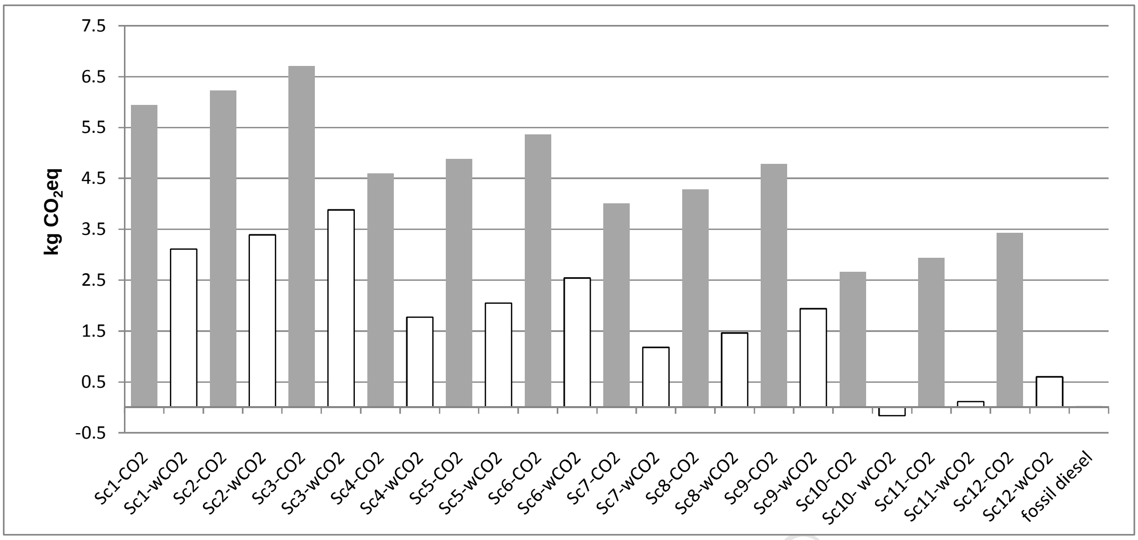
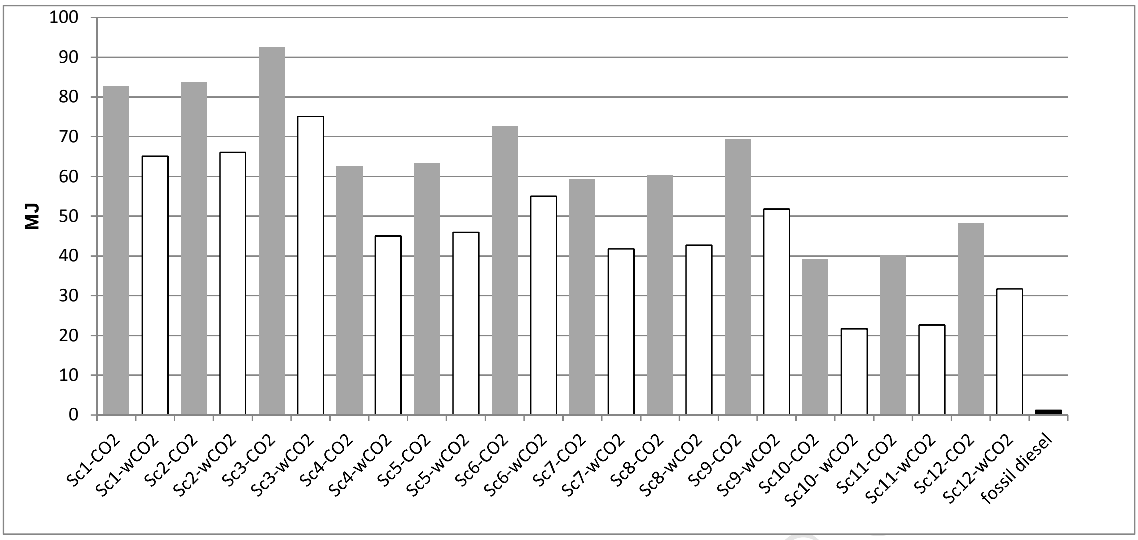
HARVESTING
ALGAL BIOMASS
DRYING
DRY BIOMASS
OIL EXTRACTION
ALGAL OIL
TRANS-ESTERIFICATION
RESIDUAL BIOMASS
GLYCEROL
ANAEROBIC DIGESTION
GLYCEROL
BIOGAS

Fig. 1

656



657
658 Fig. 2
659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Sc3-wCO2
Sc4-CO2
Sc4-wCO2
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Sc9-wCO2
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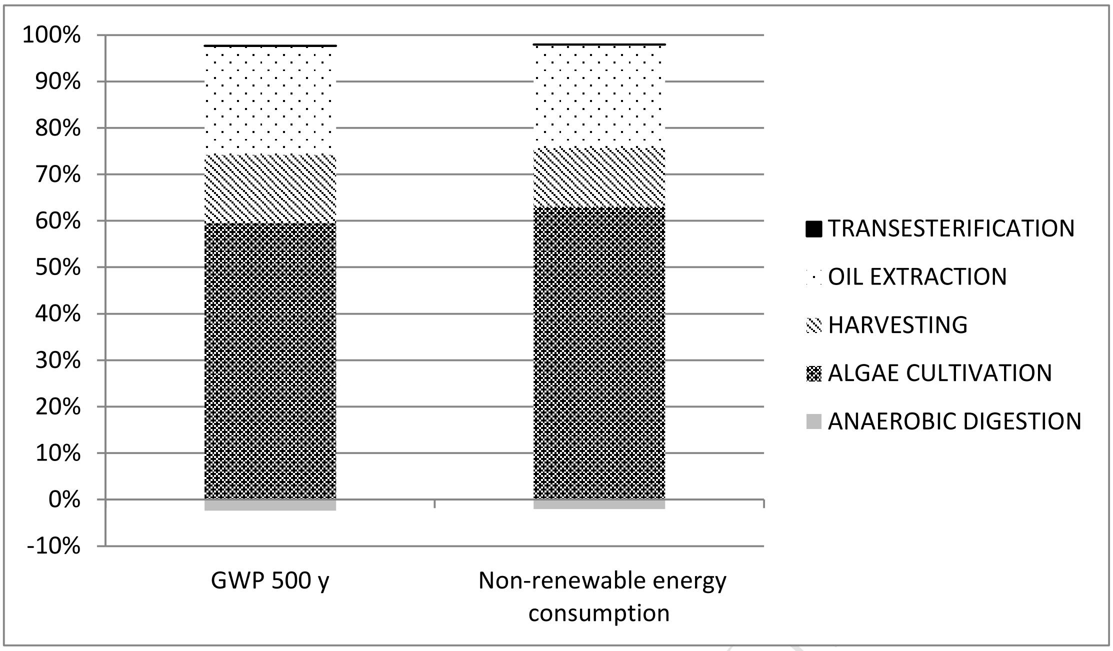
fossil diesel

Fig. 3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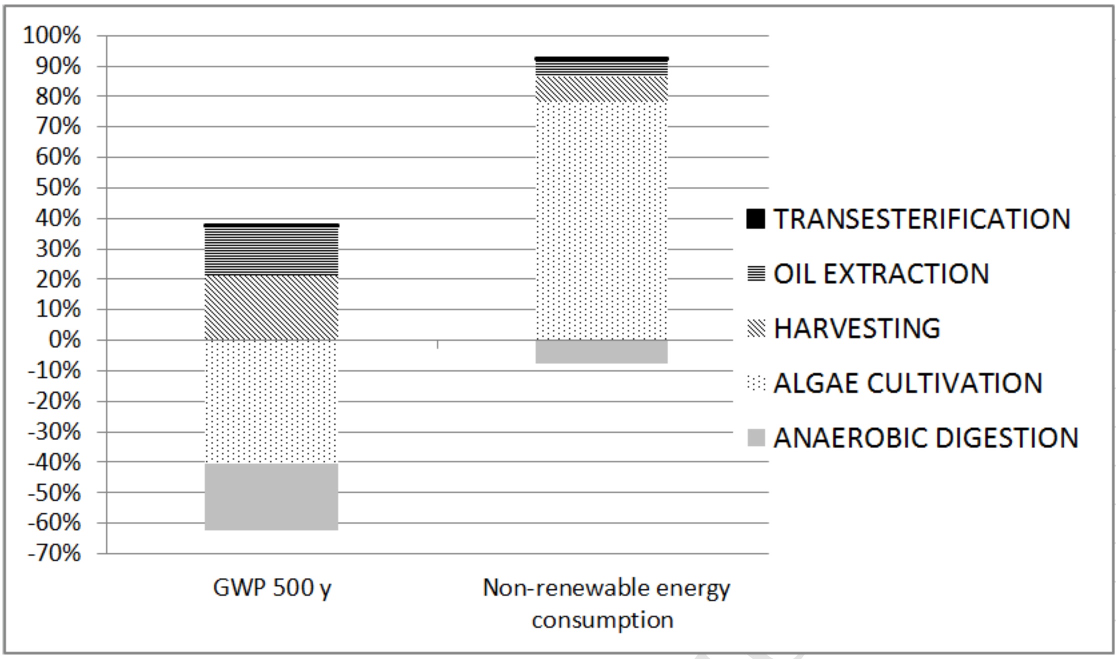
OIL EXTRACTION
HARVESTING
ALGAE CULTIVATION
ANAEROBIC DIGESTION

Fig. 4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GWP 500 y
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TRANSESTERIFICATION
OIL EXTRACTION
HARVESTING
ALGAE CULTIVATION
ANAEROBIC DIGESTION

Fig. 5

Supporting information for "*Application of LCA approach to Energy and Greenhouse Gas Emission impact of biodiesel production from microalgae cultivated in PBRs: a case study in Denmark*" submitted by Monari et al. (2013)

1. Detailed description of LCI data

The following detailed tables describe which flows are used and their correspondent processes in Gabi and which database has been used. The processes considered are cultivation (Table 1.1), harvesting and drying (Table 1.2), algal oil extraction (Table 1.3), transesterification (Table 1.4), anaerobic digestion (Table 1.5) and glycerol use (Table 1.6).

FLOWS USED FOR CULTIVATIO		
Flows	**Process in Gabi**	**Database**
Carbon dioxide (CO_2)	RER: carbon dioxide liquid at plant	Ecoinvent
Water	RER: tap water at user	Ecoinvent
Total electricity consumption in cultivation	DK: electricity production mix	Ecoinvent
LDPE sheet	RER: polyethylene LDPE, granulate at plant	Ecoinvent
Steel	RER: reinforcing steel at plant	Ecoinvent
Ammonium nitrate	RER: ammonium nitrate, as N, at regional storehouse	Ecoinvent
Mono calcium phosphate	RER: single superphosphate, as P_2O_5, at regional storehouse	Ecoinvent
WASTEWATER CULTIVATION		
Water	Water (wastewater, untreated) [Production residues in life cycle]	Ecoinvent
Nitrogen	Nitrogen (N-compounds) [Inorganic emissions to air]	Ecoinvent
Phosphorus	Phosphorus [Inorganic emissions to air]	Ecoinvent

Table 1.1: cultivation phase

HARVESTING		
Flows used for harvesting	**Process in Gabi**	**Database**
Electricity consumption in flocculation	DK: Electricity production mix	Ecoinvent
Aluminium sulphate	RER: aluminium sulphate powder at plant	Ecoinvent
Lime	CH: lime hydrated packed at plant	Ecoinvent
CENTRIFUGATION		
Electricity consumption in centrifugation	DK: Electricity production mix	Ecoinvent
DRYING		
Heat	RER: heat, unspecific at chemical plant	Ecoinvent

Table 1.2: harvesting and drying phases

EXTRACTION WITH HEXANE		
Flows for algal oil extraction	**Process in Gabi**	**Database**
Electricity consumption in hexane extraction	DK: electricity production mix	Ecoinvent
Heat	RER: heat unspecific at plant	Ecoinvent
Hexane	RER: hexane at plant	Ecoinvent
SCO_2 EXTRACTION		
CO_2 liquid	RER: carbon dioxide liquid at plant	Ecoinvent
Electricity	DK: electricity production mix	Ecoinvent

Table 1.3: algal oil extraction phase

TRANSESTERIFICATION		
Flow	**Process in Gabi**	**Database**
Electricity consumption	DK: Electricity production mix	Ecoinvent
Heat	RER: Heat unspecific at plant	Ecoinvent
Methanol	GLO: methanol at plant	Ecoinvent

Table 1.4: transesterification phase

ANAEROBIC DIGESTION		
PRODUCTION OF BIOGAS		
Flow	**Process in Gabi**	**Database**
Electricity	CH:electricity, low voltage, at grid	Ecoinvent
Plant for Anaerobic digestion	CH: anaerobic digestion plant, biowaste	Ecoinvent
Transport	CH: transport, lorry 20-28t, fleet average	Ecoinvent
Transport for municipal waste	CH: transport, municipal waste collection, lorry 21t	Ecoinvent
Heat	RER: heat, natural gas, at boiler condensing modulating >100kW	Ecoinvent
Municipal solid waste	CH: disposal, municipal solid waste, 0 % water, to municipal incineration [municipal incineration]	Ecoinvent
Biogas from biowaste	CH: biogas, from biowaste, at storage [fuels]	Ecoinvent
ELECTRICITY FROM BIOGAS		
Lubricating oil	RER: lubricating oil, at plant	Ecoinvent
Cogen unit for electricity	RER: cogen unit 160kWe, components for electricity only	Ecoinvent
Disposal of oil	CH: disposal, used mineral oil, 10% water, to hazardous waste incineration	Ecoinvent
Cogen unit for electricity and heat	RER: cogen unit 160kWe, common components for heat+electricity	Ecoinvent
Biogas	CH: biogas, production mix, at storage [fuels]	Ecoinvent

Table 1.5: anaerobic digestion

USE OF GLYCERINE TO PRODUCE PROPYLENE GLYCOL		
Flow	**Process in Gabi**	**Database**
Electricity use	UCTE: electricity, medium voltage, production UCTE, at grid [production mix]	Ecoinvent
Heat	RER: heat, natural gas, at industrial furnace >100kW	Ecoinvent
Transport in street	RER: transport, lorry >16t, fleet average [Street]	Ecoinvent
Transport in railway	RER: transport, freight, rail [Railway]	Ecoinvent
Chemical plant	RER: chemical plant, organics	Ecoinvent

Table 1.6: glycerol use phase

2. LCIA: the relative contributions of each unit process in cultivation phase

In this section, it is possible to observe the different processes used for cultivation and their relative weights to GWP and non renewable energy consumption for each case: freshwater cultivation and "pure CO_2", wastewater cultivation and "pure CO_2", freshwater cultivation and waste CO_2, wastewater cultivation and waste CO_2.

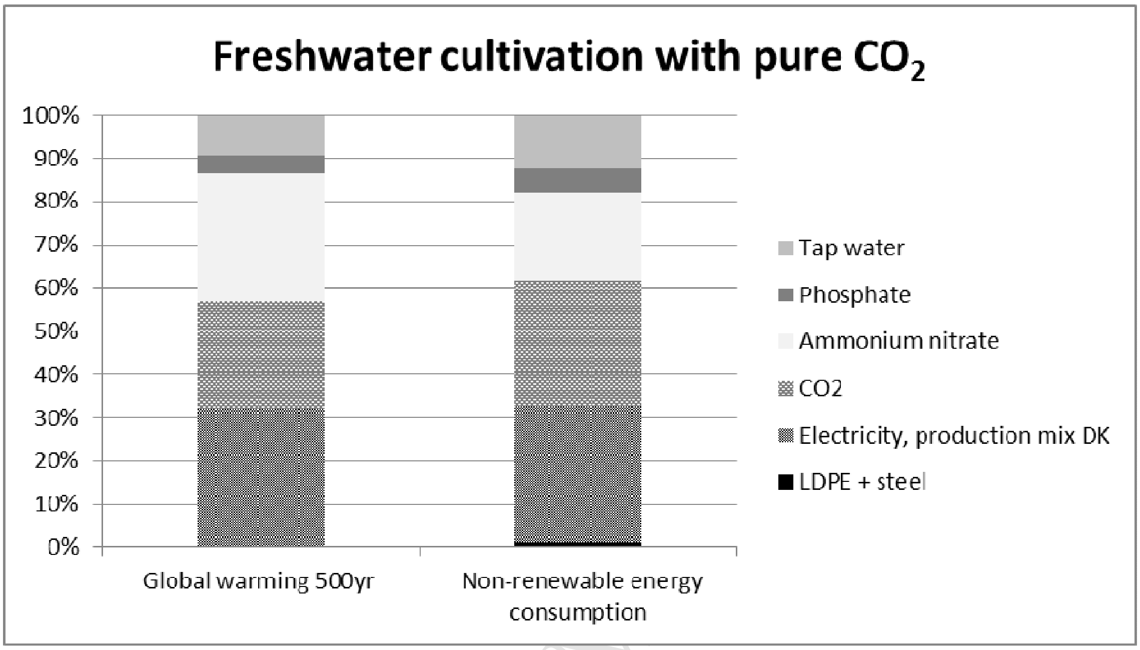

Figure 2.1: contribution of each process unit in freshwater cultivation when "pure" CO_2 is used. In this case the unit processes considered are: tap water in which phosphate, ammonium nitrate and CO_2 are added, electricity for mixing and pumping CO_2 and LDPE for PBR construction

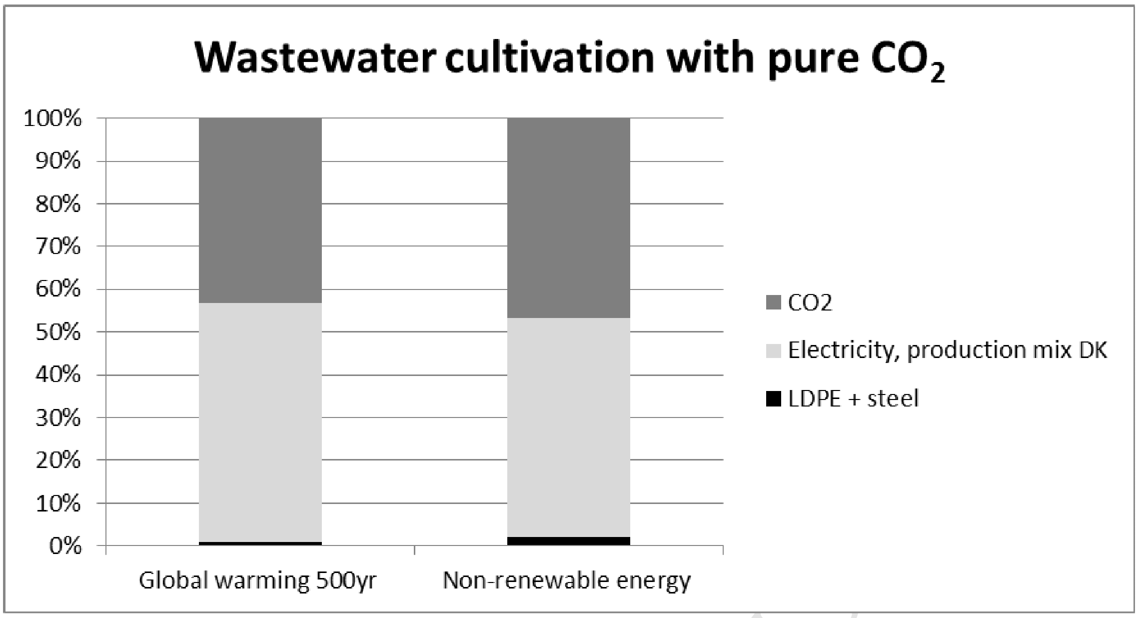

Figure 2.2: contribution of each process unit in wastewater cultivation when "pure" CO_2 is used. In this case the unit processes considered are: wastewater (already enriched by phosphorus and nitrogen) in which CO_2 is added, electricity for mixing and pumping CO_2 and LDPE for PBR construction. In this case, the nutrients are not added to the water

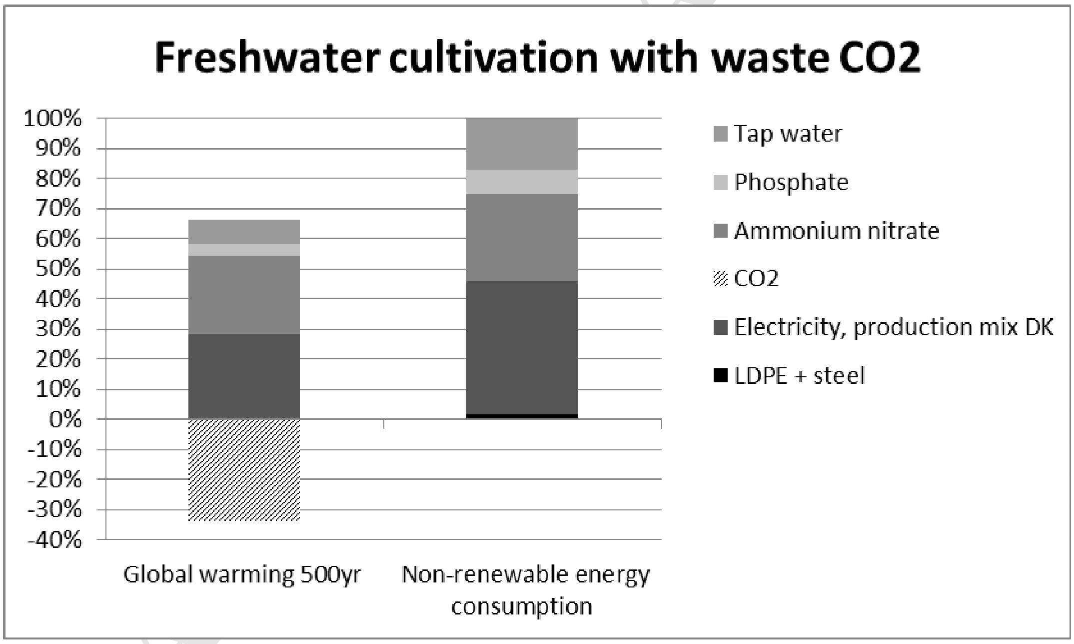

Figure 2.3: contribution of each process unit in freshwater cultivation when waste CO_2 from a nearby cement industry is used for algal flow. In this case the unit processes considered are: tap water in which phosphate, ammonium nitrate and CO_2 are added, electricity for mixing and pumping CO_2 and LDPE for PBR construction. Since CO_2 is a waste flow, the negative contribution of CO_2 indicates that the flow does not take into account its production process

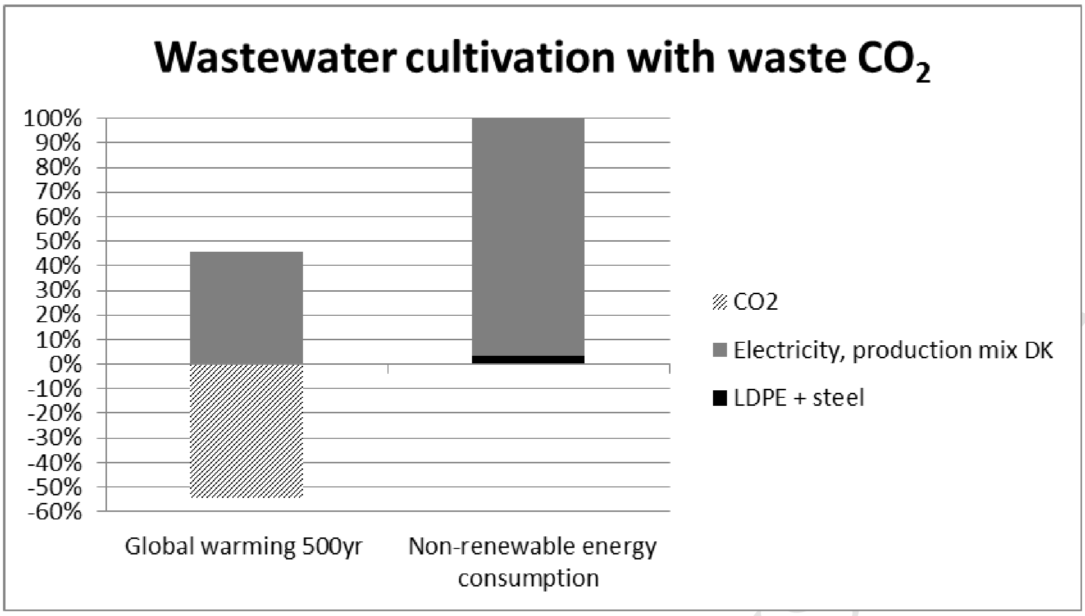

Figure 2.4: contribution of each process unit in wastewater cultivation when waste CO_2 from a nearby cement industry is considered. In this case the unit processes considered are: wastewater (already enriched by phosphorus and nitrogen) in which CO_2 is added, electricity for mixing and pumping CO_2 and LDPE for PBR construction. In this case, the nutrients are not added to the water and since CO_2 is a waste flow, the negative contribution of CO_2 indicates that the flow does not take into account its production process