Research Article Composition of Algal Oil and Its Potential as Biofuel

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First test flights using blends with algae oil are already carried out and expectations by the aviation and other industries are high. On the other hand technical data about performance of cultivation systems, downstream processing, and suitability of algae oil as fuel are still limited. The existing microalgae growing industry mainly produces for the food and feed market. Energy efficiency is so far out of scope but needs to be taken into account if the product changes to biofuel. Energy and CO_2 balances are used to estimate the potential of algae oil to fulfil the EU sustainability criteria for biofuels. The analysis is supported by lab tests as well as data gained by a pilot scale demonstrator combined with published data for well-known established processes. The algae oil composition is indicator of suitability as fuel as well as for economic viability. Approaches attaining high value fractions are therefore of great importance and will be discussed in order to determine the most intended market.

1. Introduction

The energy demand is growing worldwide. The total energy consumption has increased from 196 EJ (10^{18} Joule) in 1973 to more than 350 EJ in 2009 and the tendency is rising [1]. About 80% of this energy demand is delivered from fossil fuels with the consequence of an increase of greenhouse gas emissions in the atmosphere that provokes serious climate changes by global warming. Furthermore, the fossil fuels supplies are constantly diminishing. In consequence, the development of CO₂-neutral fuels is one of the most urgent challenges facing our society and essential in order to meet the planned internationally specified targets, like the reduction of CO₂ emissions in the range of 10–20% by 2020 (e.g., European Union). Therefore, there is an acute demand for sustainable, CO₂-neutral resources to replace the demand of liquid fuels in the near future.

The potential of microalgae as renewable source for biofuel production is very promising due to higher growth rates and the capability to accumulate higher amounts of lipids (from 20% until 80% of dry weight) [2] than conventional oil crops (not more than 5% of dry weight) [3] and therefore the oil yield per hectare obtained from microalgae can greatly exceed the yield from oil plants like rapeseed, palm, or sunflower. Another advantage of microalgae over plants is their metabolic flexibility. That means that a variation in the biochemical composition of the biomass (towards higher lipid, carbohydrates or protein accumulation) can be regulated by varying the cultivation conditions [4]. Photobioreactors can be located on nonarable land and microalgae can grow in seawater or brackish water. Therefore, there is no competition for resources with classical agriculture. Furthermore, in large-scale applications production during the whole year will be possible employing effective process engineering tools for inoculation, maintenance, harvesting, and so forth, much more than possible in agriculture.

In general the water demand for the cultivation of microalgae is low in comparison with crops, especially when closed systems (photobioreactors) are applied. The production of 1 liter of biofuel from oil crops requires around 3,000 liters of water [5]. To obtain 1 liter of biofuel from microalgae with 50% lipid content, 10 to 20 liters are needed, taking into account the stoichiometric demand to fix 1 mol CO_2 from 1 mol of water during the photosynthesis and that the cells themselves consist of up to 85% of water [6]. Although the water demand is in practice much higher because reactor cooling is necessary in closed photobioreactors or for compensation of evaporation in open systems, the values are one to two orders of magnitude lower than for conventional agriculture [7]. These low values reconfirm the high potential and motivation for using microalgae as sustainable feedstock for biofuels.

The drawbacks of the current state of microalgal biotechnology are the high investments costs and the high demand on auxiliary energy for biomass production and for lipid processing to biodiesel leading to high costs for biomass and biodiesel. The use of residual nutrient sources and nutrient recycling are one of the keys for a sustainable production of biodiesel from microalgae. Wastewater can be used to supply nitrogen and phosphorus, main nutrients needed for the cultivation of algal biomass. The use of residual algal biomass after lipid extraction for example as feed (because of the high vitamin content) is a key factor in biorefinery concepts in order to improve economic feasibility. The rest biomass can be also fermented to produce methane or ethanol. Therefore, the ongoing research and development efforts are focused on the improvement of both the economic feasibility and sustainability for the production of biofuels from microalgae. The biorefinery concept of fermentation of the rest biomass to biogas will be included on the energy and CO₂ balances shown in this paper.

2. Energy and CO₂ Balances for CO₂ Capture Growing Microalgae

2.1. Theoretical Potential for Energy Capture. Microalgae are phototrophic microorganisms. That means that they use CO_2 as inorganic carbon source and sunlight as energy source to synthesize organic compounds. In this process called photosynthesis, O_2 is released as a by-product. The overall equation of the oxygenic photosynthesis can be described as

$$\operatorname{CO}_2 + \operatorname{H}_2\operatorname{O} \xrightarrow{\text{sunlight}} [\operatorname{CH}_2\operatorname{O}] + \operatorname{O}_2,$$
 (1)

where $[CH_2O]$ represents the smallest building block of a carbohydrate and is the first organic product of photosynthesis. These sugar molecules are starting points for anabolic reactions to synthesize biomass compounds. Further auxiliary energy is required for these metabolic pathways. The organic carbon content in microalgae under nonlimiting nutrient conditions is about 50% by weight. In this case the stoichiometric CO₂ demand is 1.83 kg (0.5 · 44 g/mol CO₂ 12 g/mol C) per each kilogram of dry algal biomass produced. Under accumulation of storage components like carbohydrates or lipids with different degrees of reduction the CO₂ fixation varies between 1.65 kg·kg⁻¹ (0.45 · 44 g/mol CO₂ /12 g/mol C) for high starch content (50%) and ca. 2.33 kg·kg⁻¹ (0.634 · 44 g/mol CO₂ /12 g/mol C) for 50% lipid content respectively.

The conversion efficiency of light into biomass is expressed by the photoconversion efficiency (PCE) and is defined as the energy gained by a conversion process compared

TABLE 1: Microalgae biomass yield per square meter of surface area as function of PCE for three different solar irradiations assuming a lower heating value of $30 \text{ MJ} \cdot \text{kg}^{-1}$ dry matter.

	Germany	Mediterranean area	Dessert	
PCE\Irradiation	3,500	7,000	9,000	$MJ \cdot m^{-2} \cdot a^{-1}$
1.0%	1.2	2.3	3.0	$kg_{DM}\!\cdot\!m^{-2}\!\cdot\!a^{-1}$
2.0%	2.3	4.7	6.0	$kg_{DM}\!\cdot\!m^{-2}\!\cdot\!a^{-1}$
3.0%	3.5	7.0	9.0	$kg_{DM}\!\cdot\!m^{-2}\!\cdot\!a^{-1}$
4.0%	4.7	9.3	12.0	$kg_{DM}\!\cdot\!m^{-2}\!\cdot\!a^{-1}$
5.0%	5.8	11.7	15.0	$kg_{DM}\!\cdot\!m^{-2}\!\cdot\!a^{-1}$

to the available sunlight supplied to the conversion process. In case of microalgae it is the ratio of the lower heating value of dry algae biomass divided by the sunlight supplied per ground area to the algae cultivation:

$$PCE = \frac{\text{biomass}[W/m^2]}{\text{sunlight}[W/m^2]}.$$
 (2)

Establishing maximal PCE under sunlight is of great interest because it implies attaining maximal biomass yield achievable from a given area of crop plantation or algal culture [4].

The theoretical biomass outputs as function of PCE for three different solar irradiations are given in Table 1.

The listed range In Table 1 from 1% to 5% corresponds to the actually achieved PCEs on different cultivation systems and will be explained more detailed later on. The next paragraphs describe the maximal achievable PCE according to biological characteristics of microalgae.

The entire energy from incident sunlight cannot be converted into biomass because of several physiological properties from both microalgae and higher plants that reduce the efficiency of photosynthesis. Figure 1 shows the energy losses between the incident solar radiation and the formation of a carbohydrate. Because of the pigment absorption properties, only the photosynthetic active radiation (PAR) part from the whole solar radiation spectrum can be used for photosynthesis. The PAR consists of photons in the wavelength range between 400 nm and 700 nm resulting in a 55% loss of the total incident radiation. The aim of light absorption by the pigments is to excite the chlorophyll molecules to eject electrons so that the radiation energy can be converted into chemically stored energy in form of ATP (adenosine tri phosphate) and NADPH₂ (nicotinamide adenine dinucleotide phosphate). ATP and NADPH₂ are needed to fix CO₂ and to synthesize further biomass. Only photons with wavelengths of 680 nm and 700 nm (red) exhibit the exact energy content required for the excitation of chlorophyll. Higher frequency photons (blue) exhibit higher energy content as required and are used with less efficiency as red photons. Excess energy is dissipated as heat or fluorescence, a mechanism known as nonphotochemical quenching. Consequently, there is an energy loss of 21% of the absorbed PAR energy due to the degradation of PAR to the chlorophyll excitation energy at 700 nm. Furthermore about 35% energy losses are generated

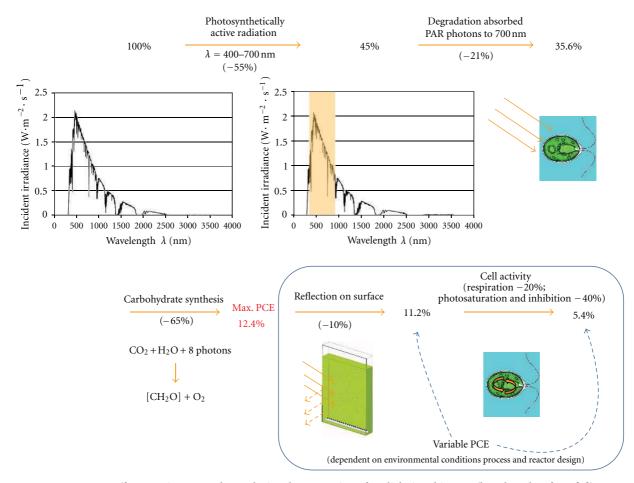


FIGURE 1: Effects causing energy losses during the conversion of sunlight into biomass (based on data from [4]).

by the several enzymatic steps needed for the fixation of CO_2 into carbohydrate. In this case a photon demand of 8 photons to fix one mole of CO_2 to carbohydrates is required [4].

The addition of all the energy losses described above, gives a theoretical maximum PCE of 12.4% (see Figure 1), a value often referred to as photosynthetic efficiency (PE). The resulting achievable value of PE is independent of the photobioreactor technology or process strategy and cannot be improved by genetically engineering of the cells. Lipid production leads to an increase of the heating value of the biomass from $20 \text{ MJ} \cdot \text{kg}^{-1}$ for oil-poor algae (between 20% and 30% dry weight) to $30 \text{ MJ} \cdot \text{kg}^{-1}$ for oil-rich algae (50% dry weight) [8]. For a constant PCE that leads to lower measured areal productivity in terms of dry mass. Furthermore, the photon demand will increase if proteins and lipids are synthesized reducing the PCE due to ineffective metabolic steps.

The real PCE in outdoor algal cultures decreases to a value between 1.5% and 5% depending on the photobioreactor system applied (race ways pond or closed systems), mixing conditions, and weather conditions. Reasons for the further losses of radiation are the reflection of light on water bodies at the reactor surface (~10%), photosaturation and photoinhibition (~40%), and respiration (~20%) [4]. If the PCE loss due to photosaturation can be reduced for example by means of light dilution (for further explanation see Subsection 2.3) short time PCE of 8% can be measured [9]. Usual PCE of crop biomass does not exceed 1% PCE in temperate regions [10, 11] but can reach values of almost 4% during short-term experiments under optimized conditions [12]. Those maximum yields are peak values and hardly observed on average.

The PCE values mentioned above will be subject to further decrease during lipid production. If microalgae are cultivated under nitrogen limitation, they cannot synthesize the required protein and nucleic acid molecules for further growth and switch their metabolism towards the accumulation of energy and carbon reserve like starch or lipids depending on the algal strain. Lipid accumulation means that the photon demand to fix CO₂ increases in comparison with starch accumulation because the synthesis of lipids from the first organic product of photosynthesis (CH₂O) requires additional enzymatic steps.

No quantitative measurements or assessments of PCE values for growth during lipid synthesis can be found in the literature for the moment. On this area experimental works in labor scale are actually running at the Department of Bioprocess Engineering KIT as a part of a collaboration project with EnBW AG to obtain PCE values of the optimized process for lipid accumulation [13].

2.2. Biodiesel from Microalgae. Biodiesel is a mixture of fatty acids of diverse lengths esterified with an alcohol, typically methanol. Typical crop plants for biodiesel production are rapeseed, palm, sunflower, and soybean due to their high lipid content. Recent efforts to substitute microalgae biomass for these conventional crops are driven by the higher growth rates and lipid content of several algae species (Table 2). Biodiesel from microalgal feedstock might even be especially interesting for general aviation due to its good flow properties at low temperatures and high energy density.

The European norm EN 14214 gives an overview of biodiesel quality requirements and test methods. Table 3 shows a comparison of properties of microalgal oil, petroleum diesel, and norm standards.

The suitability of microalgal biomass as biofuel feedstock is closely related to the length and degree of saturation of its fatty acids as specified by the four key figures iodine value, oxidation stability, cetane number, and the cold filter plugging point. Beside other quality standards the feedstock needs to comply with target ranges defined by the European norm, which is fulfilled in case of algal biodiesel as shown in Table 3.

A high iodine number represents a high degree of unsaturation of fatty acids in biodiesel, which is unfavourable because fatty acids with higher content of double bonds are prone to oxidative damage. Otherwise, unsaturated fatty acids are beneficial for flow properties, especially at lower temperatures and therewith result in an advantageous cold filter plugging point. Lipids rich in long chain fatty acids with a low degree of saturation exhibit a high cetane number, indicating a short ignition delay time and high combustion quality whereas an exceeding degree of saturation might collide with the request for a reasonable cold filter plugging point by precipitation at low temperatures [15].

A comparison of fatty acid profiles of some selected algae strains and conventional biodiesel feedstock (Table 4) shows that there are fatty acids of comparable chain length extractable from all feedstocks whereas algae oils can contain considerably higher amount of polyunsaturated fatty acids. The resulting lower cold filter plugging point makes biodiesel from microalgae suitable as fuel for aviation. The fatty acid profile of microalgae and crops is not constant and varies depending on the culture conditions and the growth stage of the culture when harvested. Optimal culture conditions for the desired fatty acid profile can be easier regulated during the microalgal growth in photobioreactors than by oil crops on the landfill but at the cost of higher energy and CO₂ demand. These conditions are species specific. No comparison data about the improve of quality for biodiesel in counterbalance with the higher energy demand for the regulation of optimal conditions are described at the moment in the literature and will not be considered for the energy and CO₂ balances.

Strain selection can be performed with a special focus not only on generally applied selection criteria, such as lipid content and areal lipid productivity, but also on the fatty acid profiles matching biodiesel requirements. Further selection criteria for microalgae need to be considered as they mainly influence the ability to produce biomass at a large scale in the

TABLE 2: Lipid content of selected algae species [2].

Microalgae species	Lipid content (% dry weight)
Botryococcus braunii	25-75%
Nannochloropsis sp.	31-68%
Schizochytrium sp.	50-77%
Neochloris oleoabundans	35-54%
Nitzschia sp.	45-47%

respective environments. Amongst them strain-specific optimal temperature range, salinity of the cultivation medium, and the ability to be maintained for longer periods in nonaxenic cultures need to be taken into account. Therefore, the choice of the most adequate cultivation system and process strategy will be decisive in order to achieve high areal lipid productivities. An overview and comparison of different types of reactors used nowadays for microalgae cultivation is given in the Subsection.

2.3. Microalgae Cultivation. Traditionally, microalgae are cultivated in the so-called open ponds, which are natural or artificial bodies of standing water typically 20 to 30 cm deep. The CO_2 is taken from the atmosphere. The expected upper performance for open ponds is a PCE around 0.5%, so basically not better than for terrestrial energy plants. The first means to increase productivity is better mixing, which can be achieved by employing paddle wheels to move the water through annular channels, thus improving gas transfer and bringing all cells to the bright surface at least statistically. In more recent applications an additional supply of CO₂ is foreseen, leading to expected PCEs below 2%. The current activities to produce biodiesel from microalgae are actually based on these raceway ponds because of their low costs and low demands on auxiliary energy when normalized to the pond volume. However, such open facilities exhibit high water evaporation per ground area (1 to $3 \text{ m}^3 \cdot \text{m}^{-2}$ and year depending on the region) and a high risk of contamination. High evaporation allows operation without undermining the idea of using arid areas only by employing sea water algae and building the plant close to the sea or where brackish water is available [19]. Microalgal dry biomass concentrations do not usually exceed $2 g \cdot L^{-1}$. This has a direct impact on the subsequent solid/liquid separation steps and on the demand of auxiliary energy for harvesting, which is high, normalized to the biomass produced.

To avoid these disadvantages the attention of research and application has shifted to closed photobioreactors. Here the culture volume is separated from the environment by transparent walls leading to less evaporation, less contact interface for possible invasion by competing microalgae, contaminating bacteria, or multicellular grassers or predators. Furthermore, CO_2 can be applied very efficiently with only small losses in the off-gas and can be controlled close to the 1 to 2 mbar, which have been shown to be optimal for growth. Other values like pH or temperature could in principle be controlled as well, but current applications do not make much use of these options. However, there is a deeper

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TABLE 3: Comparison of selected properties of algal biooil and typical conventional diesel with respect to the European norm for biodiesel
[14].

Fuel property	Algal biodiesel	Petroleum diesel	EN14214 standard
Higher heating value (MJ/kg)	41	45.9	_
Kinematic viscosity (mm ² /s) 40°C	5.2	1.2–3.5	3.5-5.2
Density (kg/L)	0.864	0.83-0.84	0.86-0.90
Carbon (wt%)	76	87	_
Hydrogen (wt%)	≤12,7	13	_
Oxygen (wt%)	≥11,3	0	_
Sulphur (wt%)	0	0.05 max	<10 max. 0,02
Boiling point (°C)		180–340	_
Flash point (°C)	115	60-80	>101
Cloud point (°C)		-15 to 5	_
Pour point (°C)	-12	-35 to -15	_
Cetane number	_	51	>51

TABLE 4: Fatty acid profiles of biodiesel feedstock (wt.%) resulting from gas chromatographic analysis.

Fatty acid	Double bound position ²	Rapeseed [15]	Sunflower [15]	Nannochloropsis salina [16]	Phaeodactylum tricornutum [17]	Botryococcus braunii [18]
C12:0 ¹	_			5.0		0.7
C14:0	—		—		4.5	0.8
C15:0	—		—	0.5	—	0.5
C16:0	—	4.9	6.2	37.5	25.8	21.0
C16:1	9		0.1	23.3	37.5	2.0
C16:2	7, 10		_	_	—	6.5
C16:3	7, 10, 13		—		—	15.2
C17:0	—		—	0.4	—	0.1
C18:0	—	1.6	3.7	0.9	1.3	2.9
C18:1	9	33.0	25.2	11.9	—	3.2
C18:2	9, 12	20.4	63.1	1.5	5.1	13.6
C18:3	9, 12, 15	7.9	0.2	_	2.0	33.0
C20:0	—		0.3	0.1	—	0.2
C20:1	11	9.3	0.2	_	—	
C22:0	—		0.7		—	0.1
C20:4	5, 8, 11, 14		—	3.3	1.6	
C20:5	5, 8, 11, 14, 17		—	15.3	13.1	
C22:0	—		—	0.4	—	
C22:1	13	23.0	0.1	_	—	
C24:0	_	_	0.2	_	_	0.2
C24:1	15	_	_	_	_	

¹ Fatty acid with chain length of 12 carbon atoms and 0 double bonds. The same nomenclature applies for all other fatty acids.

²Double bound position beginning from the carboxyl group.

reason why closed reactors achieve higher biomass concentrations and higher productivities than ponds. This can be understood by employing growth kinetics of the algae for designing photobioreactors. A typically growth kinetics depending on light irradiation plotted as photosynthetically active photon flux density (PFD) is shown in Figure 2. The basic point is that a linear increase with increasing light intensity is only achieved for low light conditions. Direct incident sunlight, as applied on open ponds, cannot be used by the algal cells or is even inhibiting.

In this situation closed reactors offer the option to "dilute" the light over a larger surface. In this way the algae cells are exposed only to a small fraction of the full sun light intensity and grow close to the point of their optimal efficiency, so that PCE of 5% and under well-controlled conditions of up to 8% can be achieved [9]. However, such values have never been obtained for real hectares and real years. For quite some years large tubular reactor installations of more than one ha are in operation [20, 21], where the light dilution is obtained by glass tubes forming fences. Here the light dilution factor is given by the ratio of the illuminated glass surface and the foot print area. The medium is pumped through the pipes in turbulent flow. Fresh CO_2 can be applied either pure or mixed with air. Such tubular reactors offer good productivities for

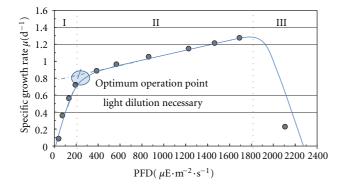


FIGURE 2: Specific growth rate as function of light irradiation plotted as photosynthetically active photon flux density (PFD) for *Chlamydomonas reinhardtii*; 1000 μ E·m⁻²·s⁻¹ corresponds approximately to 200 W·m⁻². The PCE can be read as gradient from the origin to a given point on the curve. Therefore, PCE is at its optimum only in range I, where the circle indicates the point of highest growth rate.

high value products, but show other disadvantages. The costs for installation can go up to over several hundred \in per m², being too high to ensure economic feasibility considering commodity markets. Furthermore, pumping energy exceeds several hundred or even thousand W·m⁻³, what prohibits energy positive microalgae production.

The second development line for closed photobioreactors is given by the so-called flat plate reactors. These designs consist basically of two parallel oriented vertical plates including the water body. Mixing and mass transfer are achieved by bubbles entering the single plates from the bottom similarly to bubble column or airlift reactors. Their solar light capturing surfaces consist of transparent materials with long shelf lives, for example, polyethylene (PE), polyvinylchloride (PVC, with some ecologic considerations) or poly(methyl methacrylate) (PMMA). In a simple case the plates are formed by plastic bags on scaffolds of mash wire, see Figure 3. The light path length, defined as the thickness of the plate reactor, is in this case close to 10 cm, but can vary in more recent designs between 1 and 5 cm. Low cost and a comparatively small energy input of less than $100 \,\mathrm{W} \cdot \mathrm{m}^{-3}$ make this reactor design in principle feasible for biofuel production. But still there is potential to achieve even better systems. The energy demand for bubbling can be reduced by choosing a low plate design, therewith reducing hydrodynamic pressure at the bottom of the reactor. To keep light dilution at a high level, the plates have to be arranged closer to each other. Furthermore, the smaller the total culture volume per footprint area (water coverage) is, the less energy is wasted for mixing. It is necessary to understand that the reaction volume is the intracellular space and not the chemically inactive medium between the cells. Consequently, a short light path length leads to low water coverage and high dry biomass concentrations. This demand for low height designs and low water coverage also supports a reduction of the material expenditure. Concluding, the energy input is one important cost factor and likewise significantly influences the net energy gain.

TABLE 5: Power input (excluding light) and algae biomass productivity in dry matter per cultivation volume and day of different reactors in outdoor experiments.

Reactor type	Power input $[W \cdot m^{-3}]$	Productivity $(g \cdot L^{-1} \cdot d^{-1})$	Reference
Helical tube reactor (outdoor)	3,200	1.4	[22]
Bubble column (outdoor)	109	0.4 (10 days average)	[23]
Flat panel airlift	100-200	circa 0.76 (average)	[24]

Table 5 gives an overview of power input of several reactors in outdoor experiments.

But there are other issues. Besides lower energy input especially the temperature problem has to be addressed. During outdoor operation the medium heats up during daytime depending on the changing irradiation and the air temperature. Microalgae, at least the strains usually cultivated nowadays, exhibit a quite narrow temperature growth profile. So cooling, for example, by spraying and evaporating water on the surface is necessary even in Middle-Europe in summer. One solution is to embed the plates in a separate water body, which at least dampen the temperature oscillations. The company Solix Biofuels [34] for example, cultivates algae in submerged flat plastic bags. The fundamental setup of the third-generation reactor concept (3G) is depicted in Figure 3. Major advantage of the submerged reaction compartments is the fact that additional temperature control is not necessary in the system because the surrounding water acts as temperature buffer. Moreover, construction costs for the reactor are reduced as there is no need for a special scaffold supporting the flat reaction compartments. According to the information given by the official website Solix produces 5,000 to 8,000 gallons of algal oil per acre, per year (circa 42 to 67 t ha⁻¹ \cdot a⁻¹, assuming an oil density of 900 kg \cdot m⁻³) [35]. A fourth generation reactor including membrane aeration is currently under development.

With regard to economic considerations investment costs should not exceed 20 ${ { \in \cdot m^{-2} }}$ for biofuel production and operating costs need to be minimized. A similar approach has been developed by Proviron [36] (Figure 4) and represents another applied advancement of classic flat plate reactors. Their major focus was set on development of an efficient low cost reactor suitable for large-scale outdoor applications. Their approach comprises the incorporation of flat growth compartments (less than 1 cm thick) within water-filled plastic bags without any rigid structure. The major part of the setup consists of water-filled chambers that are separated from the reaction compartments. Water diffuses the impinging solar radiation, which should result in an equalized light distribution within the water-filled chamber. At the same time, temperature is regulated without any additional energy input. Moreover, the water-filled chambers themselves constitute the scaffold of the reactor, analogous to Solix reactors. In the future the low auxiliary energy demand of 20 kW \cdot ha⁻¹ should be further reduced with control strategies that aim at an adaption of aeration to light. According to the company

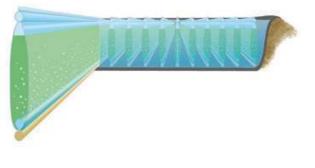




FIGURE 3: Solix Biofuels demonstration facility: schematic representation of the 3G reactor setup and photography of the production facility [34].



FIGURE 4: Submerged flat panels in a *Proviron* photobioreactor [36].

outlook investment costs are expected to drop from currently $200,000 \in ha^{-1}$ to $100,000 \in ha^{-1}$.

Besides sunlight and CO_2 all systems need to be supplied with water and nutrition.

 CO_2 has in principle to be supplied in order to meet the stoichiometric demand of the cells and to keep the partial pressure in the medium at a level high enough with respect to the uptake kinetics to allow sufficient flow across the cell membrane. CO_2 can be provided from industrial flue gases, for example, from biogas or gas combustion, while flue gas from hard coal combustion is less favourable because of heavy metal dust particles. Other CO_2 sources can be found in chemical processes like lime kiln or ethylene factories.

In tubular reactors pure CO_2 can be applied because mixing is done by pumps. In flat plate reactors, additional air is supplied for mixing purposes leading to a gas mixture of usually less than 10% CO_2 molar fraction. As no cheap and reliable CO_2 sensors are available yet, pH is taken as controlled value. As the pH is also influenced by other factors (e.g., ammonia uptake) this is not always an optimum solution. CO_2 feeding on demand, for example, with strongly reduced rates during the night is one option out of the tools of control engineering. Following the fluorescence signal given by the cells to optimize agitation and harvesting cycles on line is another idea under discussion. The cultivation process has to be controlled in a way that the nitrogen source (ammonia or nitrate) is depleted at the end of a first process phase, while the cells start to accumulate lipids. The exact timing of this switching process is another issue to be addressed during development of optimal production processes.

Future production processes will further integrate biology, process and control engineering, and environmental conditions. Further screening and genetic engineering is targeted to a fatty acid profile adapted to the technical needs. The above-mentioned light saturation can be diminished by the so-called antenna reduced mutants. While wild-type cells dissipate energy under high light conditions, these cells can take up only as many photons as they can process further in the metabolism. This makes higher cell densities and higher efficiencies in closed reactors possible. Other attempts to further enhance production are more extensive approaches to combine open ponds with light dilution by applying light conducting glass cones or other structures on the surface of creaks.

The span of the values for productivity and energy demand shown in Table 6 are usually based on pilot plant facilities. Linear extrapolation between productivity and energy demand is not possible. Firstly, productivities are influences by operational parameters themselves. High mixing energy supports high growth rates by better mass transfer, but also by the so-called intermittent light effect [37]. This phenomenon describes the abilities of the cells to store light energy in the time range of ms, in this way making profit from fast mixing cycles between dark and light parts of the reactors. While a given reactor geometry with respect to light dilution may be optimal in a given region, it may be suboptimal in another, where higher light dilution is necessary on cost of higher energy and material demand. Secondly, a measured PCE cannot be simply applied to another production site. Even if light dilution is considered properly, the effect of environmental factors on the PCE value like temperature—usually assumed as being from Arrhenius type [38]-diffuse light, and day length can hardly be anticipated. Growth-model-based attempts for extrapolation using local weather data or landscape characteristics have nevertheless been proposed [39, 40] but not finally proven. Practical experiences are actually missing, whether the advantage of a higher irradiation of a maximum ratio of 2.5 between middle Europe and North Africa can really be exploited facing problems of temperature control, CO₂ and nutrition logistics and higher light dilution factors. So there are already ideas to cultivate microalgae in moderate northern areas making use of long days with many sunny hours in the summer and switching off the plant in winter, a strategy that saves at least maintenance and operational energy.

2.4. Downstream Processing. The first step in product recovery is the harvesting of the biomass that comprises separation and dewatering of the algae cells. Dewatering in this context means removal of the intercellular medium to an extent of about 90% resulting in a pasty pumpable product, while the intracellular water (>80%) keeps unchanged. Harvesting can be done in principle by filtration or centrifugation [41]. The task of solid/liquid separation in bioprocesses is not new as such including low value products like yeasts or bacteria. Therefore, applicable devices are available on the market. However, microalgal suspensions exhibit for the moment low solid concentration of less than, for example, $1 g \cdot L^{-1}$ and a low added value with respect to biofuels. This fact leads to changes in operation of classical approaches towards lower energy consumption and also triggered the design of new separation devices (e.g., Evodos [42]).

TABLE 6: Cultivation-system-related parameters (own estimates based on/completed with [9, 25, 26]).

	Raceway ponds	Closed systems	
Photoconversion efficiency	1.5	5	%
Typical biomass concentration	0.5 to 1	2 to 9	kg/m ³
Spec. energy demand cultivation (per area)	0.5 to 4	2.5 to 15*	W/m ²

* Based on the volumetric demand of 50 to 150 W/m^3 for closed systems and a volume/surface ratio from 0.05 to $0.1 \text{ m}^3/\text{m}^2$ [23, 27].

Filtration is the classical method in bioseparation. Harvesting in open ponds is sometimes still done using manual sieves [43]. Press filtration is the standard operation procedure in sludge dewatering [44]. Filter aid drum filtration is applied to produce compressed yeast from yeast "milk" [44]. Basic problems are the compressible filter cakes with low specific permeability and the high maintenance demand. The process development to overcome these problems goes in two different directions. One is the application of shear forces tangential to the filtration membrane with the effect of shear forces to prevent the formation of a filter cake [45]. The other one is belt filtration with continuous cake removal and belt drying by pressing [46].

Centrifugation is regarded as the most effective separation method with respect to large scale [20, 47, 48]. As the separation principle depends on cell size and on specific weight, the problems are already charted. The cell diameter is of $5-10\,\mu\text{m}$ in the range of yeasts, which are usually separated by decanter centrifuges or disk stack separators [49]. This latter choice is often made for microalgae as well [20, 48]. While the specific weight for yeast is in the range of 1.1 kg·L⁻¹, the density of algae depends on the oil content and can go down to values lower than the water density, making centrifugation more and more difficult and leading to flotation as a better approach. Disk stack separators and also newly designed centrifugal devices especially for microalgae (e.g., Evodos [42]) are operated at, for example, 3,000-4,000 g to reduce cell damage and claim efficiencies even below 1 kWh/m³.

Prior to the application of separation devices a preconcentration can be employed. Flocculation can be used in order to ease the following dewatering steps. Diverse mineral salts (e.g., aluminium sulphate, ferric chloride) or organic polymers (chitosan) can serve as flocculants. However, addition of chemicals is expensive, can interfere with extraction, or is even regarded as harmful to the environment [29]. Autoflocculation, for example, by stopping aeration with the effect of a pH shift, might avoid these detriments. The success depends on the presence of polysaccharides or specific proteins on the cell surface and is therefore strain and process dependent.

Data for the energy demand of solid/liquid separation is given from the device suppliers in terms of energy per processes liquid volume, but what counts at the end is the energy demand per cell dry mass. In practice this means that the energy demand can vary by almost a factor of 100 comparing different cultivation techniques. Typical values for energy use in solid/liquid separation are given in Table 7.

Moreover, solid/liquid separation developments on the biological level could be helpful, like approaches for cultivation in biofilms attached on solid surfaces or the employment of filamentous algae, which can be harvested much easier than single cells in suspension culture.

The second step in product recovery after the harvesting of the biomass is the extraction of lipids and lipid conditioning to biodiesel. Microalgae contain two main classes of lipids, polar lipids (including phospholipids and glycolipids) and neutral lipids (mono-, di-, triglycerides, isoprenoids, and waxes). The latter are accumulated under stress conditions, like nitrogen limitation, and exhibit most desirable properties for conversion to biodiesel [28, 50]. As described above, high specific lipid contents are attained on the expense of low biomass productivities [51]. These technical challenges must be approached by sophisticated cultivation strategies, such as phased or spatial separation of high biomass productivity and lipid accumulation in two different process phases or bioreactor compartments.

Conventional biodiesel processing of algae biomass requires harvesting of biomass, drying of the crude material, and subsequent hexane extraction of lipids. The latter are then subjected to a transesterification process. During this process the three fatty acids of one molecule triacylglyceride (TAG) consecutively react with alcohol, typically methanol provided in molar excess, to form fatty acid methyl esters (FAMEs) and glycerol. Figure 5 shows the steps for biodiesel processing.

To increase efficiency of the extraction process previous cell disruption can be taken into account in order to enhance solvent access to the storage lipids [50]. The higher content of algal polyunsaturated fatty acids may induce adverse side reactions during transesterification [55]. However, some of these fatty acids, for example, eicosapentaenoic acid (C20:5), attain higher prices on the healthcare, nutrition, or other markets [26, 56]. Therefore, gentle transesterification conditions and an additional separation step of products could improve profitability of the overall process. This is especially true at an early stage of development while scales are not big enough to deteriorate prices but biofuels from microalgae are still more expensive than petroleum diesel.

Biomass with low residual water content is prerequisite for the typical transesterification with a homogenous base catalyst as the remaining water leads to undesirable saponification reactions. Soap formation also occurs in consequence of a high content of free fatty acids found in some algae and is to be prevented because saponification impairs the subsequent separation process and leads to partial consumption of the catalyst [28, 57, 58]. Although base catalysis is favoured due to faster reaction kinetics and requirement of lower reaction temperatures saponification can be avoided when acid catalysts are used instead of alkaline ones, acid or base catalysts necessitate an additional neutralization process step. However, further attempts aim at replacing homogenous with heterogeneous catalysts. The latter reduce complexity of TABLE 7: Electricity demand and ratio of separation for different separation technologies (adapted data from [9, 25, 26, 28]). A detailed review is available from [29].

	Centrifuge	Filtration	
Specific electricity demand	3.6 to 14.4	1.8	MJ/m ³
Ratio of separation	90	80	%

the process, cost, and the use of hazardous chemicals because separation and recovery of the catalyst as well as neutralization are dispensable [57, 59, 60].

Further attempts to introduce more fundamental changes in the overall process in order to replace toxic chemicals, for example, hexane, and to increase overall efficiency include extraction of lipids with supercritical carbon dioxide. In small-scale experiments extraction has proved to be efficient and shows energetic advantages over conventional extraction [50, 61]. Applicability in larger scale requires further assessment and might be energy intensive due to the demand of high pressure. Carbon dioxide can be recycled after extraction or alternatively fed into photobioreactors. One additional advantage of this method is the ease of modification, for example, utilization of cosolvents, to attain certain selectivity in the extraction therewith enabling sequential extraction of different lipid classes. Experiments show that under specific process conditions triacylglycerides can be extracted with pure supercritical carbon dioxide in a first reaction step whereas phospholipids can be recovered only after addition of another solvent, such as ethanol [50].

A promising approach to eliminate the cost and energy intensive drying process of biomass suggests a two-step process that can be applied directly to wet biomass. In a first step lipids are hydrolysed inside the cells at high temperatures (ca. 250°C) resulting in an easily filterable solid that contains the majority of free fatty acids while phosphorous and nitrogen can be recovered from the filtrate. In a second reaction step the retentate was subjected to the esterification with ethanol at supercritical conditions. The overall process therewith does not require the drying process, facilitates the straightforward recovery of nutrients, and eliminates the use of toxic chemicals [62].

2.5. Energy and CO_2 Balances of Algal Biofuels. Calculations on the energy and CO_2 balances for the production of biodiesel from microalgae on the base of the technological stateof-the-art for algal cultivation and lipid extraction will be shown in this section. The energy and CO_2 balances are determined for both cultivation technologies: (i) raceway ponds and (ii) closed photobioreactors considering the process as given in Figure 6. After cultivation the microalgae are harvested. The oil is extracted and converted into biodiesel. The algae press cake is digested for biogas production. The biodiesel as well as the biogas is balanced to substitution of fossil fuel. As microalgae biodiesel production in an industrial scale does not exist, figures from commercial vegetable oil (production capacity of around 100,000 tons per year) conversion to biodiesel are used.

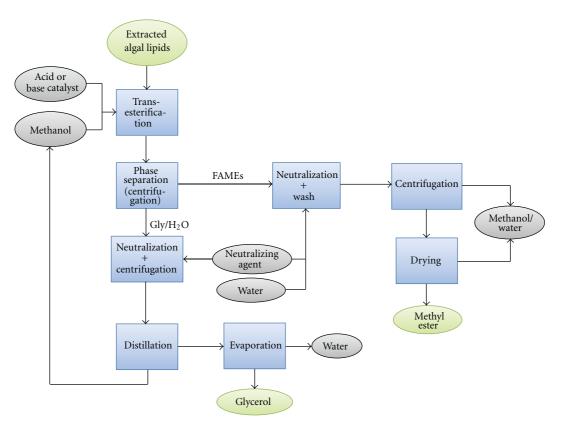


FIGURE 5: Scheme of conventional biodiesel processing [52–54].

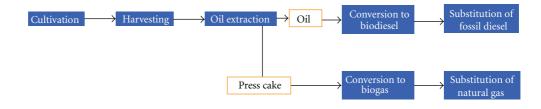


FIGURE 6: Scheme of evaluated value chain.

The results are presented for a solar radiation of 7,000 MJ·m⁻²·a⁻¹ (corresponding to the radiation in Mediterranean area) and PCEs of 1.5% for raceway ponds and 5% for closed photobioreactors, which are good case scenarios. The specific energy demand for cultivation with raceway ponds is set to $1 \text{ W} \cdot \text{m}^{-2}$ surface area and mainly applies for mixing of the culture media. For closed photobioreactors it is set to $100 \text{ W} \cdot \text{m}^{-3}$ (corresponding to $5 \text{ W} \cdot \text{m}^{-2}$). As for raceway ponds the energy demand for closed photobioreactors is predominantly used for mixing and also for gas supply, and therefore in both cases it is exclusively electrical energy. Potential additional energy consumption for, for example, temperature control is neglected.

The technology of choice for harvesting is centrifugation as it is the state of the art today. Since harvesting microalgae with biomass concentrations of just a few g per litre of culture media strongly determines the feasibility of an energetic utilization, other options need to be considered in future. The oil is assumed to be extracted with a yield and energy demand comparable to that of, for example, rape seed oil production. The energy demand for transesterification of the oil is set to 0.72 MJ electric and 1.7 MJ thermal per kg of oil in analogy to rapeseed oil [30]. Furthermore 0.1 kg of methanol is added and roughly 0.13 kg crude glycerine is produced for each kg of oil processed.

The specific CO_2 emissions applied to the CO_2 balances are 385 g per kWh electricity produced considering the European average and 260 g per kWh thermal energy assuming a natural gas fired boiler [31]. A summary of all input parameters is given in Table 8. As described above, only the main processes cultivating and utilizing microalgae are taken into account while this work should not be considered as life cycle assessment.

The energy balances are shown in Figures 7 and 8. At first sight both cultivation technologies show a positive energy balance. The energy consumed compared to the energy

PCE for raceway ponds	1.5	%
Biomass concentration for cultivation with raceway ponds	0.8	kgDM*/m ³
Electricity demand for cultivation with raceway ponds	1	W/m ²
PCE for closed photobioreactors	5	%
Biomass concentration for cultivation with closed photobioreactors	4.0	kgDM*/m ³
Specific reactor volume for cultivation with closed photobioreactors	0.05	m^3/m^2
Electricity demand for cultivation with closed photobioreactors	100	W/m ³
Lower heating value microalgae biomass	30	MJ/kgDM*
CO ₂ bound in microalgae biomass	2.5	kgCO ₂ /kgDM*
Lipid content of microalgae biomass TAG***	40	%
Lower heating value microalgae oil	38.2	MJ/kg
Lower heating value microalgae press cake	23	MJ/kgDM*
Separation efficiency of centrifuge	90	%
Electricity demand centrifuge	3.6	MJ/m ³
Oil separation efficiency	90	%
Electricity demand for oil extraction	0.35	MJ/kgDM*
Heat demand for oil extraction	1.75	MJ/kgDM*
Conversion rate algae oil into biodiesel	90	%
Electricity demand conversion to biodiesel	0.043	MJ/kg
Heat demand for conversion to biodiesel	0.750	MJ/kg
Lower heating value of biodiesel	37	MJ/kg
Conversion efficiency microalgae press cake to biogas	90	%
Electricity demand conversion to biogas	0.48	MJ/nm ^{3**}
Heat demand for conversion to biogas	1.5	MJ/nm ^{3**}
Lower heating value biogas	20	MJ/nm ^{3**}
CO ₂ emission of electricity	107	gCO ₂ /MJ
CO ₂ emission of heat	72	gCO ₂ /MJ
Lower heating value of fossil diesel	42.5	MJ/kg
CO ₂ emission of fossil diesel	3.1	kgCO ₂ /kg
Lower heating value of natural gas	43.2	MJ/kg
CO ₂ emission of natural gas	2.4	kgCO ₂ /kg

TABLE 8: Summary of estimated parameters assumed for calculation ([9, 26, 30–33] and own calculations).

^{*}DM: dry matter; ^{**}nm³: standard cubic meter, ^{***}TAG: triacylglyceride.

yield for raceway ponds accounts for circa 70% and in case of closed photobioreactors for circa 77%. Closed photobioreactors show a much higher yield per surface area and therefore might be favourable considering land use as well as water demand as mentioned earlier. Unfortunately, the positive energy balance does not withstand a closer look for both cases as it is comparing apples to oranges. Values shown are the lower heating values of microalgal biodiesel and biogas compared to the electricity and heat consumed for their production. Assuming a highly efficient combined cycle power plant with an electric efficiency of 60% leads to the conclusion that in both cases less power can be produced from the fuels as is needed for their generation. The additional heat needed for processing is even neglected. The most significant energy consuming process steps are cultivation and harvesting. Energy input for these needs to be significantly reduced if microalgae technology will enter the commodity market for sustainability but also for cost reasons.

Due to the high power demand compared to the energy yield the CO_2 balances are also negative, as presented in Figure 9. Vast efforts need to be done to meet today's as well

as futures European Union sustainability criteria for biofuels. Requested CO_2 reductions are 35% today rising to 50% until 2017 and even 60% to 2018 [63]. Whether and how development of microalgae cultivation and harvesting technology will be capable of meeting these goals is open.

3. Conclusion and Outlook

The numerous benefits of biofuel generation from microalgae are motivation for further analysis and development: biomass yields of up to 100 to 150 tons dry matter per ha and year with lipid contents of around 40%, biodiesel yields of 40 to 50 tons per ha and year by far exceed the most promising yields from land crops. Fresh water demand can be reduced applying closed photobioreactors, using brackish water or seawater by a factor of one to two orders of magnitude. Algae oil quality generally is suitable for biofuel production. Because of the high amount of polyunsaturated fatty acids, microalgal biofuel shows good flow properties under low temperatures reducing the risk of cold filter plugging and making it even suitable for aviation fuel. Special is the ability

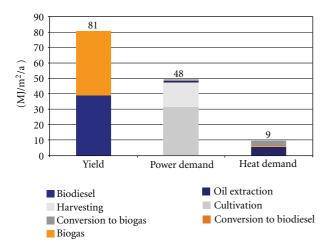


FIGURE 7: Comparison of energy yield and energy consumption producing biodiesel and biogas from microalgae applying raceway ponds (solar radiation 7,000 MJ/m²/a).

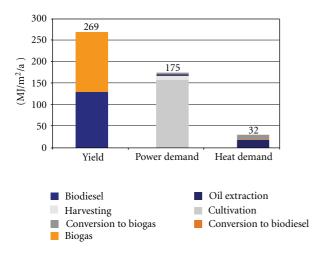


FIGURE 8: Comparison of energy yield and energy consumption for biodiesel and biogas production from microalgae applying closed photobioreactors (solar radiation 7,000 MJ/m²/a).

to influence the oil composition by the selection of the strain cultivated and the cultivation conditions themselves. This might be one piece of the solution to overcome the so far bad economics.

State-of-the-art for cultivation systems are raceway ponds with low investment cost but also low yields producing for food and feed market while high value products are produced with closed photobioreactors accepting high costs sometimes even applying artificial lighting. Both systems are not optimized to commodity markets needs especially concerning low overall efficiency due to auxiliary energy demand, sophisticated process control, and low product concentrations. Reduced energy and water demand as well as cheap nutrient supply are of lower rank while high product quality is required. The energy as well as greenhouse gas balances for those production systems including downstream processing of the microalgae biomass into biofuels show negative results. Reasons are the excessive auxiliary power

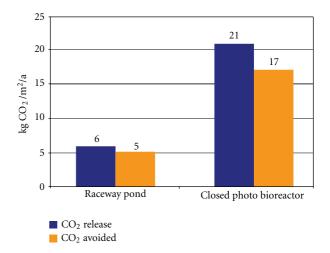


FIGURE 9: CO_2 emissions released during microalgae biodiesel and biogas production compared to emissions avoided due to substitution of fossil fuels (solar radiation 7,000 MJ/m²/a).

demand for cultivation and harvesting. At least a balanced scenario seems to be feasible assuming best case conditions. To meet the EU sustainability criteria for biofuels, which means a greenhouse gas emission reduction of at least 35% today and 60% starting from 2018, is from today's perspective very ambitious. Whether algae technology will be able to meet those targets is questionable.

However, key issues to meet commodity markets needs are identified and feasible solutions are under investigation. Closed photobioreactors show not only a much higher yield per surface area but also larger potential for process adaptation and control specific to the strain selected and products produced. Therefore, photobioreactors seem to be more promising. Possible solutions to reduce energy demand and increase yields are enhanced reactor designs, optimized process control, adapted separation technologies as well as integral concepts considering biology, process design for cultivation, and downstream processing to meet the different requirements for the specific target markets addressed in biorefinery concepts. But R&D needs are still high to make algae technology a serious part of the solution of sustainable fuel supply in future. Although pilot projects are ongoing so far, there is no demonstration facility where such measures are implemented showing the promised performance and cost reductions or even being suitable for up scaling.

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