REVIEW

Biodiesel production from microalgae and other applications

Imane Haoujar ⊙ . Karim Radouane . Ammar B. Altemimi . Francesco Cacciola ⊙

© The Author(s) 2023

Abstract Due to their extensive application potential in biopharmaceuticals, nutraceuticals, and renewable energy, microalgae have recently drawn considerable attention worldwide. Biofuels, bioactive medicinal products, and food ingredients are made from microalgae, a renewable, sustainable, and economic resource. A number of microalgae species have shown remarkable pharmacological and biological properties that have made them useful as value-added products. The cost, renewable nature, and environmental concerns of biofuels make them a perfect substitute for liquid fossil fuels. As well as carbohydrates, lipids, and other bioactive metabolites, microalgae convert atmospheric CO2 into useful products. There are some limitations and challenges to overcome before microalgae can be upgraded from pilot stage to industrial scale, though they are feasible sources of bioenergy and biopharmaceuticals in general. Enhancing microalgae growth rate and product synthesis is the most challenging and crucial issue, as is dewatering algae cultures, pretreating biomass, and optimizing fermentation processes. Various bioactive compounds as well as microalgae advantages for biofuel production are discussed in this review.

Keywords Microalgae . Algae cultures . Biofuel production . Growth rate . Pretreating biomass . Value-added products

Introduction

Photosynthetic algae can grow in a variety of aquatic habitats, including lakes, ponds, rivers, oceans, and even wastewater. Microalgae can grow in reservoirs, deserts, and other symbiotic conditions and tolerate wide ranges of temperature, salinity, and pH values (Barsanti et al. 2008). It is generally believed that algae can be classified into *Rhodophyta*, *Phaeophyta*, and *Chlorophyta*, according to their color. Biofuels, health supplements, pharmaceuticals, and cosmetics can be made with microalgae, which contain carbon compounds (Brennan and Owende 2010). Furthermore, they can be used for the treatment of wastewater and the mitigation of atmospheric CO_2. In addition to polysaccharides, lipids, pigments, proteins, vitamins,

Imane Haoujar (✉)
Laboratory of Biotechnology and Applied Microbiology, Department of Biology, Faculty of Sciences of Tetouan, Abdelmalek Essaadi University, 93000 Tetouan, Morocco
e-mail: imane.haoujar2015@gmail.com

Karim Radouane
Department of human sciences option of agricultural extension, National School of Agriculture of Meknes, 50000 Meknes, Morocco

Ammar B. Altemimi
Food Science Department, College of Agriculture, University of Basrah, 61004 Basrah, Iraq

Ammar B. Altemimi
College of Medicine, University of Warith Al-Anbiyaa, 56001 Karbala, Iraq

Francesco Cacciola (✉)
Department of Biomedical, Dental, Morphological and Functional Imaging Sciences, University of Messina, 98125 Messina, Italy
e-mail: cacciolaf@unime.it

bioactive compounds, and antioxidants, microalgae produce a wide range of bioproducts (Brennan and Owende 2010). In the biorefinery industry, microalgae have become a new focus due to its potential for renewable and sustainable biofuel production. Genetic engineering and growth enhancement techniques could improve their potential as renewable bioproduct sources in the future. Over the last few decades, industrial microalgae cultivation has increased dramatically for biofuels and bioproducts (Plaza et al. 2009). Global energy consumption continues to increase due to the rapid growth of the world's population. Since fossil fuels are unsustainable and nonrenewable, their intensive use worldwide leads to their depletion and exhaustion. There is currently a growing opportunity for biofuels around the world to replace fossil fuels in a sustainable way. Some developed countries are currently producing biofuels at a commercial level. Several biomass resources can be used for the production of biofuels, including food crops, plant wastes, woody parts of trees, garbage, and algae, among others. Biofuels have proven to be excellent alternatives to petroleum fuels in a variety of ways. In addition to being renewable, biomass-based biofuels contribute significantly less to global warming and environmental pollution.

CO_2 emissions from fossil fuel combustion are responsible for most global warming. With a total of 35.3 billion tons of CO_2 released to date, fossil fuels are responsible for 29 gigatons of CO_2 released each year (Paul Abishek et al. 2014). Petroleum-based fuels emit high levels of sulfur while biofuels like algal fuels have oxygen levels of 10 to 45%. A biofuel is an environmentally friendly, local, accessible, sustainable, and reliable fuel derived from renewable resources. Using microalgae algae-based fuels can reduce global CO_2 emissions while being eco-friendly, nontoxic, and nonpolluting. The fixation of CO_2 by algal biomass has been reported to be 1.83 kg per kg of biomass; additionally, SO_X and NO_X are used by some species as nutrients (Gendy and El-Temtamy 2013). A crucial aspect of biofuels generation is selecting and developing biomass in order to optimize energy structure and cost. Greenhouse gas emissions, environmental sustainability and economic sustainability are directly related to the choice of biomass for biofuel production (Cerri et al. 2017). A focus is currently being placed on microalgae as a raw material for biofuel production in order to balance and compensate the ever increasing demand for biofuels, food, feeding, and valuable chemicals (Ho et al. 2010; Paul Abishek et al. 2014). Bioenergy from microalgae biomass is being industrialized in several Asian, European, and American countries.

Photosynthesis by microalgae converts 9–10% of solar energy (average sunlight irradiance) into biomass each day, which is about 280 tons per hectare per year (Melis 2009; Formighieri et al. 2012). The yield of this culture system is lower in both outdoor and indoor systems at a larger scale of cultivation. The actual yield of photobioreactors is lower due to the loss absorbed active radiation. It is therefore necessary to shake and mix the culture properly to ensure uniform light distribution (Medipally et al. 2015).

There are many microalgae species suitable for biodiesel production because they contain high lipid contents of 50–70% and may even reach 80% as with the microalga *B. braunii*, which accumulates 80 percent oil (Mata et al. 2010). Biodiesel can be produced from microalgae by producing 58,700 L/hac of algal oil (Gouveia and Oliveira 2009; Gendy and El-Temtamy 2013; Medipally et al. 2015). Currently, algal fuels are in their infancy, and much improvement is needed before investors and consumers find them appealing.

Algae culturing

Microalgae are adapted to scavenge for resources in the environment, store them, and increase their efficiency in utilizing those resources. Microalgae typically require sufficient carbon sources and light to carry out photosynthesis in order to grow biomass (which consists of 40–50% carbon) (Moheimani 2005; Mata et al. 2010). Adaptations such as biochemical adaptations and physiological adaptations can occur within their internal structure, as well as the release of a wide range of compounds aimed at, for example, making nutrients available and limiting competitors' growth (Richmond 2004). As a response to changes in environmental conditions, microalgae can shift from autotrophic to heterotrophic, mixotrophic to photoheterotrophic metabolisms. Some organisms, for instance, can grow (Chojnacka and Marquez-Rocha 2004):

- Using light as a sole source of energy and converting it to chemical energy is called photoautotrophism.

- Organic compounds are used exclusively as carbon and energy sources heterotrophically.

- In a mixotrophic ecosystem, photosynthesis is the primary source of energy, although organic compounds and CO_2 are equally important.

- Depending upon the concentration of organic compounds and the intensity of light available, amphitrophy is a subtype of mixotrophy that enables organisms to live either autotrophically or heterotrophically.

- Light is required for organic compounds to be metabolized photoheterotrophycally, also called photoorganitrophy, photoassimilation, and photometabolism.

Photoheterotrophs and mixotrophs can be distinguished based on the source of energy they use to perform growth and produce specific molecules. Microalgae growth stoichiometry can also be used to distinguish the metabolism involved. There are several strains that grow under photoautotrophic, heterotrophic, and mixotrophic conditions, including *Chlorella vulgaris*, *Haematococcus pluvialis*, and *Arthrospira (Spirulina) platensis*. Photoautotrophic, heterotrophic, or photoheterotrophic growth is possible in strains such as *Selenastrum capricornutum* and *Scenedesmus acutus* (Chojnacka and Marquez-Rocha 2004). Besides organic carbon (they need sugars, proteins, and fats as carbon sources), vitamins, salts, and other nutrients (nitrogen and phosphorus), algae also require an equilibrium between operational parameters (pH, temperature, oxygen, carbon dioxide and light intensity) (Williams 2002).

With the right climate and the right nutrients, microalgae can grow profusely. When they are in the exponential growth phase, they usually double their biomass within 24 hours or within 3.5 hours (Chisti 2007).

Fig. 1 shows a growth curve for algae based on batch culture (solid line) and nutrients concentration (dashed line), with five phases clearly defined: (1) lag phase, (2) exponential phase, representing the maximum growth rate under the specified conditions, (3) linear phase, (4) stationary phase, and (5) decline phase. As the stationary phase progresses, the dashed curve (in Fig. 1) moves in the opposite direction, showing nutrients being depleted. Cultures of exponentially growing algae usually contain more proteins, whereas those of stationary growth usually contain more carbohydrates and glycogen (De Pauw et al. 1984).

Increasing the volume of algae or creating a semi-sterile environment can lead to a premature collapse, or other species more suited to outdoor conditions can take over. Consequently, high-density cultures should be defined better in the exponential growth phase, when they are developed under unbalanced growth conditions. In addition to abiotic factors like light (quality and quantity), temperature, nutrient concentration, oxygen, carbon dioxide, pH, salinity, and toxic chemicals, algal growth is also influenced by biotic factors like pathogens (bacteria, fungi, viruses) and competition from other algae. The operation factor is the shear generated by mixing, the dilution rate, the depth, the harvest frequency, and the addition of bicarbonate.

Light

Growing microalgae is limited by light intensity. Biochemistry and biomass yield of microalgae are directly affected by light intensity and duration. Modeling a bioreactor or open pond system should take into account that light intensities vary inside the culture and reduce as culture depth increases. To grow and accumulate biomass to the fullest extent, algae species have different light requirements (De Meester et al. 2016).

Microalgae cannot grow efficiently at very low and very high light intensities (Mata et al. 2010; Ye et al. 2012; Altonen et al. 2020). When photosynthetic CO_2 is uptaken and released in equal quantities, there
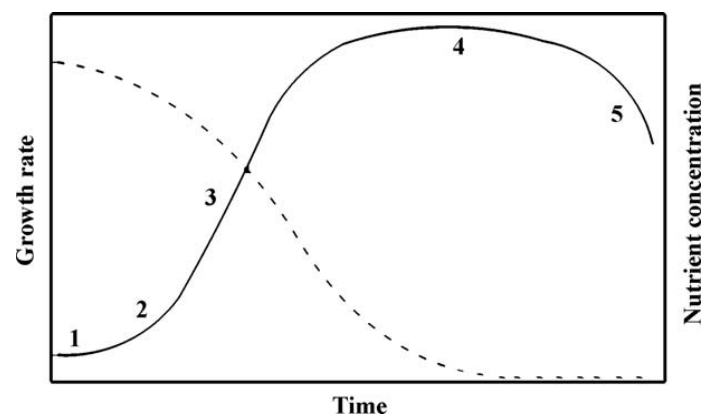

Fig. 1 Schematic representation of algae growth rate in batch culture (solid line) and nutrients concentration (dashed line)

is no net growth. When photorespiration and photoinhibition are balanced, the photosynthetic rate will increase with increasing light intensity. For maximum CO2 assimilation, it is necessary to determine optimal light intensity experimentally with a minimal rate of photorespiration and minimal photoinhibition (Ye et al. 2012).

The algal photosynthesis process requires specific light and dark periods. To produce carbon skeletons, ATP and NADPH are synthesized by light (Cheirsilp and Torpee 2012). The growth of microalgae is directly related to light intensity and duration up to the saturation point. Under different light intensities and for varying periods of time, Khoeyi et al. (2012) showed different growth rates and biomass yields for the same algae strain. Biomass productivity and growth rate decreased as light duration decreased (Jacob-Lopes et al. 2009). Algae grow most effectively when exposed to 16 hours of light and 8 hours of darkness. To avoid photooxidation and growth inhibition, microalgae require adequate light intensity and duration (Carvalho et al. 2011). Algae in the lower layers should have sufficient penetration and uniform distribution of light to avoid photoinhibition. In addition to LED lights, fluorescent tubes are also suitable for this purpose (Wu 2016). According to Mata et al. (2010), aerated cultures of microalgae under 12,000 lx intensity during daylight hours yielded a higher biomass yield. Reducing light intensity, however, reduces biomass. Using 5000 lx red LED light, Khan et al. (2016) reported maximum biomass and carbohydrate productivity for Microcystis aeruginosa. Most recently, Daliry et al. (Daliry et al. 2017) reported that Chlorella Vulgaris grows fastest and produces most lipids when exposed to light intensities of 5000–7000 lux. Most microalgae species prefer light intensities of 200–400 μM photons/m2/s. In order to prevent photoinhibition, high light intensity or continuous culture mixing is necessary, which can be achieved by increasing the photons/m2/s level (Altonen et al. 2020).

As a result, light directly affects algal biomass yields as well as carbohydrate synthesis and accumulation. The experimental study by Kitaya et al. (Kitaya et al. 2005) proved that some microalgal species can be effectively grown at a light intensity of 100 mol/m2/s.

Temperature

In both closed and open outdoor systems, the temperature is the most important limiting factor. The temperature of some microalgae can be decreased up to 15 °C below their optimal, but a temperature increase of only 2–4 °C may cause the cultures to die When the carboxylase activity is reduced at low temperatures, an excessive amount of energy is produced if the light conditions remain the same. Some cells can handle these conditions as a result of the imbalance that creates light saturation. The chlorophyll content of cultures obtained at 5 °C was lower than those at 27 °C, for example (Maxwell et al. 1994). As a result of the excessive light, cells adapted their photosynthetic apparatus to grow at 5 °C. The same acclimation has been found with *Dunaliella Salina* (Krol et al. 1997). Similarly, *Skeletonema costatum* increased its chlorophyll content when growth temperature was lowered. The overproduced energy was consumed by a carboxylase activity that gained activity. A closed culture system may also overheat on some hot days, when the temperature inside the reactor may reach 558 degrees Celsius.

While microalgae are able to grow under high temperatures only in certain species, other species are not. In addition to being thermophilic, *Chlorella sorokiniana* grows optimally at temperatures reaching 40 °C. In spite of this, growth beyond an optimal temperature is rarely reported for mesophilic or thermophilic species. The deleterious effects of a higher temperature are greater than those of a low temperature. Moreover, when optimal temperatures are exceeded, photosynthesis, respiration, and growth decline primarily due to the imbalance between energy demand and ATP production, but also due to the inactivation or denaturing of photosynthesis-related proteins (Raven and Geider 1988). Both *Microcystis aeruginosa* and *Scenedesmus acutus* showed lower respiration rates and higher photosynthesis rates during elevated temperature acclimation, Staehr and Birkeland (2006) found.

It is generally recognized that every microalga species has an optimal growth temperature (Table 1). At optimal growth temperatures, cells can undergo photosynthesis without undergoing any biochemical or physiological changes. For mesophilic species, the maximum growth rate is reported between 20 and 25 °C. However, thermophilic strains (*Chaetoceros*, *Anacystis nidulans*) can reach 40 °C, and psychrophilic strains can reach 17 °C (*Asterionella formosa*).

Nutrients

Various microalgae species have different nutritional requirements, but all species have the same basic needs. Algal growth depends on the macronutrients nitrogen, phosphorus, and carbon ($CH_{1.7}O_{0.4}N_{0.15}P_{0.0094}$) (Juneja et al. 2013) lipids. It is also necessary for some marine microalgae species to receive silicon as a macronutrient. Different species of microalgae can contain different amounts of macronutrients such as nitrogen and phosphorus. As nitrogen and phosphorus concentrations decreased from 31.5 to 10.5 mg/l, chlorella growth declined (Aslan and Kapdan 2006). Nitrogen levels directly affect the growth of cells in culture. While microalgae culture may produce more carbohydrates and lipids under nitrogen limitation, they can also produce less growth and biomass. It has been proven that *Chlorella vulgaris* produces 3.43 grams of biomass at 0.5 grams of nitrogen per liter (Daliry et al. 2017).

There is no evidence that Mo, K, Co, Fe, Mg, Mn, B, and Zn are necessary in trace quantities for microalgae to grow, yet they are crucial for many of the enzymatic processes in alga cells (Gardner-Dale et al. 2017) but their successful implementation requires an understanding of how design decisions influence nutrient uptake over daily (i.e., diel. Nitrates and phosphates are the most common forms in which inorganic nitrogen and phosphorus are absorbed. As an alternative to inorganic nitrogen sources, urea is also a useful and cost-effective source.

Algae cultures can be supplied with carbon in organic forms, such as glycerol or acetate, or as CO_2. In order to cultivate microalgae on a large scale, however, environmental CO_2 must be used as a carbon source, in addition to its low cost and ability to minimize CO_2 emissions. Inorganic nutrients P, N, and C are essential for the growth of microalgae. Low biomass is caused by nutrient deficiency in microalgae (Khozin-Goldberg and Cohen 2006; Hu et al. 2008; Prathima Devi and Venkata Mohan 2012)17.5 and 0μM ($K2HPO4$). Microalgae accumulate carbohydrates and lipids according to their nutrient supply (Prathima Devi and Venkata Mohan 2012) growth. To produce microalgae biomass commercially, the culture must grow rapidly; therefore, providing the right nutrients is essential. It is possible to enhance microalgae growth by using some strongly limiting substances.

Mixing

In addition to homogenization of the cell distribution, mixing also facilitates the transfer of gases, heat, and metabolites among the cells. In addition, turbulence is necessary in large-scale reactors to accelerate microalgae cell circulation from the dark to the light zones. Microalgae can be damaged by shear stress due to high liquid velocities and degrees of turbulence (as a result of mechanical mixing or air bubble mixing) (Eriksen 2008).

Salinity

A microalgae's growth and cell composition can be influenced by salinity, both in an open and closed

Table 1 The optimal growth rates for 15 species

Species	Tmin	Topt	Tmax	References
A. formosa	-7.3	20.1	29.8	Butterwick et al. (2005)
Ceratium furca	8.4	24.4	32.1	Baek et al. (2008)
Ceratium furcoides	6.9	22.3	30	Butterwick et al. (2005)
Ceratium fusus	4.2	26.5	30.7	Baek et al. (2008)
Chlorella pyrenoidosa	5.2	38.7	45.8	Sorokin and Krauss (1962)
Cryptomonas marssonii	-2.4	15.9	30.3	Butterwick et al. (2005)
Dinobryon divergens	-5.8	17	28.4	Butterwick et al. (2005)
D. tertiolecta	5.0	32.6	38.9	Eppley and Sloan (1966)
Nannochloropsis oceanica	-0.2	26.7	33.3	Sandnes et al. (2005)
P. tricornutum	-27.7	22.5	25.2	Kudo et al. (2000)
Porphyridium cruentum	5.8	19.1	30	Dermoun and Chaumont (1992)
Scenedesmus sp.	-3.1	26.3	32.7	Xin et al. (2011)
S. costatum	8	24.5	33	Butterwick et al. (2005)
Tychonema bourrelyi	0.4	21.8	30	Butterwick et al. (2005)

system. Due to high evaporation during hot weather conditions, every alga has its own optimum salinity range. There are three main ways in which salinity changes affect phytoplankton (Moheimani 2005): The changes in cellular ionic ratios are induced by three factors: (1) osmotic stress; (2) ion (salt) stress; and (3) the selective ion permeability of the membranes. Adding fresh water or salt as needed is the easiest way to control salinity.

Producing microalgae is significantly affected by the cost of using fresh water. Microalgae grow best in areas where freshwater is scarce or nonexistent. Therefore, saline water is an inevitable option for growing microalgae in large scales. This can be in the form of seawater or underground saline water (Borowitzka et al. 2013). Nevertheless, using any type of saline water poses another challenge. The growth of microalgae is accelerated by increased irradiance in areas with high levels of sunlight. Increasing light intensity prevents evaporation, which increases salinity levels, especially in open ponds (Ishika et al. 2017). High-salinity water can be suitable for three types of microalgae: marine, halotolerant, and halophilic. There are different types of microalgae that react differently to high salinity, and salinity is an important parameter for their growth (Yeesang et al. 2011). According to Laing and Utting (1980), *Tetraselmis suecica* ranges from 2.5–3% and *Isochrysis galbana* from 1.5–2.5%. As a result of salinity, microalgae can undergo three types of stress: osmotic stress, ion stress due to uptake or loss of ions, and ionic ratio changes (Kirst 1990). Inhibition of photosynthesis can be caused by changes in osmotic pressure or swelling, which may result in the cells burst (Fogg 2001). As salinity levels increase in a culture, stress is more likely to be induced on the cells. Stressed cells grow slower and accumulate lipids during the growth phase, when cell numbers are increasing. It is crucial to understand how changes in salinity, either gradual (from evaporation) or sudden (from recycling medium), affect growth when saline water is used for microalgae cultivation (Wang et al. 2009).

Pretreatment of algal biomass

In addition to algae and mould, fungi, yeast, and bacteria are some of the most common biological contaminants observed in wastewater. Microalgae cultures cultivated in raceway ponds collapsed due to protozoa and contamination by other algae.

The Moheimani (2005) method suggests that after removing unwanted organisms, the culture needs to be subjected to a temporary change in light, temperature, pH, or other environmental factors in order to decrease contaminants. In closed cultivation reactors, the closed environment, greater control over culture parameters, and higher cell concentrations make it possible to cultivate some important microalgae without contamination.

Stress conditions

Authors have examined various factors that affect algal growth:

- A reduction in the medium pH caused by CO_2 inhibited the growth of algae according to Moheimani (2005). As determined by this study, a plate photobioreactor should maintain a pH range between 7.7 and 8.0, and an outdoor raceway pond should maintain a pH range between 9.1 and 9.6 for Pleurochrysis carterae. A depth of 16 to 21 centimeters was also determined as the optimal operating depth for the outdoor raceway pond by this author.

- Using shallow suspensions of CO_2 at near neutral pH is difficult to control since the bubbles do not have enough time to absorb CO_2, resulting in great atmospheric losses of CO_2. There is a considerable loss of CO_2 to the atmosphere when CO_2, which is supplied in shallow suspensions at near neutral pH, is supplied due to a lack of residence time of the bubbles (Richmond 2004).

- There are two major uncatalyzed reactions that may accelerate CO_2 absorption into alkaline waters, according to Weissman and Goebel (1987): hydration of CO_2 and subsequent acid-base reactions to form bicarbonate ions, or direct reaction of CO_2 with hydroxyl ions to form bicarbonate. According to this author, the first reaction occurs faster at pH values below 8, while the second occurs at pH levels above 10. Both can be important between 8 and 10.

- Chiu et al. (2009) found that *Nannochloropsis oculata* cultures produced more biomass and lipids with increased CO_2 concentration.

- De Morais and Costa ([2007](#)) also found similar results from *Scenedesmus obliquus* and *Chlorella kessleri* cultures isolated from a treatment pond in a coal-fired thermoelectric power plant in Brazil, suggesting that these microalgae are capable of fixing carbon dioxide in thermoelectric power plants.

- Aeration and light intensity are also important factors. According to Mata et al. ([2010](#)) a bioreactor with airlift grew better than a bioreactor without airlift.

The reason for this is that aeration improves the mixing of the microalgal culture, preventing sedimentation, maintaining homogeneous conditions, and improving vitamin and nutrient contact. Increasing light intensity increases cell density and specific growth rate, but growth is inhibited above a certain limit as light intensity increases.

- Using the mineral medium carbon dioxide (and bicarbonate) as a carbon source and nitrate as nitrogen source, Thomas ([1984](#)) investigated photosynthetic algae species that grow autotrophically. In this study, the proximate chemical compositions of algae are examined in relation to nitrogen as well as salt stress. Environmental stress results in an alteration of the basic cellular composition of the tested species. In *Botryococcus*, the highest concentration of lipids, with the greatest proportion being hydrocarbons, was found under non-stressed conditions. Lipid levels for the green algae averaged 23% per organic weight, while *Nitzschia* sp. averaged 12% and Isochrysis has a 7% rate.

- The marine strain *C. vulgaris* was also found to accumulate considerable amounts of lipid when exposed to high iron concentrations, as demonstrated by Liu et al. ([2008](#)). The presence of high concentrations of iron in the initial medium may modify some metabolic pathways related to lipid accumulation in *C. vulgaris*.

- In their study, Illman et al. ([2000](#)) found that nitrogen reduction led to increased lipid content in all five Chlorella strains studied, including *C. emersonii, C. minutissima. C. vulgaris* that were gained an increase in lipid contend of 63%, 56% and 40% biomass by dry weight respectively.

- A study by Thomas et al. ([1984](#)) examined the effect of nitrogen stress on the neutral lipid content of algae, concluding that cultivation conditions deficient in nitrogen increased neutral lipid content, but this cannot be summarised as a single trend. In nitrogen-stressed cultures of *Botryococcus, Isochrysis*, and *Dunaliella*, this behavior is observed. In *Botryococcus*, neutral lipids account for the majority of the total lipids. Neutral lipids are produced most during the resting stage of algae, and the greatest amount occurs when the algae move from a green to a brown growth phase. While the *Dunaliella bardawil* and *Dunaliella salina* lipid fractions increased by 10%, the *Botryococcus* lipid fraction decreased by 10%. As a result of nitrogen stress, these halotolerant green algae started storing carbohydrates. In contrast, Isochrysis accumulated more carbohydrates and lipids in nitrogen deficient conditions, and lipids accounted for about one-fourth of the organic mass of Isochrysis. Nitrate deficiency generally decreased protein levels, chlorophyll levels, carbohydrate levels, and lipid levels, depending on the species. Algae under environmental stress manifest neutral lipid content by shifting to lipid storage. The neutral lipids are primarily multibranched and polyunsaturated hydrocarbons rather than straight chain saturated hydrocarbons.

- As Macedo and ([2001](#)) found, the concentration of lipids in *Spirulina* increases approximately 3 times as nitrogen concentration decreases and temperature decreases, with nitrogen concentration decrease having the greatest effect on lipid content.

Harvesting and biomass concentration

During the algal harvesting process, biomass is recovered from the culture medium, which can account for 20–30% of the cost of biomass production (Grima et al. [2003](#)). To perform solid-liquid separation, a suitable harvesting method may require one or more steps and be achieved physically, chemically, or biologically, in order to remove large quantities of water and process large algal biomass volumes. While there is no universal harvesting method, research continues to be carried out in this area, as any algal species can be harvested in an appropriate and economical manner.

Sifting, centrifugation, filtration, and ultrafiltration, along with flocculation or flotation combined, are the most common harvesting procedures. As a result of flocculation, the microalgal cells are aggregated into larger balls that are easier to sedimentate, centrifuge, and filter (Grima et al. [2003](#)). Microstraining, belt filtering, flotation with floating collection, and sedimentation were cited as four primary harvesting methods by Weissman and Goebel ([1987](#)). Separation of biomass is performed using size and density

discrimination. The mechanical simplicity and large unit size of microstrainers make them a popular harvesting method. Microalgae harvesting has recently been revived by the availability of very fine mesh polyester screens. The cells must be flocculated prior to microstrain, according to subsequent studies.

It is possible to recover large quantities of biomass using filter presses that operate under pressure or vacuum, but filtration may be relatively slow and unsatisfactory for certain applications. Additionally, in a different study it has been demonstrated that filtration is more effective at removing bacteria that have large dimensions, such as *Coelastrum proboscideum* and *S. platensis*, but it is not as effective at recovering bacteria with a smaller diameter, such as *Scenedesmus*, *Dunaliella*, or *Chlorella* (Grima et al. 2003). For small-scale production processes, microfiltration and ultrafiltration are more suitable alternative filtration methods than conventional filtration to recover algal biomass. In addition, membrane replacement and pumping are more expensive with these filtration processes.

Processing and components extraction

It is extremely difficult to produce low cost commodities (fuels, feeds, and foods) without processing, and it is extremely difficult to produce higher value products (β-carotene and polysaccharides). Considering that processing is highly specific and heavily depends on the desired result, it can be difficult to discuss. There is a common practice of dehydrating biomass, which also extends its shelf-life. Spray-drying, drum-drying, freeze-drying, and sun-drying are some of the most common methods of drying microalgae, such as *Chlorella*, *Scenedesmus*, and *Spirulina* (Richmond 2004). In low-value products such as biofuel or protein, spray-drying is not economically feasible because algal biomass is high in water content. In order to release the metabolites of interest, the microalgae cells must be disrupted after drying. Microalgae walls and products can be processed using a variety of methods, depending on the type of product to be obtained, such as cell homogenizers, autoclaves, ultrasounds, and spray drying, or non-mechanical means (organic solvents, base, acid, such as freezing, osmotic shock, and enzyme reactions).

As an example, studying various methods of astaxanthin recovery, autoclaved and mechanically disrupted biomass yielded 3 times more astaxanthin than other methods (Richmond 2004). Algal cells are broken down and turned into fine powder by lyophilizing.

Microalgal biomass must be extracted for the production of biodiesel. Lipids are usually extracted via solvent from lyophilized biomass, being an efficient and fast extraction method that decreases degradation by a small amount. In addition to hexane and ethanol (96%), hexane-ethanol mixtures (96%) can also be used to obtain purified fatty acids to a purity of 98% (Richmond 2004). The use of ethanol is not recommended if the purpose of the extraction is in order to extract only lipids, as it can also extract sugars, salts, hydrophobic proteins, amino acids, and pigments. Various methods of oil extraction from vegetable sources were also studied, including ultrasound and microwave-assisted extraction. According to Cravotto et al. (2008) these methods provide similar yields and oil extraction times to conventional extraction methods. A multimode microwave oven that can operate both with open and closed vessels has been developed by these authors' research team in order to achieve this goal. The extraction was combined with simultaneous double sonication at 19 and 25 kHz as well as simultaneous ultrasound/multimode microwave irradiation. According to these results, these novel methods can significantly enhance oil extraction with higher efficiency compared to conventional methods. A 50%–500% increase in yield was achieved at a low or moderate cost, and minimal toxicity was introduced.

As ultrasound disrupted the tough algal cell wall, extraction yield improved considerably from 4.8% (in soxhlet) to 25.9% for marine microalgae *Crypthecodinium cohnii* (Cravotto et al. 2008).

Microalgae as potential sources of bioactive compounds and value added products

A blockage to commercialization remains the expensive cost of biomass production and downstream processing of microalgal biomass (Misra et al. 2014). As a result of their bioactive compounds, microalgae are also potentially useful in pharmaceutical, cosmetic, and nutritional applications. Polyunsaturated fatty acids (PUFAs), Phycobiliproteins, enzymes, pigments, sterols, and toxins are some of the bioactive compounds found in microalgae. A brief overview of microalgae-derived bioactive compounds is presented in this section. Microalgae produce many important compounds, and some of them have highly valuable bioactive properties.

Carotenoids

An important part of our daily diet are carotenoids, a group of terpenoid pigments with C40 backbones. Various species of aquatic life, microorganisms, and terrestrial plants produce over 1100 carotenoids that have been discovered to date (Lui et al. 2021). A variety of plants receive their yellow, orange, and red colors from these colorful molecules. During oxygenic photosynthesis, carotenoids serve as accessory pigments in the photosynthesis apparatus to harvest light or as structural molecules to stabilize protein folding (Herrero et al. 2013). Carotenoids are ingested, which produce vitamin A and help repair retinal damage caused by free radicals (Del Campo et al. 2007).Their anti-cancer, anti-inflammatory, and anti-obesity properties can also reduce the risk of some diseases. Because of this, carotenoids are widely used in food, feed, cosmetics, and pharmaceuticals (Del Campo et al. 2007).

In microalgae, carotenoids are classified as primary and secondary. Primary carotenoids are those that are accumulated during photosynthesis (Nisar et al. 2015). Astaxanthin, zeaxanthin, fucoxanthin, and fucocyanin are mainly discussed in this review.

A microalgal cell's biochemical components are affected by external environmental changes due to its flexible metabolic system. A high light intensity, salinity, nitrogen starvation, and high or low temperatures are all stress factors. For seawater culture of microalgae, salt stress treatment is far more attractive than several environmental abiotic stress factors because it is affordable, easy to operate, and offers great commercialization potential.

Proteins and enzymes

As well as having strong therapeutic effects on health, some proteins, peptides, and amino acids are necessary for the normal functioning of cells and tissues. Food is usually the only source of these nutrients when the human body cannot synthesize them. Many species of microalgae produce high levels of essential amino acids and proteins, which are useful for feeding people and preventing a variety of illnesses. As much protein as can be produced by microalgae can be produced by other sources of proteins, such as eggs, meat, and milk (Sousa et al. 2008). There is significant nutritional value to microalgae proteins. Microalgae produce 2.5–7.5 tons/Ha/year of proteins (Bleakley and Hayes 2017). Various types of proteins can be obtained from *Chlorella*, a green microalgae. *Arthrospira* is another microalgae rich in protein. As a result of the activation of cholecystokinin by microalgae and plants, cholesterol levels can be reduced. In addition, they are involved in enzymatic reactions (Smee et al. 2008). Cyanovirin, a protein produced by Nostoc, has been reported to have antiviral properties against HIV and influenza viruses (Zappe et al. 2008). As an example, *Isochrysis galbana* produces a vital enzyme carbonic anhydrase that converts CO_2 into carbonic acid and bicarbonate, while *Anabaena* and *Porphyridium* produce SOD (superoxide dismutase), which protects against oxidative damage.

Vitamins

There are many vitamins in microalgae, including A, B1, B2, B6, B12, C, E, pantothenic acid, biotin, folic acid etc. Among the nutrients found in *Isochrysis galbana* are vitamins A and E, nicotinic acid, folic acid, pantothenic acid, riboflavin, biotin, thiamine, cobalamin, pyridoxine, chlorophyll (a and C), diadinoxanthin and fucoxanthin and *Euglena gracilis* antioxidant can produce vitamins such as β-carotene and vitamins E and C (Mulders 2013).

Plants do not synthesize vitamin B12, which is a water-soluble vitamin. Vegans and vegetarians often suffer from vitamin B12 deficiency. Microalgae, such as *Chlorella* sp., and *Pleurochrysis carterae* can synthesize or contain vitamin B12 (Kumudha et al. 2015).

There are many microalgae capable of producing vitamin E, including *Tetraselmis suecica*, *Nannochloropsis oculata*, *Dunaliella tertiolecta*, *Porphyridium cruentum* and *Chaetoceros calcitrans*. Microalgae contain a higher amount of vitamin E than plants, according to a number of studies. The production of vitamin E from microalgae is valuable in this regard (Santiago et al. 2018).

Besides containing different vitamins, microalgae are rich in antioxidants. *Haslea ostrearia* is rich in tocopherols (vitamin E). Vitamins E and C are abundant in *P. Cruentum* and *Skeletonema marinoi*, as well

as pancreatic beta-carotene (vitamin A) (Mus et al. 2013). Among the vitamins and nitrates that *D. Salina* produces are A, B, C, D, thiamine, biotin, pyridoxine, and nicotinic acid. *Phaeodactylum tricornutum* and *Haematococcus pluvialis* were selected for their high levels of (carotene, fucoxanthin, astaxanthin, eicosapentaenoic, and arachidonic acid). Approximately 80% of the nutrients (vitamins E, B, C and pigments) were produced (Hosseini Tafreshi and Shariati 2009).

Lipids

The proportion of microalgal lipid ranges from 20% to 70%, and the composition of the fatty acids in algal cells is determined by genetic and physiological factors, as well as environmental factors (Ravindran et al. 2016).

There are two types of algal lipids; the polar lipids and the non-polar lipids. Polyunsaturated fatty acids (PUFAs) make up most of the polar lipids (also known as structural lipids). Humans and aquatic animals require these PUFAs for nutrition. Cell membranes are composed of sterols and polar lipids, which provide the matrix for metabolic processes. Cell signaling pathways also use it as an intermediate. Compared to polar lipids, nonpolar lipids are commonly known as storage or neutral lipids. Transesterifying these storage lipids can convert these saturated fatty acids into energy (biodiesel), especially triacylglycerols (TAGs) (Sharma et al. 2011). Biodiesel production and other algal biofuels require the analysis of lipid profiles in biomass feedstocks. Therefore, microalgae are more likely to accumulate lipids when they are under phosphate or nitrogen limitation. Various factors influence the composition of fatty acids in a species. These factors include the composition of the growth medium, the rate of aeration, the temperature, the ratio of light to dark, and the intensity of artificial illumination (Bellou and Aggelis 2013).

Polyunsaturated fatty acids

In addition to contributing to tissue integrity, polyunsaturated fatty acids are beneficial for the health of the body. The human body cannot produce omega-3 and omega-6 fatty acids, which are crucial to human health. The intake of DHA, linoleic acid, eicosapentaenoic acid, arachidonic acid, and gamma-linolenic acid is therefore essential from external sources such as foods or cosmetics. Research has shown that these acids reduce cholesterol levels, delay aging, protect membrane integrity, and prevent cardiovascular disease (Hu et al. 2002; Bannenberg et al. 2017). Researchers have explored several microalgae species for their ability to synthesize these valuable fatty acids, including *Porphyridium cruentum, Arthrospira platensis, Odontella,* and *I. galbana*. While Pavlova lutheri produces large amounts of polyunsaturated fatty acids, *A. platensis* produces stigmasterol, sitosterol, and eicosapentaenoic acid (Ghosh et al. 2015).

Omega-3 polyunsaturated acids such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are essential for the treatment of many diseases including arthritis, heart disease, and headaches. Fish oils are limited in quantity and are unable to supply the required amount of EPA and DHA in a sustainable and promising manner. Microalgae are the only alternative because they are a sustainable and promising source of these two essential fatty acids (De Jesus Raposo et al. 2013; Armenta and Valentine 2013). The metabolic
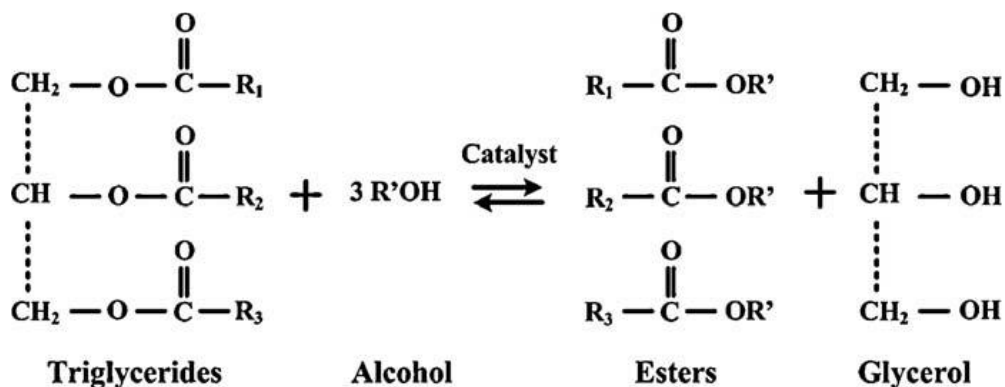

Fig. 2 Transesterification of triglycerides (overall reaction)

pathways of EPA and BHA have been altered through genetic manipulation in recent years to enhance their production (Adarme-Vega et al. 2014). There has recently been interest in the production of EPA and DHA from *Phaeodactylum tricornutum* (Draaisma et al. 2013; Hamilton et al. 2014; Koller et al. 2014). EPA and DHA, polyunsaturated fatty acids found in omega 3 compounds, have been genetically modified in the diatom *P. tricorornitum. P. tricornutum* strains genetically engineered to produced DHA and EPA have been reported to yield 36.5% and 23.6% of DHA and EPA per total fatty acids of biomass, indicating their commercial viability (Chauton et al. 2015). The low yield of these products, such as EPA and DHA, is a result of several hurdles and challenges that need to be overcome before they can be produced on a commercial scale (Hamilton et al. 2016). In order to increase production scale, several aspects must be optimized, including strain selection, culture development, and product induction and extraction methods. Carbon sources and light strength greatly affect algae growth and Omega-3 polyunsaturated fatty acid production (Yongmanitchai and Ward 1991; Wen and Chen 2003; Gardner et al. 2012).

Microalgae for biodiesel production

Vegetable oils or animal fats are transesterified to produce biodiesel, which consists of alkyl fatty acid esters. Approximately 90–98% of these lipid feedstocks are triglycerides. Small amounts of monoglycerides are also present, as well as free fatty acids (1–5%) and residual phospholipids, phosphatides, carotenes, and tocopherols (Bozbas 2008). The process of transesterification involves three separate reversible steps, starting with the conversion of triglycerides into diglycerides, then the conversion of diglycerides to monoglycerides, which is then converted into esters (biodiesel) and glycerol (by-product). According to Fig. 2, long chain hydrocarbons, such as fatty acids, are the radicals R1, R2, and R3. Transesterification occurs in the presence of a catalyst (usually NaOH) and oil or fat and an alcohol (usually methanol). In general, the molar ratio of 6:1 is used for accurate reaction completion, despite the alcohol:oil theoretical molar ratio of 3:1. As a result, one pound of oil produces about one pound of biodiesel, which is about 1:1 between feedstock mass input and biodiesel mass output. Transesterification reactions can be sped up with the use of homogeneous or heterogeneous, acidic or basic catalysts, though some processes (methanol and ethanol) may not require such catalysts (Warabi et al. 2004). Homogeneous alkali catalysts (such as NaOH or KOH) are commonly used in batch reactors in industrial processes. There have been recent improvements to this process, including the possibility of operating it continuously with a shorter reaction time, such as microwave assisted reactors (Cravotto et al. 2008; Azcan and Danisman 2008), cavitation reactors (Gogate 2008; Gogate and Kabadi 2009), and ultrasonic reactors (Kalva et al. 2009; Deshmane et al. 2009).

Microalgae lipid content and productivities

An increase in oil yield can be achieved by induced accumulation of lipids in many microalgae species (Sheehan et al. 1998). Most species contain between 1 and 70% lipids, but some have a higher percentage of lipids under certain conditions (Spolaore et al. 2006; Chisti 2007; Li et al. 2008). Several differences between marine and freshwater microalgae species can be seen in Table 1, which shows lipid content, biomass productivity, and lipid productivity (Zhu and Lee 1997; Mata et al. 2010). Table 2 shows that microalgae can have oil content up to 75% by weight of dry biomass, but low productivity (e.g. for *Botryococcus braunii*). Several different species (*Neochloris, Dunaliella, Chlorella, Isochrysis, Schizochytrium, Tetraselmis Phaeodactylum, Porphyridium, Crypthecodinium, Nannochloris, Nannochloropsis, Cylindrotheca, Nitzschia*) are possible to achieve higher productivity levels with oil levels between 20 and 50%.

In terms of biodiesel production, *Chlorella* is an excellent option. The selection of the most appropriate species has to take into account other factors, since other species are as efficient and productive as this one. For example, microalgae can develop under specific conditions or if nutrients are available. When selecting the most appropriate species or strains for biodiesel production, all of these parameters need to be taken into account simultaneously. Additionally, different microalgae species have different fatty acid compositions, which can affect biodiesel's characteristics. Some of them belong to the v3 and v6 families of saturated and unsaturated fatty acids. Seven fresh water microalgae species were analyzed by Thomas et al. (1984) and showed to synthesize C14:0, C16:0, C18:1, C18:2, and C18:3 fatty acids. According to this author, individual fatty acid chains vary in intensity according to species, including C16:4 and C18:4 in *Ankistrodesmus*

sp., C18:4 and C22:6 in I*sochrysis* sp., C16:2, C16:3, and C20:5 in *Nannochloris* sp., and C16:2, C16:3, and C20:5 in *Nitzschia* sp). Nutritional factors, environmental conditions, and growth phases may influence fatty acid composition. *B. braunni*, for example, accumulated C18:1 in response to nitrogen deficiency and salt stress (Thomas et al. 1984). Various algae species also have different fatty acid compositions, according to other authors (Poisson et al. 2002; Natrah et al. 2007; Gouveia and Oliveira 2009).

Microalgae biodiesel value chain stages

The schematic of biodiesel production from microalgae is shown in Fig. 3. Selecting the suitable species for the culture conditions and product is the first step (Pulz and Gross 2004). A number of factors need to be taken into consideration before starting a culture, air (carbon dioxide), pH, such as light, temperature and nutrient concentration. A variety of methods can be used for the harvest of microalgae, sedimentation, centrifugation, such as microscreening, flocculation and membrane filtration.

Drying under vacuum removes water from the harvested biomass until it achieves a constant weight. Oil is extracted by pulverizing the dried biomass with a mortar and pestle.

In the extraction of oil from microalgae, three methods are known: steam extraction, solvent extraction and supercritical fluid extraction (Demirbaş 2008). Soxhlet extraction uses hexane as a solvent and takes four hours to complete. Several other solvents can be used, including ethanol, petroleum ether and a mixture of hexane and ethanol. A number of methods can be used for converting raw oils into biodiesel: direct use, microemulsions, thermal cracking (pyrolysis) and transesterification among others. Ma and Hanna (1999) and Leung et al. (2010)especially alkali-catalyzed transesterification. When the raw materials (oils or fats provide comprehensive descriptions and comparisons of these methods. Transesterification, or alcoholysis, is the most common process for reducing oil viscosity due to its ability to produce fatty esters (biodiesel) (Demirbaş 2008). Transesterification is a process by which triglycerides are converted first into diglycerides, then into monoglycerides, and then into esters (biodiesel) and glycerol (byproducts). There
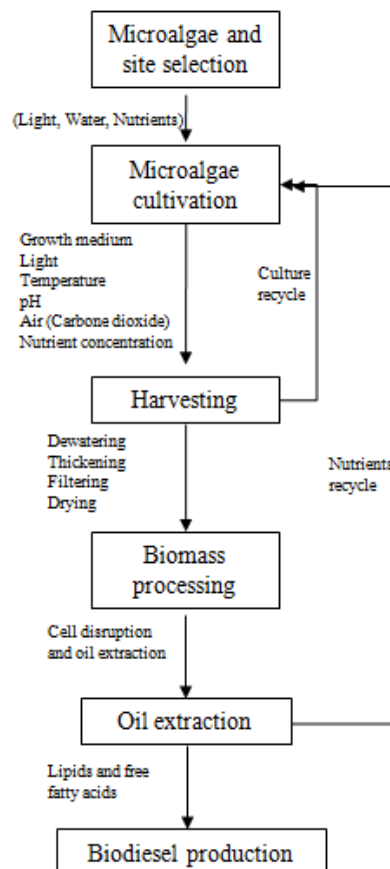

Fig. 3 Production of biodiesel from microalgae

are other sources of information about microalgae-based biodiesel production (Brennan and Owende 2010; Mata et al. 2010).
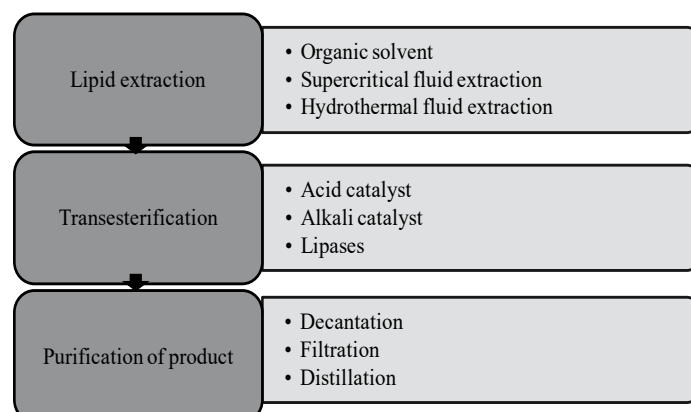
Transesterification of microalgal oil to biodiesel

Chemical transesterification approach

Alkali or acid catalysts can be used to transesterify microalgal lipids into biodiesel. An overview of downstream biodiesel production processes is shown in Fig. 3. Oils and fats can be transesterified to biodiesel faster and more efficiently with alkali-catalyzed transesterification compared to acid catalysis (Christopher et al. 2014). Oils with low acid values are preferable for biodiesel production since they avoid yield losses due to basic transesterification. In this case, soap can be produced in parallel with the free fatty acids, which results in the consumption of the catalyst and a reduction in its efficiency. Moreover, saponification increases viscosity, resulting in gels that complicate biodiesel separation (Yan et al. 2014). In order to remove these free fatty acids, an esterification step is recommended since microalgal oil has a high acid value (Tabernero et al. 2012)together with an evaluation of feasibility is performed in this article. The plant employs the microalgae *Chlorella* protothecoides to obtain biomass. The subsequent oil extraction is done with supercritical carbon dioxide. Based on previous studies, it was possible to determine the mass and energy balances and to design the equipment of the main process. A non-conservative study reveals the no feasibility of the production plant unless the residues are sold (two different and real prices were chosen. As a result of esterification, free fatty acids are converted into alkyl-esters by adding an alcohol, which produces biodiesel and water. A potassium hydroxide (KOH) catalyst is used to transesterify the oil after centrifugation and separation from the catalyst. An ester bond between fatty acids and the backbone of glycerol is cleaved by the catalyst during transesterification (Chisti 2008). After liberating fatty acids, FAME is formed by reacting them with methanol. The forward reaction is driven by a large amount of methanol in excess of stoichiometric requirements (6:1 molars) in laboratory scale experiments using a small quantity of crude microalgal lipids to drive the forward reaction and ensure quantitative transesterification (Stojković et al. 2014). Numerous authors have studied the kinetics of transesterification in biodiesel production (Darnoko and Cheryan 2000; Komers et al. 2002; Vicente et al. 2005) diglycerides (DG).

Enzymatic transesterification approach

The use of enzyme catalysts can result in high yield and quality biodiesel from low quality feedstocks with a high content of free fatty acids (Atadashi et al. 2012). Biodiesel can be converted into biodiesel with up to 98% purity using enzyme catalysts (Tran et al. 2012; Lai et al. 2012). To prevent enzyme activity loss, the enzymatic transesterification reaction often occurs at lower temperatures than acidic or alkaline reactions (Gog et al. 2012). There is a range of temperatures that work best for certain lipases used in the biodiesel synthesis process, between 30°C and 55°C (Iso et al. 2001; Haas et al. 2002). The optimal temperature range for converting microalgal oil extracted from *Chlorella vulgaris* ESP-31 was reported by Tran et

Lipid extraction	• Organic solvent • Supercritical fluid extraction • Hydrothermal fluid extraction
Transesterification	• Acid catalyst • Alkali catalyst • Lipases
Purification of product	• Decantation • Filtration • Distillation

al. (2012) to be 25-40°C. Transesterification of triglycerides into biodiesel is insignificantly affected by water when enzymes are involved. Enzymes in the surrounding region are, however, influenced by water (Atadashi et al. 2012). Lipase-catalyzed transesterification is essentially non-aqueous, and water plays an important role in influencing the catalytic activity and stability of lipase in this medium (Gog et al. 2012). When the enzyme is dissolved in an organic solvent, some water is usually required to maintain its activity. Glycerol, the main byproduct of transesterification, may be attenuated by adding water to the reaction mixture (Tran et al. 2012). Increasing mass transfer resistance is one of the major reasons why immobilized lipase is less active when there is an accumulation of glycerol in the reaction mixture. It has been extensively reviewed elsewhere how water affects enzymatic transesterification (Atadashi et al. 2012). A complex separation, purification, and immobilization procedure limits lipase production at a commercial scale due to its high cost. In 1979, Atkinson et al developed a technique called biomass solid particle (BSP) that immobilized whole cell biocatalysts on porous solid supports (Gog et al. 2012). The conversion of soybean oil to biodiesel by whole-cell lipases immobilized on polyurethane foam BSPs has been found to be 90%, but no experiments have been conducted on microalgae oil lipases (Ban et al. 2002). As a result of inefficient mass transfer, immobilized whole cell lipase has a slow reaction rate. It is possible to mitigate this limitation by using packed bed reactors with whole cell biocatalysts and by operating them continuously (Fukuda et al. 2009).

Advantages of using microalgae for biodiesel production

Research publications and articles have described microalgae as having many advantages over other biodiesel feedstocks. It is easy to cultivate them, they grow without much attention, they use unsuitable water, and they are easy to get nutrition from (Renaud et al. 1999; Tsukahara and Sawayama 2005; Chisti 2008; Hossain et al. 2008; Li et al. 2008; Rodolfi et al. 2009).

It takes microalgae a few days to complete an entire growth cycle through photosynthesis which converts sunlight into chemical energy (Sheehan et al. 1998). Furthermore, they grow almost anywhere, as long as they have sunlight and some simple nutrients, although specific nutrients and adequate aeration can accelerate their growth (Renaud et al. 1999; Pratoomyot et al. 2005; Aslan and Kapdan 2006).

Many types of microalgae are capable of adapting to a variety of environmental conditions. In this manner, it is possible to identify species with specific growth characteristics or adaptation to local environments, which is not possible with other current biodiesel feedstocks (such as soybean, rapeseed, sunflower and palm oil). As compared to conventional forestry, agricultural crops, and aquatic plants, they have significantly higher growth rates and productivity, and require much less land. Compared to rapeseed or soybean crops, algae biomass contains up to 49 or 132 times as much oil (w/w), for a 30% (w/w) oil content (Chisti 2007). Thus, arable soil is less competitive with other crops, including crops used by humans. Biodiesel, methane, hydrogen, and ethanol are among the renewable fuels that microalgae can be converted into. Compared to petroleum diesel, algae biodiesel emits fewer particulates, CO, hydrocarbons, and SOx while reducing particulate matter, CO, and hydrocarbon emissions. Some engine types, however, may emit more NOx (Delucchi 2003).

Microalgae can also be used for other purposes besides biofuel production. We are currently considering the following possibilities:

- A company or process can reduce its GHG emissions by removing CO_2 from industrial flue gases through algae bio-fixation (Wang et al. 2008).

- Algae uses these water contaminants as nutrients to grow after NH_4^+, NO_3, PO_4^3 are removed from the wastewater (Wang et al. 2008).

- Depending on the N:P ratio of the algae biomass, it can be used as organic fertilizer, ethanol, methane, or burned for energy cogeneration (electricity and heat) (Wang et al. 2008);

- Due to their ability to grow in harsher conditions, and their reduced requirement for nutrients, they can be grown in areas that are not suitable for agriculture, regardless of seasonal weather changes. Wastewater can also be used as a culture medium without requiring freshwater, which prevents them from competing for arable land.

- A variety of fine chemicals and bulk products can also be extracted from microalgae, depending on its species, some of which have valuable applications in different industries, including fats, polyunsaturated

Table 2 Lipid content and productivities of different microalgae species

Marine and freshwater microalgae species	Lipid content (% dry weight biomass)	Lipid productivity (mg/L/day)	Volumetric productivity of biomass (g/L/day)	Areal productivity of biomass (g/m²/day)
Ankistrodesmus sp.	24.0-31.0	-	-	11.5-17.4
Botrycoccus braunii	25.0-75.0	-	0.02	3.0
Chaetoceros muelleri	33.6	21.8	0.07	-
Chaetoceros calcitrans	14.6-16.4/39.8	17.6	0.04	-
Chlorella emersonii	25.0-63.0	10.3-50.0	0.036-0.041	0.91-0.97
Chlorella protethecoides	14.6-57.8	1214	2.00-7.70	-
Chlorella sorokiniana	19.0-22.0	44.7	0.23-1.47	-
Chlorella vulgaris	5.0-58.0	11.2-40.0	0.02-0.20	0.57-0.95
Chlorella sp.	10.0-48.0	42.1	0.02-2.5	1.61-16.47/25
Chlorella pyrenoidosa	2.0	-	2.90-3.64	72.5-130
Chlorella	18.0-57.0	18.7	-	3.50-13.90
Chlorococcum sp.	19.3	53.7	0.28	-
Crypthecodinium cohnii	20.0-51.1	-	10	-
Dunaliella salina	6.0-25.0	116.0	0.22-0.34	1.6-3.5/20-38
Dunaliella primolecta	23.1	-	0.09	14
Dunaliella tertiolecta	16.7-71.0	-	0.12	-
Dunaliella sp.	17.5-67.0	33.5	-	-
Ellipsoidion sp.	27.4	47.3	0.17	-
Euglena gracilis	14.0-20.0	-	7.70	-
Haematococcus pluvialis	25.0	-	0.05-0.06	10.2-36.4
Isochrysis galbana	7.0-40.0	-	0.32-1.60	-
Nitzschia sp.	16.0-47.0	-	-	8.8-21.6
Oocystis pusilla	10.5	-	-	40.6-45.8
Pavlova salina	30.9	49.4	0.16	-
Pavlova lutheri	35.5	4.02	0.14	-
Phaedactylum tricornitum	18.0-57.0	44.8	0.003-1.9	2.4-21
Porphyridium cruentum	9.0-18.8/60.7	34.8	0.36-1.50	25
Scenedesmus obliquus	11.0-55.0	-	0.004-0.74	-
Scenedesmus quadricauda	1.9-18.4	35.1	0.19	-
Scenedesmus sp.	19.6-21.1	40.8-53.9	0.03-0.26	2.43-13.52
Skeletpnema sp	13.3-31.8	27.3	0.09	-
Skeletonema costatum	13.5-51.3	17.4	0.08	-
Spirulina platensis	4.0-16.6	-	0.06-4.3	1.5-14.5/24-51
Spirulina maxima	4.0-9.0	-	0.21-0.25	25
Thalassiosira pseudonana	20.6	17.4	0.08	-
Tetraselmin suecica	8.5-23.0	27.0-36.4	0.12-0.32	19
Tetraselmis sp.	12.6-14.7	43.4	0.30	-
Monodus subterraneus	16.0	30.4	0.19	-
Monallanthus salina	20.0-22.0	-	0.08	12
Nannochloris sp.	20.0-56.0	60.9-76.5	0.17-0.51	-
Nannochloropsis oculata	22.7-29.7	84.0-142.0	0.37-0.48	-
Nannochloropsis sp.	12.0-53.0	37.6-90.0	0.17-1.43	1.9-5.3
Neochloris oleoabundans	29.0-65.0	90.0-134.0	-	-
Nitzschia sp.	16.0-47.0	-	-	8.8-21.6
Oocystis pusilla	10.5	-	-	40.6-45.8
Pavlova salina	30.9	49.4	0.16	-
Pavlova lutheri	35.5	40.2	0.14	-
Phaepdactylum tricornitum	18.0-57.0	44.8	0.003-1.9	2.4-21
Porphyridium cruentum	9.0-18.8/60.7	34.8	0.36-1.50	25
Scenedesmus obliquus	11.0-55.0	-	0.004-0.74	-
Scenedesuus quadricauda	1.9-18.4	35.1	0.16	-
Scenedesmus sp.	19.6-21.1	40.8-53.9	0.03-0.26	2.43-13.52
Skeletonema sp.	13.3-31.8	27.3	0.09	-
Skeletonema costatum	13.5-51.3	17.4	0.08	-
Spirulina platensis	4.0-16.6	-	0.06-4.3	1.5-14.5/24-51
Spirulina maxima	4.0-9.0	-	0.21-0.25	25

*Source: Thomas et al. 1984; Zhu and Lee 1997; Renaud et al. 1999; Illman et al. 2000; Poisson et al. 2002; Grima et al. 2003; Richmond et al. 2004; Moheimani 2005; Spolaore et al. 2006; De Morais et al. 2007; Chisti 2007; Natrah et al. 2007; Demirbaş 2008; Li et al. 2008; Eriksen 2008; Gouveia et al. 2009; Chiu et al. 2009; Rodolfi et al. 2009; Mata et al. 2010.

fatty acids, antioxidants, oils, pigments, sugars, natural dyes, bioactive compounds, and others (Raja et al. 2008).

- Microalgae can revolutionize the biotechnology field in many ways, including biofuels, pharmaceuticals, cosmetics, food additives, nutrition, aquaculture, and pollution prevention, because of their wide range of high-value biological derivatives and their potential commercial applications (Raja et al. 2008; Rosenberg et al. 2008).

Conclusion

Biofuels, bioactive medicines, and food ingredients can be obtained from microalgae, which are tiny factories. Since microalgae have many advantages as a sustainable feedstock for biodiesel production, they may be a potential source for producing third generation biodiesel; however, more research will need to be conducted to identify the best microalgae species for biodiesel production and improve their yield.

The process for producing biodiesel from microalgae should remain the same in the future, but more research is needed to understand how algal lipids, especially triglycerides, are synthesized. Due to their poor performance in many environmental impacts, other potential sources of biodiesel cannot compete with microalgae for creating realistic biodiesel production.

Conflicts of interest The authors declare no conflict of interest.

References

Adarme-Vega TC, Thomas-Hall SR, Schenk PM (2014) Towards sustainable sources for omega-3 fatty acids production. Curr Opin Biotechnol 26:14–18. https://doi.org/10.1016/j.copbio.2013.08.003

Altonen BL, Arreglado TM, Leroux O (2020) Characteristics, comorbidities and survival analysis of young adults hospitalized with COVID-19 in New York City. PloS One 15:e0243343

Amini Khoeyi Z, Seyfabadi J, Ramezanpour Z (2012) Effect of light intensity and photoperiod on biomass and fatty acid composition of the microalgae, Chlorella vulgaris. Aquac Int 20:41–49

Armenta RE, Valentine MC (2013) Single-cell oils as a source of omega-3 fatty acids: An overview of recent advances. J Am Oil Chem Soc 90:167–182. https://doi.org/10.1007/s11746-012-2154-3

Aslan S, Kapdan IK (2006) Batch kinetics of nitrogen and phosphorus removal from synthetic wastewater by algae. Ecol Eng 28:64–70

Atadashi IM, Aroua MK, Abdul Aziz AR, Sulaiman NMN (2012) The effects of water on biodiesel production and refining technologies: A review. Renew Sustain Energy Rev 16:3456–3470. https://doi.org/10.1016/j.rser.2012.03.004

Azcan N, Danisman A (2008) Microwave assisted transesterification of rapeseed oil. Fuel 87:1781–1788. https://doi.org/10.1016/j.fuel.2007.12.004

Baek SH, Shimode S, Han MS, Kikuchi T (2008) Growth of dinoflagellates, Ceratium furca and Ceratium fusus in sagami bay, Japan: the role of nutrients. Harmful Algae 7:729–739

Ban K, Hama S, Nishizuka K (2002) Repeated use of whole-cell biocatalysts immobilized within biomass support particles for biodiesel fuel production. J Mol Catal B Enzym 17:157–165

Bannenberg G, Mallon C, Edwards H, Yeadon D, Yan K, Johnson H, Ismail A (2017) Omega-3 long-chain polyunsaturated fatty acid content and oxidation state of fish oil supplements in New Zealand. Sci Rep 7:1488. https://doi.org/10.1038/s41598-017-01470-4

Barsanti L, Coltelli P, Evangelista V, Frassanito AM, Passarelli V, Gualtieri P (2008) Oddities and curiosities in the algal world. In: Algal toxins: nature, occurrence, effect and detection. Springer, pp 353–391

Bellou S, Aggelis G (2013) Biochemical activities in Chlorella sp. and Nannochloropsis salina during lipid and sugar synthesis in a lab-scale open pond simulating reactor. J Biotechnol 164:318–329. https://doi.org/10.1016/j.jbiotec.2013.01.010

Bleakley S, Hayes M (2017) Algal proteins: extraction, application, and challenges concerning production. Foods 6:33

Borowitzka MA, Moheimani NR. Sustainable biofuels from algae. Mitig Adapt Strateg Glob Change 2013;18:13–25

Bozbas K (2008) Biodiesel as an alternative motor fuel: Production and policies in the European Union. Renew Sustain Energy Rev 12:542–552. https://doi.org/10.1016/j.rser.2005.06.001

Butterwick C, Heaney SI, Talling JF (2005) Diversity in the influence of temperature on the growth rates of freshwater algae, and its ecological relevance. Freshw Biol 50(2):291–300

Brennan L, Owende P (2010) Biofuels from microalgae—a review of technologies for production, processing, and extractions of biofuels and co-products. Renew Sustain Energy Rev 14:557–577

Carvalho AP, Silva SO, Baptista JM, Malcata FX (2011) Light requirements in microalgal photobioreactors: an overview of biophotonic aspects. Appl Microbiol Biotechnol 89:1275–1288

Cerri CEP, You X, Cherubin MR, Moreira CS, Raucci GS, Castigioni BDA (2017) Assessing the greenhouse gas emissions of Brazilian soybean biodiesel production. PLoS One 12:e0176948

Chauton MS, Reitan KI, Norsker NH, Tveterås R, Kleivdal HT (2015) A techno-economic analysis of industrial production of marine microalgae as a source of EPA and DHA-rich raw material for aquafeed: Research challenges and possibilities. Aquaculture 436:95–103. https://doi.org/10.1016/j.aquaculture.2014.10.038

Cheirsilp B, Torpee S (2012) Enhanced growth and lipid production of microalgae under mixotrophic culture condition: effect of light intensity, glucose concentration and fed-batch cultivation. Bioresour Technol 110:510–516

Chisti Y (2007) Biodiesel from microalgae. Biotechnol Adv 25:294–306. https://doi.org/10.1016/j.biotechadv.2007.02.001

Chisti Y (2008) Biodiesel from microalgae beats bioethanol. Trends Biotechnol 26:126–131

Chiu SY, Kao CY, Tsai MT, Ong SC, Chen CH, Lin, CS (2009) Lipid accumulation and CO_2 utilization of *Nannochloropsis oculata* in response to CO_2 aeration. Bioresour Technol 100:833–838

Chojnacka K, Marquez-Rocha FJ (2004) Kinetic and stoichiometric relationships of the energy and carbon metabolism in the culture of microalgae. Biotechnol 3:21–34

Christopher LP, Hemanathan Kumar, Zambare VP (2014) Enzymatic biodiesel: Challenges and opportunities. Appl Energy 119:497–520. https://doi.org/10.1016/j.apenergy.2014.01.017

Cravotto G, Boffa L, Mantegna S, Perego P, Avogadro M, Cintas P (2008) Improved extraction of vegetable oils under high-intensity ultrasound and/or microwaves. Ultrason Sonochem 15:898–902

Daliry S, Hallajisani A, Mohammadi Roshandeh J, Nouri, H, Golzary A (2017) Investigation of optimal condition for *Chlorella vulgaris* microalgae growth. Glob J Environ Sci Manag 3:217–230

Darnoko D, Cheryan M (2000) Kinetics of palm oil transesterification in a batch reactor. J Am Oil Chem Soc 77:1263–1267. https://doi.org/10.1007/s11746-000-0198-y

De Jesus Raposo MF, de Morais RMSC, de Morais AMMB (2013) Health applications of bioactive compounds from marine microalgae. Life Sci 93:479–486

De Meester N, Gingold R, Rigaux A, Derycke S, Moens T (2016) Cryptic diversity and ecosystem functioning: a complex tale of differential effects on decomposition. Oecologia 182:559–571

De Morais MG, Costa JAV (2007) Carbon dioxide fixation by *Chlorella kessleri*, *C. vulgaris*, *Scenedesmus obliquus* and *Spirulina sp.* cultivated in flasks and vertical tubular photobioreactors. Biotechnol Lett 29:1349–1352

Dermoun D, Chaumont D (1992) Modelling of growth of *Porphyridium cruentum* in connection with two interdependent factors: light and temperature. Bioresour and Technol 42:113–117

De Pauw N, Morales J, Persoone G (1984) Mass culture of microalgae in aquaculture systems: Progress and constraints. Hydrobiologia 116:121–134. https://doi.org/10.1007/BF00027650

Del Campo JA, García-González M, Guerrero MG (2007) Outdoor cultivation of microalgae for carotenoid production: current state and perspectives. Appl Microbiol Biotechnol 74:1163–1174

Delucchi M (2003) A lifecycle emissions model (LEM): lifecycle emissions from transportation fuels, motor vehicles, transportation modes, electricity use, heating and cooking fuels, and materials. Available online at: www.its.ucdavis.edu/publications.html

Demirbaş A (2008) Production of biodiesel from algae oils. Energy Sources Part Recovery Util Environ Eff 31:163–168. https://doi.org/10.1080/15567030701521775

Deshmane VG, Gogate PR, Pandit AB (2009) Ultrasound-assisted synthesis of biodiesel from palm fatty acid distillate. Ind Eng Chem Res 48:7923–7927. https://doi.org/10.1021/ie800981v

Draaisma RB, Wijffels RH, Slegers P (2013) Food commodities from microalgae. Curr Opin Biotechnol 24:169–177. https://doi.org/10.1016/j.copbio.2012.09.012

Eriksen NT (2008) The technology of microalgal culturing. Biotechnol Lett 30:1525–1536. https://doi.org/10.1007/s10529-008-9740-3

Eppley RW, Sloan PR (1966) Growth rates of marine phytoplankton: correlation with light absorption by cell chlorophyll a. Physiol Plantarum 19(1):47–59

Fogg G (2001) Algal adaptation to stress—some general remarks. In: Rai LC, Gaur JP, editors, Algal adaptation to environmental stresses. Berlin: Springer p.1–19

Formighieri C, Franck F, Bassi R (2012) Regulation of the pigment optical density of an algal cell: filling the gap between photosynthetic productivity in the laboratory and in mass culture. J Biotechnol 162:115–123

Fukuda H, Kondo A, Tamalampudi S (2009) Bioenergy: Sustainable fuels from biomass by yeast and fungal whole-cell biocatalysts. Biochem Eng J 44:2–12

Gardner RD, Cooksey KE, Mus F, Macur R, Moll K, Eustance E (2012) Use of sodium bicarbonate to stimulate triacylglycerol accumulation in the chlorophyte *Scenedesmus* sp. and the diatom *Phaeodactylum tricornutum*. J Appl Phycol 24:1311–1320. https://doi.org/10.1007/s10811-011-9782-0

Gardner-Dale DA, Bradley IM, Guest JS (2017) Influence of solids residence time and carbon storage on nitrogen and phosphorus recovery by microalgae across diel cycles. Water Res 121:231–239. https://doi.org/10.1016/j.watres.2017.05.033

Gendy TS, El-Temtamy SA (2013) Commercialization potential aspects of microalgae for biofuel production: an overview. Egypt J Pet 22:43–51

Ghosh T, Paliwal C, Maurya R, Mishra S (2015) Microalgal rainbow colours for nutraceutical and pharmaceutical applications. In: Bahadur B, Venkat Rajam M, Sahijram L, Krishnamurthy KV (eds) Plant Biology and Biotechnology: Volume I: Plant diversity, organization, function and improvement. Springer india, New Delhi, pp 777–791

Gog A, Roman M, Toşa MM, Paizs C, Irimie FD (2012) Biodiesel production using enzymatic transesterification – Current state and perspectives. Renew Energy 39:10–16. https://doi.org/10.1016/j.renene.2011.08.007

Gogate PR (2008) Cavitational reactors for process intensification of chemical processing applications: A critical review. Chem Eng Process Process Intensif 47:515–527. https://doi.org/10.1016/j.cep.2007.09.014

Gogate PR, Kabadi AM (2009) A review of applications of cavitation in biochemical engineering/biotechnology. Biochem Eng J 44:60–72. https://doi.org/10.1016/j.bej.2008.10.006

Gouveia L, Oliveira AC (2009) Microalgae as a raw material for biofuels production. J Ind Microbiol Biotechnol 36:269–274

Grima EM, Belarbi EH, Fernández FA, Medina, AR, Chisti Y (2003) Recovery of microalgal biomass and metabolites: process options and economics. Biotechnol Adv 20:491–515

Haas MJ, Piazza GJ, Foglia TA (2002) Enzymatic approaches to the production of biodiesel fuels. In: Lipid biotechnology. CRC Press pp 680–694

Hamilton ML, Haslam RP, Napier JA, Sayanova O (2014) Metabolic engineering of *Phaeodactylum tricornutum* for the enhanced accumulation of omega-3 long chain polyunsaturated fatty acids. Metab Eng 22:3–9

Hamilton ML, Powers S, Napier JA, Sayanova O (2016) Heterotrophic production of omega-3 long-chain polyunsaturated fatty acids by trophically converted marine diatom *Phaeodactylum tricornutum*. Mar Drugs 14:53. https://doi.org/10.3390/md14030053

Herrero M, Mendiola JA, Plaza M, Ibañez E (2013) Screening for bioactive compounds from algae. In: Advanced biofuels and bio-products. Springer pp 833–872

Ho SH, Chen WM, Chang JS (2010) *Scenedesmus obliquus* CNW-N as a potential candidate for CO_2 mitigation and biodiesel production. Bioresour Technol 101:8725–8730

Hossain AS, Salleh A, Boyce AN, Chowdhury P, Naqiuddin M (2008) Biodiesel fuel production from algae as renewable energy. Am J Biochem Biotechnol 4:250–254

Hosseini Tafreshi A, Shariati M (2009) *Dunaliella* biotechnology: methods and applications. J Appl Microbiol 107:14–35

Hu FB, Bronner L, Willett WC, Stampfer MJ, Rexrode KM, Albert CM, Hunter D, Manson JE (2002) Fish and omega-3 fatty acid intake and risk of coronary heart disease in women. JAMA 287:1815–1821. https://doi.org/10.1001/jama.287.14.1815

Hu Q, Sommerfeld M, Jarvis E, Ghirardi M, Posewitz M, Seibert M, Darzins A (2008) Microalgal triacylglycerols as feedstocks for biofuel production: perspectives and advances. Plant J 54:621–639

Ishika T, Moheimani NR, Bahri PA. Sustainable saline microalgae co-cultivation for biofuel production: a critical review (2017). Renew Sustain Energy Rev78:356–68

Illman AM, Scragg AH, Shales SW (2000) Increase in *Chlorella* strains calorific values when grown in low nitrogen medium. Enzyme Microb Technol 27:631–635

Iso M, Chen B, Eguchi M, Kudo T, Shrestha S (2001) Production of biodiesel fuel from triglycerides and alcohol using immobilized lipase. J Mol Catal B Enzym 16:53–58

Jacob-Lopes E, Scoparo CHG, Lacerda LMCF, Franco TT (2009) Effect of light cycles (night/day) on CO_2 fixation and biomass production by microalgae in photobioreactors. Chem Eng Process Process Intensif 48:306–310

Juneja A, Ceballos RM, Murthy GS (2013) Effects of environmental factors and nutrient availability on the biochemical composition of algae for biofuels production: A Review. Energies 6:4607–4638. https://doi.org/10.3390/en6094607

Kalva A, Sivasankar T, Moholkar VS (2009) Physical mechanism of ultrasound-assisted synthesis of biodiesel. Ind Eng Chem Res 48:534–544. https://doi.org/10.1021/ie800269g

Khan MI, Lee MG, Seo HJ, Shin JH, Shin TS, Yoon YH (2016) Enhancing the feasibility of *Microcystis aeruginosa* as a feedstock for bioethanol production under the influence of various factors. BioMed Res Int. doi:10.1155/2016/4540826

Khozin-Goldberg I, Cohen Z (2006) The effect of phosphate starvation on the lipid and fatty acid composition of the fresh water eustigmatophyte monodus subterraneus. Phytochem 67:696–701. https://doi.org/10.1016/j.phytochem.2006.01.010

Kitaya Y, Azuma H, Kiyota M (2005) Effects of temperature, CO2/O2 concentrations and light intensity on cellular multiplication of microalgae, *Euglena gracilis*. Adv Space Res 35:1584–1588. https://doi.org/10.1016/j.asr.2005.03.039

Kirst G (1990) Salinity tolerance of eukaryotic marine algae. Annu Rev Plant Biol 41:21–53

Koller M, Muhr A, Braunegg G (2014) Microalgae as versatile cellular factories for valued products. Algal Res 6:52–63

Komers K, Skopal F, Stloukal R, Machek J (2002) Kinetics and mechanism of the KOH — catalyzed methanolysis of rapeseed oil for biodiesel production. Eur J Lipid Sci Technol 104:728–737

Krol M, Maxwell DP, Huner NPA (1997) Exposure of *Dunaliella salina* to low temperature mimics the high lightinduced accumulation of carotenoids and the carotenoid binding protein (Cbr). Plant Cell 38(2):213–216

Kudo I, Miyamoto M, Noiri Y, Maita Y (2000) Combined effects of temperature and iron on the growth and physiology of the marine diatom *Phaeodactylum tricornutum* (Bacillariophyceae). J Phycol 36:1096–1102

Kumudha, A, Selvakumar S, Dilshad P, Vaidyanathan G, Thakur MS, Sarada R (2015) Methylcobalamin—A form of vitamin B12 identified and characterised in *Chlorella vulgaris*. Food Chem 170:316–320

Lai JQ, Hu ZL, Wang PW, Yang Z (2012) Enzymatic production of microalgal biodiesel in ionic liquid [BMIm][PF6]. Fuel 95:329–333. https://doi.org/10.1016/j.fuel.2011.11.001

Laing I, Utting SD (1980) The influence of salinity on the production of two commercially important unicellular marine algae. Aquaculture 21:79–86

Leung DYC, Wu X, Leung MKH (2010) A review on biodiesel production using catalyzed transesterification. Appl Energy 87:1083–1095. https://doi.org/10.1016/j.apenergy.2009.10.006

Li Y, Horsman M, Wang B, Wu N, Lan, CQ (2008) Effects of nitrogen sources on cell growth and lipid accumulation of green alga *Neochloris oleoabundans*. Appl Microbiol Biotechnol 81:629–636

Liu C, Hu B, Cheng Y, Guo Y, Yao W, Qian H (2021) Carotenoids from fungi and microalgae: A review on their recent production, extraction, and developments. Bioresour Technol 337:125398

Liu ZY, Wang GC, Zhou BC (2008) Effect of iron on growth and lipid accumulation in *Chlorella vulgaris*. Bioresour Technol 99:4717–4722

Ma F, Hanna MA (1999) Biodiesel production: a review. Bioresour Technol 70:1–15. https://doi.org/10.1016/S0960-8524(99)00025-5

Macedo RVT, Alegre RM (2001) Influência do teor de nitrogênio no cultivo de *Spirulina maxima* em dois níveis de temperatura: Parte II: PRODUÇÃO DE LIPÍDIOS. Food Sci Technol 21:183–186

Mata TM, Martins AA, Caetano NS (2010) Microalgae for biodiesel production and other applications: a review. Renew Sustain Energy Rev 14:217–232

Maxwell DP, Fal S, Trick CG, Huner N (1994) Growth at low temperature mimics high-light acclimation in *Chlorella vulgaris*. Plant Physiol 105(2):535–543

Medipally SR, Yusoff FM, Banerjee S, Shariff M (2015) Microalgae as sustainable renewable energy feedstock for biofuel production. BioMed Res Int 2015

Melis A (2009) Solar energy conversion efficiencies in photosynthesis: minimizing the chlorophyll antennae to maximize efficiency. Plant Sci 177:272–280

Misra R, Guldhe A, Singh P, Rawat I, Bux F (2014) Electrochemical harvesting process for microalgae by using nonsacrificial carbon electrode: A sustainable approach for biodiesel production. Chem Eng J 255:327–333

Moheimani NR (2005) The culture of coccolithophorid algae for carbon dioxide bioremediation. Murdoch University

Mulders KJM, Weesepoel Y, Lamers PP, Vincken JP, Martens DE, Wijffels RH (2013) Growth and pigment accumulation in nutrient-depleted *Isochrysis galbana* T-ISO. J Appl Phycol 25:1421–1430

Mus F, Toussaint JP, Cooksey KE, Fields, MW, Gerlach R, Peyton BM, Carlson RP (2013) Physiological and molecular analysis of

carbon source supplementation and pH stress-induced lipid accumulation in the marine diatom *Phaeodactylum tricornutum*. Appl Microbiol Biotechnol 97:3625–3642

Natrah FMI, Yusoff FM, Shariff M, Abas F, Mariana NS (2007) Screening of malaysian indigenous microalgae for antioxidant properties and nutritional value. J Appl Phycol 19:711–718

Nisar N, Li L, Lu S, Khin NC, Pogson BJ (2015) Carotenoid metabolism in plants. Mol Plant 8:68–82

Paul Abishek M, Patel J, Prem Rajan A (2014) Algae oil: a sustainable renewable fuel of future. Biotechnol Res Int. doi:10.1155/2014/272814

Plaza M, Herrero M, Cifuentes A, Ibanez E (2009) Innovative natural functional ingredients from microalgae. J Agric Food Chem 57:7159–7170

Poisson L, Devos M, Pencreac'h G, Ergan F (2002) Nutrition benefits and current developments of polyunsaturated fatty acids from microalgae lipids. Oléagineux, Corps Gras, Lipides 9(2):92-95

Prathima Devi M, Venkata Mohan S (2012) CO2 supplementation to domestic wastewater enhances microalgae lipid accumulation under mixotrophic microenvironment: Effect of sparging period and interval. Bioresour Technol 112:116–123. https://doi.org/10.1016/j.biortech.2012.02.095

Pratoomyot J, Srivilas P, Noiraksar T (2005) Fatty acids composition of 10 microalgal species. Songklanakarin J Sci Technol 27:1179–1187

Pulz O, Gross W (2004) Valuable products from biotechnology of microalgae. Appl Microbiol Biotechnol 65:635–648. https://doi.org/10.1007/s00253-004-1647-x

Raja R, Hemaiswarya S, Kumar NA (2008) A perspective on the biotechnological potential of microalgae. Crit Rev Microbiol 34:77–88

Raven JA, Geider RJ (1988) Temperature and algal growth. New Phytol 110:441–461

Ravindran B, Gupta SK, Cho WM, Kim JK, Lee SR, Jeong KH, Choi HC (2016) Microalgae potential and multiple roles—current progress and future prospects—an overview. Sustainability 8:1215

Renaud SM, Thinh LV, Parry DL (1999) The gross chemical composition and fatty acid composition of 18 species of tropical Australian microalgae for possible use in mariculture. Aquaculture 170:147–159

Richmond A (2004) Biological principles of mass cultivation. Handb Microalgal Cult Biotechnol Appl Phycol 125–177

Rodolfi L, Chini Zittelli G, Bassi N, Padovani G, Biondi N, Bonini G, Tredici MR (2009) Microalgae for oil: Strain selection, induction of lipid synthesis and outdoor mass cultivation in a low-cost photobioreactor. Biotechnol Bioeng 102:100–112

Rosenberg JN, Oyler GA, Wilkinson L, Betenbaugh MJ (2008) A green light for engineered algae: redirecting metabolism to fuel a biotechnology revolution. Curr Opin Biotechnol 19:430–436

Sandnes JM, Klqvist T, Wenner D, Gislerød HR (2005) Combined influence of light and temperature on growth rates of *Nannochloropsis oceanica*: linking cellular responses to large-scale biomass production. J App Phycol 17(6):515–525

Santiago-Morales IS, Trujillo-Valle L, Márquez-Rocha FJ, López Hernández JF (2018) Tocopherols, phycocyanin and superoxide dismutase from microalgae as potential food antioxidants. Appl Food Biotechnol 5: 19–27

Sharma YC, Singh B, Korstad J (2011) A critical review on recent methods used for economically viable and eco-friendly development of microalgae as a potential feedstock for synthesis of biodiesel. Green Chem 13:2993–3006

Sheehan J, Dunahay T, Benemann J, Roessler P (1998) A look back at the US Department of Energy's aquatic species program: biodiesel from algae. Natl Renew Energy Lab 328

Smee DF, Bailey KW, Wong MH, O'Keefe BR, Gustafson KR, Mishin VP, Gubareva LV (2008) Treatment of influenza A (H1N1) virus infections in mice and ferrets with cyanovirin-N. Antiviral Res 80:266–271

Sousa I, Gouveia L, Batista AP, Raymundo A, Bandarra NM (2008) Microalgae in novel food products. Food Chem Res Dev 75–112

Spolaore P, Joannis-Cassan C, Duran E, Isambert A (2006) Commercial applications of microalgae. J Biosci Bioeng 101:87–96. https://doi.org/10.1263/jbb.101.87

Staehr P, Birkeland MJ (2006) Temperature acclimation of growth, photosynthesis and respiration in two mesophilic phytoplankton species. Phycol 45(6):648–656

Stojković IJ, Stamenković OS, Povrenović DS, Veljković VB (2014) Purification technologies for crude biodiesel obtained by alkali-catalyzed transesterification. Renew Sustain Energy Rev 32:1–15. https://doi.org/10.1016/j.rser.2014.01.005

Sorokin C, Krauss RW (1962) Effects of temperature and illuminance on chlorella growth uncoupled from cell division. Plant Physiol 37:37–42

Tabernero A, Martín del Valle EM, Galán MA (2012) Evaluating the industrial potential of biodiesel from a microalgae heterotrophic culture: Scale-up and economics. Biochem Eng J 63:104–115. https://doi.org/10.1016/j.bej.2011.11.006

Thomas WH, Tornabene TG, Weissman J (1984) Screening for lipid yielding microalgae: activities for 1983. Final subcontract report. Solar Energy Research Inst, Golden, CO (USA)

Tran DT, Yeh KL, Chen CL, Chang JS (2012) Enzymatic transesterification of microalgal oil from *Chlorella vulgaris* ESP-31 for biodiesel synthesis using immobilized Burkholderia lipase. Bioresour Technol 108:119–127. https://doi.org/10.1016/j.biortech.2011.12.145

Tsukahara K, Sawayama S (2005) Liquid fuel production using microalgae. J Jpn Pet Inst 48:251–259

Vicente G, Martínez M, Aracil J, Esteban A (2005) Kinetics of sunflower oil methanolysis. Ind Eng Chem Res 44:5447–5454. https://doi.org/10.1021/ie040208j

Wang B, Li Y, Wu N, Lan CQ (2008) CO_2 bio-mitigation using microalgae. Appl Microbiol Biotechnol 79:707–718

Wang ZT, Ullrich N, Joo S, Waffenschmidt S, Goodenough U (2009) Algal lipid bodies: stress induction, purification, and biochemical characterization in wild-type and starchless *Chlamydomonas reinhardtii*. Eukaryot Cell 8:1856–68

Warabi Y, Kusdiana D, Saka S (2004) Reactivity of triglycerides and fatty acids of rapeseed oil in supercritical alcohols. Bioresour Technol 91:283–287. https://doi.org/10.1016/S0960-8524(03)00202-5

Weissman JC, Goebel RP (1987) Design and analysis of microalgal open pond systems for the purpose of producing fuels: a subcontract report. Solar Energy Research Inst, Golden, CO (USA)

Wen ZY, Chen F (2003) Heterotrophic production of eicosapentaenoic acid by microalgae. Biotechnol Adv 21:273–294. https://doi.org/10.1016/S0734-9750(03)00051-X

Williams JA (2002) Keys to bioreactor selections. Chem Eng Prog 98:34–41

Wu H (2016) Effect of different light qualities on growth, pigment content, chlorophyll fluorescence, and antioxidant enzyme activity in the red alga *Pyropia haitanensis* (Bangiales, Rhodophyta). BioMed Res Int. doi:10.1155/2016/7383918

Xin L, Hong-ying H, Yu-ping Z (2011) Growth and lipid accumulation properties of a freshwater microalga *Scenedesmus* sp. under different cultivation temperature. Bioresour Technol 102:3098–3102

Yan Y, Li X, Wang G (2014) Biotechnological preparation of biodiesel and its high-valued derivatives: A review. Appl Energy 113:1614–1631. https://doi.org/10.1016/j.apenergy.2013.09.029

Ye CP, Zhang MC, Yang YF, Thirumaran G (2012) Photosynthetic performance in aquatic and terrestrial colonies of *Nostoc flagelliforme* (Cyanophyceae) under aquatic and aerial conditions. J Arid Environ 85:56–61

Yeesang C, Cheirsilp B (2011) Effect of nitrogen, salt, and iron content in the growth medium and light intensity on lipid production by microalgae isolated from freshwater sources in Thailand. Bioresour Technol 102:3034–40

Yongmanitchai W, Ward OP (1991) Growth of and omega-3 fatty acid production by *Phaeodactylum tricornutum* under different culture conditions. Appl Environ Microbiol 57:419–425. https://doi.org/10.1128/aem.57.2.419-425.1991

Zappe H, Snell ME, Bossard MJ (2008) PEGylation of cyanovirin–N, an entry inhibitor of HIV. Adv Drug Deliv Rev 60:79–87

Zhu CJ, Lee YK (1997) Determination of biomass dry weight of marine microalgae. J Appl Phycol 9:189–194. https://doi.org/10.1023/A:1007914806640

Publisher's Note