7 Aquatic Microbial Oxygenic Phototrophs: A Short Treatise on Diverse Applications and the Future Biofuel Scenario

Mayur Mausoom Phukan, Rupesh Kumar, Kuldeep Gupta, Pritam Bardhan, Nilutpal Bhuyan, Lina Gogoi, Plaban Bora, Manabendra Mandal, and Rupam Kataki

Abstract

Man has relied upon microalgae ever since millennia. The importance of microalgal biotechnology as an exclusive niche in the industrial state of affairs is undeniably indisputable. Microalgae have been used to produce a wide variety of high value exploitable commercial products/metabolites such as antioxidants, carotenoids, vitamins, biomolecules (carbohydrates, proteins, and lipids), etc. Microalgae also hold great promise for the forthcoming biofuel industry. Microalgal biofuel are poised to be sustainable alternatives to conventional petro fuels; however, they need to overcome certain copious obstacles in order to compete in the international fuel market for an extensive commercial deployment. The scientific community is actively involved in research to establish microalgae as a biofuel podium. Progress made in this field is noteworthy, however, scientifically demanding and intellectually rigorous research seems to be the need of the hour. This article emphasizes on the non-energy and energy prospects of microalgal biomass with additional focus on the research gaps. This article aims to disseminate first-hand state-of-the-art information to help researchers, technocrats, venture capitalists, and policy makers in their futuristic endeavors pertaining to microalgal biotechnology.

M. M. Phukan
Department of Forest Science, Nagaland University, Lumami, Nagaland, India

R. Kumar
Department of Biotechnology, Royal Global University, Guwahati, Assam, India

K. Gupta · P. Bardhan · M. Mandal
Department of Molecular Biology and Biotechnology, Tezpur University, Tezpur, Assam, India

N. Bhuyan · L. Gogoi · R. Kataki (✉)
Department of Energy, Tezpur University, Tezpur, Assam, India

P. Bora
Department of Energy Engineering, Assam Science & Technology University, Guwahati, Assam, India

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7.1 Introduction

Today's world is confronted with a global energy crisis. Adding to this energy crisis is the planetary emergency of global warming resultant from extensive use of fossil fuel usage. Declining geological reservoirs and the allied environmental issues have put the whole world at stake, whereby warranting global efforts for exploring clean and carbon neutral energy sources. The answer may lie well in our past, i.e. reliance on biomass as the source of energy. The biomass sources are renewable in nature, have a wider geographical distribution, and are environmentally benign. But the real question remains, which biomass we require to satisfy the escalating energy demand? Although time and again different researchers have rolled up the dice in favor of plant derived biomass, microbial biomass may have their own story of credibility. Aquatic Microbial Oxygenic Phototrophs (AMOPs) generally refer to algae, cyanobacteria, and diatoms. Among all these organisms algae (from here on microalgae for specificity) have been a topic of intense scientific focus for issues pertaining to energy crisis, energy security, and sustainable development.

Microalgae have garnered the attention of the scientific community as a biofuel feedstock (mostly biodiesel, bio-ethanol, pyrolytic bio-oil, and bio-hydrogen). They are promising biomass species that can serve as feedstock for the forthcoming biofuel industry. These wondrous microorganisms have been extensively investigated owing to their numerous salient attributes in comparison to terrestrial energy crops. Some of the salient prominent features of microalgae in this regard are:

1. The microalgal lipid content can easily be manipulated/adjusted by altering the respective growth media composition (Meher et al. 2006).
2. They rely exclusively on atmospheric carbon dioxide as the carbon source for their growth (Schenk et al. 2008).
3. The microalgal biomass doubling time during logarithmic phase may be normally as little as 3.5 h (Chisti 2007).
4. It is quite feasible to culture microalgae in waste as well as salty water (Schenk et al. 2008).
5. The intrinsic oil content in many species of microalgae exceeds 80% (by weight of dry biomass) (Chisti 2007).
6. Algae generally have superior rates of oil and biomass production when compared to conventional crops. This may be assigned to their simple cellular structure (Becker 1994).
7. Biomass from microalgae can be harvested almost all throughout the year (in batches). This ensures a consistent and incessant supply of oil (Schenk et al. 2008).
8. Various species of microalgae have been reported to produce different types of lipids, various complex oils and hydrocarbons (Metzger and Largeau 2005), which is much conducive in various biomass conversion processes.
9. It is possible to combine algal biofuel production with flue gas carbon dioxide alleviation, treatment of waste water, and subsequent production of high value bio-actives (Demirbas 2010).

10. Algae have been reported to produce 30–100 times more energy in a given area (per hectare) in comparison to terrestrial energy crops (Demirbas 2010).

The concept of biofuel production from microalgae is not new (Chisti 1980) but presently is being followed up critically because of soaring petro fuel prices and the planetary emergency of global warming associated with fossil fuel burning (Gavrilescu and Chisti 2005). The Arab embargo of the 1970s gave a new thrust to the field of microalgal biofuel. The Department of Energy's Office of Fuels Development (United States of America) launched the historic Aquatic Species Program (ASP) in 1978. This research project investigated biodiesel production from oleaginous algal isolates which were grown in ponds, and utilized waste carbon dioxide from coal fired power plants (Sheehan et al. 1998). The ASP was later on discontinued in 1996 owing to budget curbing, but however the status report of ASP serves as an excellent blueprint for initiating research in microalgal bioenergy. Despite the ASP out of the scenario research continued in this domain and now is well evident with tons of scientific literatures being available in the scientific repositories. There are substantial scientific efforts underway worldwide to investigate the probability of renewable biofuel production from various species of microalgae. The most important in this regard are methane (here in algal biomass is subjected to anaerobic digestion) (Spolaore et al. 2006), microalgal oil derived biodiesel (Chisti 2007), photo-biologically produced bio-hydrogen (Fedorov et al. 2005), and pyrolytic bio-oil produced by thermo-chemical conversion (Pan et al. 2010). Biofuel production from microalgal biomass is possible both in theory and practice and considering the present energy scenario is of global importance. Biofuel from microalgae indisputably appears to be promising in context of the existent energy shortage scenario; however, a critical impediment to their successful commercial implementation is the reasonably cheaper rates of petro fuels.

7.2 Non-energy Based Prospects

Apart from energy based products microalgae have other potential role such as in food additive, nutraceutical, biomedical domain, etc. Figure 7.1 shows the potential use of microalgae other than its role in energy based products. Microalgae are well-known rich source of high value bio-actives such as carotenoids, antioxidants, proteins, etc., which enhances the nutritional value of the food supplements (Hudek et al. 2014). Lutein which is a predominant carotenoid is found in many microalgae like *Muriellopsis* sp., *Scenedesmus almeriensis, Chlorella* sp., etc., and has a very high nutraceutical value (Guedes et al. 2011). Apart from Lutein, Astaxanthin and β-carotene are other carotenoids which have the potential nutraceutical values. Many microalgae like *Chlamydomonas*, *Chlorella*, *Oscillatoria*, *Scenedesmu*s, *Micractinium*, *Dunaliella, Spirulina*, and *Euglena* are well known for their high protein content which may be up to 50% of their dry weight (Islam et al. 2017). In Human muscle proteins; lysine, leucine, isoleucine, and valine amino acids are predominantly present, which account for nearly 35% of all amino acids.

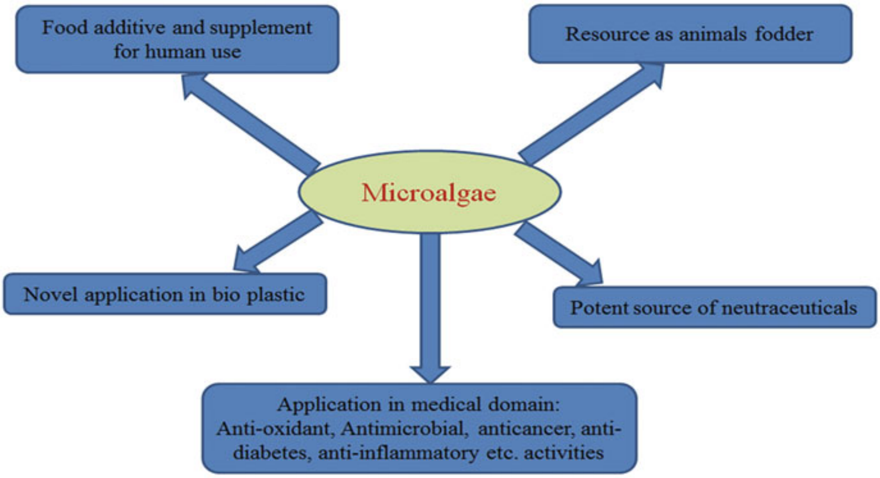

Fig. 7.1 Flowchart depicting non-energy based prospects from microalgae

Microalgae contain high amount of these amino acids. Thus they have proven application as dietary supplements to fulfill the requirement of proteins for all age groups (Dewapriya and Kim 2014). Minerals such as phosphorus, sodium, zinc, potassium, magnesium, manganese, iron, and calcium are very important for growth and nutrition in case of human and animals. Microalgae contain high amount of these minerals (Tokuşoglu 2003; Fabregas and Herrero 1990) which suggests its potential role as a nutraceutical agent. Several compounds isolated form microalgae have been reported for their potential use in biomedical domain such as naviculana polysaccharide isolated from *Navicula directa* reported as antiviral agent (Lee et al. 2006), compounds from *Phaeodactylum tricornutum* to induce leukemia cell death (Prestegard et al. 2009), marennine from *Haslea ostrearia* as antimicrobial agent (Gastineau et al. 2014), fucoxanthin and some microalgae as anti-inflammatory, anticancer, antidiabetic, and antimalarial activities (Peng et al. 2011; Lauritano et al. 2016), *Halidrys siliquosa* as anti-biofilm agent (Busetti et al. 2015). Several microalgal genera such as *Arthrospira, Chlorella, Dunaliella*, and *Haematococcus* are used as animal fodder which increases the production and quality of meat from livestock species such as pigs, ruminants, rabbits, and poultry (Madeira et al. 2017). Apart from all these biological activities, microalgae like *Chlorella, Spirulina, Nannochloropsis, Botryococcus braunii, Nannochloropsis gaditana* have been used to synthesize the bio plastics (Rahman and Miller 2017).

7.2.1 Microalgae from the Biotechnology Perspective

Microalgae are commercially used for human nutrition (*Chlorella*, *Spirulina*, *Dunaliella*) (Sathasivam et al. 2019), source of vitamins as dietary supplements (vitamin B_{12}, K_1) (Grossman 2016; Tarento et al. 2018), animal and aquatic feed

(Madeira et al. 2017; D'Este et al. 2017). A wide range of pigment molecules like carotenoids, astaxanthin which are antioxidants are produced by microalgae (Rammuni et al. 2019), polyunsaturated fatty acids (PUFAs), antimicrobial and anti-carcinogenic compounds (Kumar et al. 2019; Marrez et al. 2019), storage lipids or triacylglycerides (Xin et al. 2019), proteins, carbohydrates, and amino acids (Rizwan et al. 2018). Microalgae have garnered recent research attention particularly for bio-refinery (i.e., sustainable generation of biofuels along with high value metabolic co-products products by consolidated bio processing). In this context, oleaginous microalgae like *Nannochloropsis*, *Schizochytrium*, and *Botryococcus* (with oil content ranging from 20% to 60%, and up to 80%) have been widely explored for the biodiesel production (Bardhan et al. 2019) and other liquid biofuels (de Morais et al. 2019) (Table 7.1).

Genetic engineering tools have been used to modify microalgae for recombinant protein production, express genes to synthesize novel products, and increase the yield of natural value added products (Gangl et al. 2015). For example, *Chlamydomonas reinhardtii* was genetically engineered to produce xylitol (finds application in the food and confectionary industry as artificial sweetener) by integrating a xylose reductase gene from *Neurospora crassa* into its chloroplast genome (Pourmir et al. 2013). Furthermore, recombinant microalgae are promising cell factories for therapeutic protein production including antibodies, vaccines, and hormones (Gong et al. 2011). Recently, *Schizochytrium* sp. was genetically modified to produce a new antiviral vaccine against zika virus (Márquez-Escobar et al. 2018).

7.2.2 Microalgae from the Environmental Microbiology Perspective (Fig. 7.2)

Discharge of industrial effluent, municipal solid waste, agro-industrial waste water, pharmaceutical contaminants into freshwater systems has led to serious health issues and associated environmental hazards. In this context, microalgae mediated wastewater treatment and bioremediation of polluted contaminants has garnered recent research attention as it is solar-power driven, economically comprehensive, and sustainable strategy to mitigate these issues (Xiong et al. 2018). de Souza Leite et al. (2019) reported more than 90% removal of organic matter (in municipal and piggery wastewater) using *Chlorella sorokiniana.* Another study demonstrated the potential of using microalgae–bacteria consortium (two microalgal species, viz. *Desmodesmus* spp. and *Scenedesmus obliquus*) for the treatment of leachate/wastewater mixture along with microalgal biomass production having enhanced amount of accumulated lipids for the production of biodiesel (Hernández-García et al. 2019).

In addition to wastewater treatment, microalgae find immense application in soil as bio-fertilizers for nitrogen recovery (de Souza et al. 2019). Khan et al. 2019 demonstrated bio-refinery approach by integrating the phycoremediation potential of *Chlorella minutissima* with the subsequent production of biodiesel and organic manure. In terms of CO_2 capture process, microalgae has been found to have better (10–50 times more) CO_2 fixation ability than plants (Yadav and Sen 2017).

Table 7.1 Use of different microalgae in biotechnological application

Microalgae	Product/benefits	Application	References
Anabaena cylindrica	Vitamin K_1	Human nutrition	Tarento et al. (2018)
Spirulina platensis	Anti-inflammatory, antioxidant, and antihepatotoxic effects	Pharmaceutical, nutrition	Al-Qahtani and Binobead (2019)
Poterioochromonas malhamensis	Bio-active compounds (anti-bacterial)	Pharmaceutical	Schuelter et al. (2019)
Nannochloropsis oceanica	Biodiesel and aquaculture feed	Biofuels, shrimp feed	Ashour et al. (2019)
Thraustochytrium striatum	Astaxanthin	Nutraceuticals, cosmetics	Xiao et al. (2019)
Monoraphidium sp.	α-Linolenic acid	Infant formulas and nutritional supplements	Lin et al. (2018)
Tetradesmus obliquus	Galactooligosaccharides/β-galactosidase	Prebiotics/enzymes	Suwal et al. (2019)
Dictyosphaerium chlorelloides	Biopolymer (proteoglycan)	Food industry	Halaj et al. (2018)
Porphyridium cruentum	β-phycoerythrin	Fluorescent biomarker for diagnostics	Tran et al. (2019)
Chlorella pyrenoidosa, Scenedesmus sp., *Chlorococcum* sp.	Exopolysaccharides/antioxidant, antitumor activity	Pharmaceutical	Zhang et al. (2019)

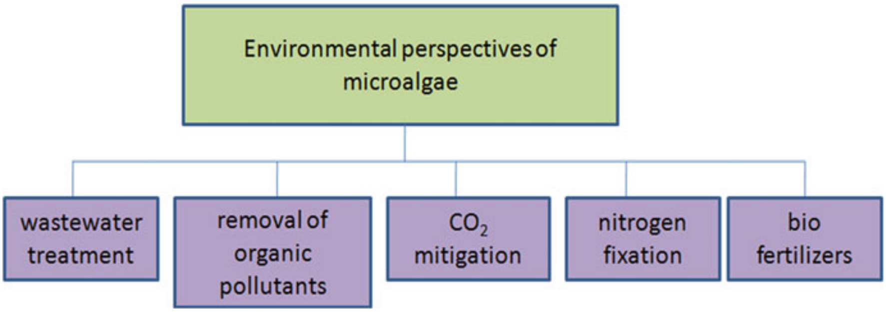

Fig. 7.2 Flowchart depicting different environmental perspective of microalgae

Table 7.2 Biotechnological applications of bio-active compounds from algae (Sharma and Sharma 2017; Priyadarshani and Biswajit 2012)

Sl. no.	Algal species	Different compounds	Uses
1	*Spirulina platensis*	Phycocyanins	Nutraceuticals, cosmetics
2	*Chlorella vulgaris*	Ascorbic acid	Health food, food supplement, food surrogate
3	*Haematococcus pluvialis*	Carotenoids, astaxanthin	Nutraceuticals, pharmaceuticals, additives
4	*Odontella aurita*	Fatty acids	Pharmaceuticals, cosmetics, baby food
5	*Porphyridium cruentum*	Polysaccharides	Pharmaceuticals, cosmetics
6	*Dunaliella salina*	Carotenoids	Nutraceuticals, food supplement, feed

Scenedesmus and *Neochloris* microalgal strains were found to be the most efficient as compared to different strains of microalgae or cyanobacteria for CO_2 capture process development (Sepulveda et al. 2019).

7.2.3 A Treatise on Commercial Applications of Microalgae

Presently, microalgae have received global interest owing to their prospective application in the renewable energy, nutraceutical, and biopharmaceutical sectors (Khan et al. 2018) (Table 7.2).

Microalgae have three essential features that can be capitalized for commercial and technical benefits (Priyadarshani and Biswajit 2012):

1. Microalgae are a highly diverse group from the genetical standpoint with an extensive array of biochemical and physiological characteristics; accordingly they are known to produce different altered and unusual fats, high value bio-active compounds, sugars, etc. (Richard and Bruce 1994).

Table 7.3 Useful substances present in microalgae (Priyadarshani and Biswajit 2012)

Pigments/carotenoids	β-carotene, astaxanthin, lutein, zeaxanthin, canthaxanthin, chlorophyll, phycocyanin, phycoerythrin, fucoxanthin
Antioxidants	Catalases, polyphenols, superoxide dismutase, tocopherols
Polyunsaturated fatty acids (PUFAs)	DHA(C22:6), EPA(C20:5), ARA(C20:4), GAL(C18:3)
Vitamins	A, B_1, B_6, B_{12}, C, E, biotin, riboflavin, nicotinic acid, pantothenate, folic acid
Other	Antimicrobial, antifungal, antiviral agents, toxins, amino acids, proteins, sterols, MAAs for light protection

2. They can cost-effectively assimilate the stable isotopes ^{13}C, ^{15}N, and ^{2}H into their biomass, and thereby into plentiful compounds they produce (Richard and Bruce 1994).
3. They represent a large, uncultivated group of organisms, and thus provide an almost accessible source of many valuable products (Richard and Bruce 1994).

Microalgae can be used as a prospective source of single-cell protein (SCP) (Priyadarshani and Biswajit 2012), a means of carbon sequestration (carbon dioxide) from stack gas, and can be utilized in sewage purification and for the production of biofuel. Microalgae have been projected as sunlight driven living-cell factories for biofuel production and several bio-chemicals used in food, aquaculture, poultry, and pharmaceutical establishments due to presence of different useful compounds (presented in Table 7.3).

7.2.4 Algae and Food

Global demand for algal foods (both micro and macroalgal) is escalating, and algae are increasingly being consumed for numerous practical benefits beyond the customary/traditional considerations of nourishment and health (Dawczynski et al. 2007). According to previous reports, algae are an exceedingly affluent source of vital and essential nutrients. They are a major source of human food in different nations, such as North & South America, France, Scotland, Ireland, Norway, Sweden, Germany, particularly in Asian countries like Japan, Peoples Republic of China, and Korea (Priyadarshani and Biswajit 2012; Kalau 2017). Even, microalgae are an affluent source of enzymes, protein, carbohydrates, and fiber. Additionally, numerous vitamins and minerals such as vitamin A_1 (Retinol), C (Ascorbic acid), B_1 (Thiamine), B_2 (Riboflavin), B_6 (Pyridoxine), Niacin (Nicotinic acid and a form of vitamin B_3), iodine, potassium, iron, magnesium, calcium, etc., are extensively found in microalgae (Priyadarshani and Biswajit 2012). Edible cyanobacteria (blue-green algae), such as *Spirulina*, *Nostoc*, and *Aphanizomenon* species have been used as food for millennia (Usharani et al. 2012). *Spirulina platensis*, a blue-green alga is increasingly gaining global reputation as a food supplement (being one of the most nutritious food source known to man) (Priyadarshani and Biswajit 2012).

Spirulina is an excellent source of proteins (Colla et al. 2007), pigments (Rangel-YaguiCde et al. 2004), poly unsaturated fatty acids (Sajilata et al. 2008) vitamins, and phenolics (Ogbonda et al. 2007).

7.2.5 Microalgae and Cosmetics

The cosmetic products from microalgae include mostly hair care and sunscreen products. Some typical microalgal species are well established in the skin care market such as *Mastocarpus stellatus, Chondrus crispus, Chlorella vulgaris, Ascophyllum nodosum, Dunaliella salina, Spirulina platensis, Nannochloropsis oculata*, *Alaria esculenta*, etc. (Priyadarshani and Biswajit 2012; Stolz and Obermayer 2005). The whole range of microalgal extracts can be largely found in skin and face care products (e.g., refreshing or re-generant care products, anti-aging cream, emollient, and as an anti-irritant in peelers (Stolz and Obermayer 2005). Some microalgal species are grossly utilized in companies in the skin care market. Some companies such as LVMH and Daniel Jouvance have landed in their personal microalgal cultivation systems. The various extracts from these species are an integral part of several cosmetics such as rejuvenating care products, anti-aging cream, sunscreen lotions, and hair care products (Sharma and Sharma 2017; Spolaore et al. 2006). There has been a soaring rise in procurement of various sunscreen products (which includes extracts from different species of microalgae) mainly due to increased public perception regarding skin cancer and photo-aging processes (Spolaore et al. 2006).

7.2.6 Microalgae and Food Colorant

Microalgal stains have marketable applications as aesthetic/cosmetic constituents and natural food colorants (Priyadarshani and Biswajit 2012). Various species of microalgae are known to produce an extensive array of colored pigments and consequently they are an appealing repertoire of natural colorants. Some microalgae cover substantial amounts of Carotene (besides β-carotene). β-carotene is used as a food colorant (major purpose in providing the yellow color to margarine), as a food additive to augment the color of fish flesh and eggs yolks, and to improve the well-being and productiveness/fertility of grain-fed cattle (Borowitzka and Borowitzka 1987).

7.2.7 As Bio-Fertilizers

Cyanobacteria (microalgae) have been known to play a cardinal role in maintenance and upsurge of fertility of soil, consequently enhancing crop growth (especially paddy), yield, and productivity as a natural bio-fertilizer (Song et al. 2005). From the cultivation perspective cyanobacteria in paddy cultivation is directly associated with their ability to fix atmospheric nitrogen and other positive effects for plants and soil (Malik et al. 2001). Cyanobacterial species like *Tolypothrix, Anabaena, Nostoc*, etc., are efficient fixers of atmospheric nitrogen and are mainly used as inoculants for cultivation of paddy crops (both in upland and low land conditions) (Priyadarshani and Biswajit 2012). Jochum et al. 2018 reported that the two strains of N_2-fixing cyanobacteria (*Anabaena* sp. UTEX 2576, *Nostoc muscorum* UTEX 2209S), and a polyculture of *Chlorella vulgaris* (UTEX 2714) and *Scenedesmus dimorphus* (UTEX 1237) had improved the efficacy of microalgae based fertilizers in paddy growth with the help of using vertical semi-closed airlift photo-bioreactor (PBR) (Jochum et al. 2018). *Anabaena* in conjugation with the water fern *Azolla* contributes nitrogen (approximately 60 kg/ha/season) and also enhances soil quality with organic substances (Priyadarshani and Biswajit 2012).

7.3 Energy Prospects from Microalgae with Special Reference to Bioenergy

Speedily dwindling geological reservoirs, swelling energy demands, and increasing global concerns about the environment have compelled mankind to search for petro fuel substitutes. In this regard, numerous plant species have been investigated as potential source of biofuel; however, several lacunas associated with terrestrial energy crops/oil crops and lignocellulosic biofuel hinder their progression and popularity. On the global forefront, microalgae have been projected as a prime bioenergy source with the potential to replace conventional petro fuels (Chisti 2007). As discussed earlier in the introduction section, algae offer several advantages as compared to oil crops/energy crops and consequently have been extensively researched as a replacement for the predominant biofuel sources like sugarcane and corn. Several developed nations, emerging economies, and reputed companies like ExxonMobil, Sapphire Energy, Algenol, Solazyme, etc., have been already working towards the concept to commercialization of microalgal biofuel.

7.3.1 Biodiesel from Microalgae

Conventionally biofuel is mostly produced from plant oils such as corn, canola, soybean, rape seed, palm oil, *Jatropha*, *Pongamia*, coconut, ground nut, sunflower, mustard, etc. But none of these feedstocks can even pragmatically satisfy even a fraction of the present burgeoning need for energy (liquid biofuel). Furthermore, the conundrum of the Food Vs Fuel debate has geared up the quest for newer, sustainable, cost efficient, and environmentally benign feedstock for biodiesel (fatty acid methyl esters) production. A possible exception that may roll up the dice in favor of sustainability in near future is biodiesel production from microalgae. Over the past few decades microalgae have been the center of bioenergy research consideration. Microalgae today lie in the vanguard of bioenergy research as an emerging and promising feedstock for biodiesel production. Microalgal biodiesel research is now one of the top notch research topics especially in the context of escalating petro fuel prices and climatic changes. Microalgae are characterized by an exceptionally speedy growth rate in comparison to plants/energy crops and additionally a significant proportion of their weight comprises oil. The microalgal oil following suitable extraction procedures can be reacted with an alcohol to get biodiesel which is renewable and environmental benign in nature. Also from the theoretical perspective, microalgae present strong candidature as a viable bioenergy feedstock for biodiesel production. The yield of oil from microalgae (on per unit area basis) is predicted to be 20,000–80,000 L/acre/year (Demirbas and Demirbas 2010). The theoretical yield is 7–31 times higher than palm oil (the next best crop for the production of biodiesel) (Demirbas and Demirbas 2010). Microalgae have been hypothesized to be the sole source of renewable biodiesel competent of meeting the international demand for liquid transportation fuels (Chisti 2007).

7.3.2 Bio-Oil from Microalgae

Biodiesel production from microalgae involves lipid extraction followed by transesterification. Following lipid extraction the microalgal de-oiled cake/remnants (low value biomasses refuse devoid of oil) are left. Finding suitable prospects for the microalgal de-oiled cake is one of the utmost challenges for the forthcoming microalgal bio-refineries (Ferrell and Sarisky-Reed 2010). Previously the microalgal de-oiled cakes were used as an aquaculture feed. But now with the energy crisis hitting the block, scientists are more concerned in finding suitable energy based options from the de-oiled cakes. One feasible alternative in this regard which would also be influential in reducing the economics of feedstock utility is pyrolysis of microalgal remnants to obtain renewable bio-oil and other value added products (bio-char and syn-gas). A few studies done in this direction by Pan et al. 2010, Wang et al. 2013 warrant the feasibility of pyrolytic bio-oil production from microalgal remnants.

In the wake of recent advancements in bioenergy research pyrolytic bio-oils have already gathered the attention of the scientific community in that they offer potential candidature not only as a chemical feedstock but also as a progressively attractive fuel option. However, it is interesting to note that there is dearth of scientific information about pyrolysis of direct microalgal biomass or its remnants in comparison to the pyrolysis of lignocellulosic biomass. A few studies have been conducted in this regard (Du et al. 2011; Miao et al. 2004; Miao and Wu 2004). Researchers have suggested that thermo-chemical conversion of de-oiled cake via pyrolysis can produce bio-oil which in some admiration is superior to bio-oil obtainable via pyrolysis of lignocellulosic biomass (Du et al. 2011, Miao et al. 2004, Miao and Wu 2004).

A major lacuna of microalgal biomass as a pyrolysis feedstock is the elevated nitrogen content in the bio-oil product. As per previous studies (Becker 2006) most of this nitrogen is present as protein in fast growing autotrophic microalgae. Additional nitrogenous ingredients of microalgae comprise nucleic acids (DNA and

RNA), chlorophyll, glucosamides, and cell wall materials though at reasonably low levels (<0.6 wt%) when compared to protein (10 wt%) (Devi et al. 1981; Becker 2006).

7.3.3 Bio-Ethanol from Microalgae

Bio-ethanol production from microalgae is not a new concept (although to a lesser extent in contrast to microalgal biodiesel). Currently, there have been incredible surges in research and development efforts to investigate the deployment of microalgae as a superior bioenergy feedstock for bio-ethanol production processes (Subhadra and Edwards 2010). The global interest in microalgae as a bio-ethanol feedstock is because they do not require arable land for cultivation and consequently do not contribute to the Food Vs Fuel debate. On to the contrary, existing bio-ethanol crops such as sugarcane, soybean, and corn contribute to the challenge of the Food Vs Fuel dispute.

Microalgae have been reported to store substantial fraction of carbohydrates in the form of glycogen, starch/cellulose, pentoses, and hexoses which can be converted into fermentable sugars via fermentation for bio-ethanol production (Wayman 1996). Bio-ethanol can be produced from microalgal biomass by utilizing amylolytic biocatalysts which facilitate starch hydrolysis and successive formation of fermentable sugars. Following fermentation of these sugars they can be distilled by means of distillation technology to obtain anhydrous bio-ethanol.

Although there is paucity of scientific literature with regard to bio-ethanol production from microalgae the process offers certain distinct advantages. Algal fermentation processes involve less energy intake and the procedure is much simple in contrast to production scheme for biodiesel (Singh and Gu 2010). Furthermore, carbon dioxide released by fermentation process can be recycled as a carbon supply for microalgae cultivation, thereby reducing greenhouse gas emissions (Singh and Gu 2010).

In today's scenario much of the scientific attention in microalgal biofuel research is concentrated in the biodiesel production from various species of oleaginous microalgae. A major production scheme for biodiesel production from oleaginous microalgae would generate enormous quantum of microalgal remnants/de-oiled cakes. Conversion of these microalgal remnants (chiefly comprising carbohydrates and proteins) into bio-ethanol may be a lucrative alternative in this regard. However, production of bio-ethanol from microalgae is at its infantile stage and warrants additional scientific investigations.

7.4 Research Gaps in Microalgal Biotechnology

Research gap literally exists in all the aspects of microalgal biotechnology. Microalgal biotechnology must enable us in getting products superior in quality, economically competitive (low at cost), and massive in scale. This is achievable only if the existent research gaps are addressed properly. Research gaps need to be addressed for cost cutting in every step of the supply chain and commercially realize the full potential of microalgal biofuel and associated value added products. An integrated bio-refinery based approach is always preferable, where in the microalgal feedstock can be converted to biofuel in conjugation with a spectrum of other valuable products. The idea of zero waste bio-refineries is very popular in modern times, where in any waste generated in the production scheme automatically becomes the feedstock for the next product. Although significant progress has been achieved in the field of microalgal biotechnology, much more strenuous research is warranted to realize economically competitive algal biofuel and other value added products. Many subareas such as microalgal biology, strain improvement, microalgal cultivation systems (mostly mass culture approaches), process optimization, microalgal harvest and dewatering, new extraction technologies, genetically engineered strains (GMOs), biomass conversion, product recovery, co-product generation, value addition, fuel processing, economic analysis, etc., still remain the cardinal check points. However, addressing all these is beyond the scope of the present article. This article will consider only the life cycle assessment part. Although microalgae based biofuel production is emerging out as a top notch domain of research, there are many challenges and technical obstacles for massive commercialization of the fuel processing technologies. Microalgal biofuel production involves complicated cultivation, harvesting, dewatering, oil extraction, conversion, and purification steps. Adoption of suitable and efficient harvesting, extraction, or conversion technology is very crucial for an overall sustainable process economics (Shi et al. 2019).

Several methodologies for assessing environmental impacts and better sustainability for chemical process industry have been followed in recent years. Life cycle assessment (LCA) study among these is viewed as of paramount importance and imperative for microalgae based biofuel productions (Shi et al. 2019; Wang et al. 2011; Lardon et al. 2009). However, these analyses also give different results as biofuel production from microalgae largely relies on assumptions pertaining to algae cultivation methods, biomass yield and lipid content parameters, oil extraction methods and post treatment processes related to purification and upgradation (if any). For example, dewatering has been proposed as the most energy consuming and GHG emitting steps in an overall LCA of algae biodiesel production (Sander and Murthy 2010).

Another LCA results reported that flocculation had the lowest impact among three algae harvesting options, namely centrifugation, filtration, and flocculation/settling. The results of the study also showed chitosan and hexane as the most suitable, energy intensive flocculant and solvent, respectively (Brentner et al. 2011).

In a recent finding, “after modeling 160 pathways for combinations of different technologies of each process stage, the overall best-case scenario for well-to-wheel study was found to be flat-plate photo-bioreactor cultivation, chitosan flocculation, supercritical methanol combined extraction and transesterification, and energy recycle through recovery of biogas, from a LCA perspective, and total life cycle GHG emission amounts to 8.05 g CO_2 eq per MJ of biodiesel (Shi et al. 2019).” Although, it is quite complicated to evaluate these manifold LCA results on harvesting and extraction unit procedures and subsequently resolve which technology leads to preferable environmental performance (Shi et al. 2019; Wang et al. 2011; Lardon et al. 2009; Sander and Murthy 2010; Brentner et al. 2011).

7.5 Concluding Remarks and Future Projections

The preceding decades have witnessed a major upsurge in microalgal biotechnology based research. Specific contributions to this pool of scientific knowledge are well evident with numerous scientific publications and patents from all around the globe. On the global vanguard there are numerous algal firms which produce and harvest algal biomass for varied applications (biofuels and value added products). Additionally, the number of startup companies attempting for commercial deployment of algal based biofuel is constantly in the rise. Algal biofuel undeniably offers a win–win situation, but still strenuous research is required for successful all round commercial deployment. A zero waste integrated microalgal bio-refinery based approach (where any waste generated becomes the feedstock for the next product) is likely to improvise the economic viability of algal biofuel. On a serious note much work needs to be realized in specific domains such as genetic and metabolic engineering of microalgal strains (strain improvement), growth and process optimization, microalgal biomass productivity, harvesting, bioreactor designing, enzymatic hydrolysis, lipid modulation, energy recycling, in expensive mass culture approaches, efficient and modern biomass conversion technologies, downstream processing, value addition (especially by-products), etc., for an enhanced inclusive understanding. Although, concept to commercialization of algal biofuel is only a matter of time, in the ensuing future algae is likely to play a paramount role in the international road transportation fuel mix. Nevertheless, it would also be interesting to see how algal biofuel would compete with new kids like hybrid cars in the near future where much progress has been made in the battery and energy efficiency technologies.

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