COBEM 2021 Florianópolis - Brasil
ABCM Associação Brasileira de Engenharia e Ciências Mecânicas
26th ABCM International Congress of Mechanical Engineering
November 22-26, 2021. Florianópolis, SC, Brazil

COB-2021-0548
MICROALGAE AS SOURCE OF RENEWABLE ENERGY: A REVIEW

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Abstract. *Microalgae are photosynthetic single-celled microorganisms that have instigated interest in many kinds of scientific research, mainly because of their versatility in several applications. They are capable of promoting the generation of electric energy, by converting the thermal energy released during incineration, via, for example, the Rankine Cycle. They are used as nutritional additives in fish and pet food, or even in nutritional bars for human consumption. Microalgae can act as agents for bioremediation and mitigation of pollutants, through the treatment of effluents, reducing the amount of greenhouse gas emissions, and also can be a solution for a more adequate disposal of agro-industrial waste. However, one of the biggest scenarios for contemplating the applications of microalgae is the context of renewable energy, mostly in the composition of biofuels, such as ethanol, biodiesel, biobutanol, biomethane, biohydrogen, and even syngas. Energy conversion from sustainable sources has increasingly attracted attention, for example, the United Nations (UN), in 2015, proposed the "2030 Agenda for Sustainable Development" as a priority to be achieved, from the 169 goals, that make up part of the 17 Sustainable Development Goals (SDGs), also known as the Global Goals. Within these objectives, those that chiefly contemplate the themes of accessible and clean energy, sustainable cities and communities, responsible consumption and production, and combating climate change stand out. Furthermore, in 2020 Brazil launched the "2050 National Energy Plan" (PNE), whose objective is the long-term planning of the country's energy sector, working specially with global energy generation trends aiming to apply them domestically before 2050. From these, microalgae represent a possible path to new sustainable energy resources, since they meet the criteria for biofuel generation. This work presents a general overview of the research related to the market for biofuels derived from microalgae from the cultivation process until the biochemical synthesis routes, including different means through which biomass may be processed according to the final intended product, and how much it can affect the green economy, aiming to impact the global market.*

Keywords: *biofuels; microalgae; renewable energy; renewable resources; sustainability.*

1. Introduction

Microalgae are single-celled microorganisms responsible for most of the oxygen production of the world (Chapman, 2013). They can grow in inhospitable conditions, since they have different characteristics that vary from one biological species to the other (Furlan *et al.,* 2020). The advantage of using photobioreactors (PBRs) as a way of growing microalgae has received much attention from laboratories and industries in recent years. That is mainly due to a lucrative interest, as they possess high-added-value bioproducts, causing their rise in popularity in the scientific community, and the promotion of research thereon (Jacob-Lopes, 2009).

Microalgae can be used for various purposes that can contribute to the sustainability scenario, for example, when applied in the production of biopolymers, they provide extremely fast bioplastics in the recyclable chain, as they are extremely biodegradable (Onem Cinar, 2020); they act directly in environmental bioremediation, purifying gases present in the polluted atmosphere, such as SO_x, NO_x, and even CO_x, synthesizing sulfates, nitrates and carbonates in their food, and releasing the O_2 in a clean and pure way into the environment, in addition to consuming phosphates, which may also be present suspended in the environment.

Some of them, such as *Haematococcus Pluvialis*, when subjected to continuous stress due to the absence of any basic condition for their survival, turn into a characteristic red color, which when extracted becomes a carotenoid which is used in food dyes or as a powerful antioxidant, popularly known as astaxanthin (Vechio, 2021). However, the greatest current interest in the production of microalgae is for their use in biofuels, in order to reduce the emission load of pollutants generated by fossil fuels.

In the production of microalgae, carbohydrates, proteins and lipids are generated. The extraction of carbohydrates may produce fermentable sugars, thus generating bioethanol and even methane, through fermentation. Meanwhile, the extraction of fatty oils, derived from lipids, through a transesterification, generates biodiesel and biokerosene. The byproducts generated by the biodiesel production chain, after gasification, result in the production of syngas, which can also generate biohydrogen when refined. There are also other means of biohydrogen production, which do not involve the consumption of biomass, but rather are a natural process of microalgae cultivation, which are photohydrolysis, or water photolysis.

2. Microalgae culture conditions

Microalgae are considered unicellular microorganisms of rapid reproduction and can be biologically characterized as autotrophic or heterotrophic, with the ability to develop in adverse environments, including saline or brackish aquatic environments (Furlan *et al.*, 2020). They can be cultivated in different ways, from open tanks - (lakes and ponds) -, as found in nature, as well as in an industrial way, through the use of photobioreactors (Harun *et al.,* 2010). Microalgae are responsible for most of the photosynthesis on our planet, as they are found in vast areas in the environment (Lehmuskero, 2018), besides that, they have the ability to promote environmental bioremediation (Schmitz, 2015), cleaning the air atmospheric, through the photosynthesis reaction, which promotes, in addition to its growth, the oxygenation of the environment (Derner *et al.*, 2006). For their appropriate growth, they require, as their food, a source of organic carbon, solar energy, and inorganic sources such as nitrates and phosphates, which are present in the media in which they are immersed (Borowitzka, 2020).

Microalgae cultivation can be classified into three types, namely: heterotrophic, autotrophic and mixotrophic. The photoautotrophs are dependent on light as a source of energy for the production of their nutrients. While heterotrophic cultures require providing a source of carbon within the culture medium for the microorganism's energetic development. On the other hand, when the food provided comes from inorganic, organic and metabolic sources (such as sugars, for example), in addition to adding light to the inoculum, we call the medium mixotrophic, as photosynthesis and feeding by organic sources occurs (Furlan, *et al,* 2020).

2.1 Critical factors

There are several factors that influence the cultivation of microalgae and consequently, impact the production of microalgae bioproducts. Light is an important parameter that one must consider when cultivating microalgae, it is the main source of energy for photoautotrophic cultures. In nature, the main source of light is solar radiation, but under laboratory conditions, it is replaced by fluorescent lamps. In both cases, light is converted by the microalgae into chemical energy and food sources. Light can interfere with crops in three ways: intensity, wavelength, and photoperiods. Both forms of lightning may partially shine on the culture. The strategy of having specific places for light exposure is called a semi photoautotrophic regime (Straka and Rittmann, 2018).

The next factor is temperature, directly impacting the availability and solubility of certain nutrients for the culture medium and directly influencing the growth rate of the microorganism. Furthermore, temperature can induce biochemical changes in the cellular structure and in the formation and quality of microalgal lipids (Yuan *et al*. 2020), thus affecting the generation of bioproducts, such as biofuels. It is undeniable that temperature governs chemical transformations or reactions, and even the speed of many metabolic reactions, and may also invalidate a crop, after all, certain species are comfortable to develop at specific temperatures, ranging from 25°C to 30°C, while other species develop in ice temperatures, or in arid temperatures. (Schmidell *et al.* 2001; Barten *et al.* 2020; Daneshvar *et al.* 2021).

Another factor is pH, directly important in the metabolic activity of microalgae. Linked with the compound solubility capacity in the environment, thus subjecting the bioavailability of nutrients such as carbonates, nitrates and even phosphates, in addition to the gas transfer. Under alkaline conditions, growth inhibition and ammonia volatilization can occur (Lu *et al.* 2020; Rossi *et al.* 2020). In addition, variations in pH beyond the ideal range ($6 < pH < 9$) physiological and morphological characteristics of microalgae can also be altered, influencing even the permeabilization of the cell membrane, thus allowing the passage of ions to change the internal biochemical composition of cells (Galès *et al.* 2020).

Finally, the most important factor of extreme necessity for the cultivation of microalgae is aeration, which guarantees homogeneity in the reaction mixture, promoting good exposure of the cells to light, in addition to good gas exchange and heat transfer. When this factor is properly matched, it prevents the formation of precipitate in the environment in which cultivation is taking place (Zhao *et al.* 2011; Daneshvar *et al.* 2021). In addition to the main function of supplying CO_2, aeration is responsible for promoting movement in cases where other means such as impellers and propellers are not present or impractical.

3. Microalgae Cultivation Systems

To produce biofuels microalgae biomass is needed, and to generate it there are several cultivation systems for either a pilot or industrial scale, the main ones being externally (open system), as open tanks and ponds, also called raceways ponds (Borowitzka, 1999, Schenk *et al.,* 2008, Brennan & Owende, 2010, Zeng *et al*., 2011, Huo *et al.,* 2012). The use of tanks and raceway ponds has been dated since 1920 in California and North Dakota, both in the USA (Jordão and Pessoa, 2011). Later, in the 1950s, they started to be explored for commercial purposes, which is documented in several studies and publications (Chen *et. al.,* 2018, Yin, *et. al.*, 2020, Nugroho and Zhu, 2019).

The major advantages of microalgae cultivation in raceway ponds are scalability, high production volumes, low operating and maintenance costs, and the simplicity of operation and construction (Chisti, 2007).

Additionally, there are closed systems, called closed tubular photobioreactors (PBRs), that gained popularity in the mid-1990s, to optimize, industrialize and try to compress the production of microalgae, then becoming a necessity over time (Hu *et al*., 1996,. Amaro *et al.,* 2011, Jiang, *et al.,* 2013). They can be classified according to their format and operation mode. The highlights are: PBRs with flat-plates, horizontal PBRs, and airlifts, also known as vertical tubular plates (Tredici, 2004). They can be built with materials like glass or plastic, which result in higher investment cost when compared with raceways ponds of comparable capacity and there is additional maintenance.

The PBRs started to expand and develop a lot of research for the microalgal biomass production branch and its respective market, with the premise of sustainable bioenergy and biofuels generation (Acién, *et al.*, 2017). The cultivation of microalgae in PBRs presents several advantages, when compared to the raceway ponds, as shown in Table 1.

Table 1: Comparison of Cultivation in Raceway *versus* PBRs (Adapted from: Azeredo, 2012).

Parameter	Raceway	PBRs
Space	High	Low
Contaminations Risk	High	Less to low
Water Loss	High	Low
Oxygen Concentration	Usually low	Continuously removed
CO_2 Loss	High	Almost none
Production Reproducibility	Variable but consistent	Possible within certain tolerances
Process Control	Limited	Possible
Material Wear Construction	Low	Usually, high
Temperature	Variable but consistent	Necessary cooling
Harvest Cost	High	Average
Manutention	Easy	Hard
Construction Cost	Average	Highest
Harvest Concentration Biomass	Low	Highest

Although many studies focused only on open systems, the bottlenecks presented above and the global demand for fuels from renewable energy resources, the search for technological routes in the public and private sectors and in partnerships has motivated researchers to consolidate processes in closed microalgae biofuel production systems. A real example of this is the Sustainable Energy Research and Development Center (NPDEAS), created in 2009, at Federal University of Paraná (UFPR, Curitiba-PR, Brazil).

With the major objective of promoting research around renewable energy and potential production of biofuels and bioproducts, the NPDEAS conducts studies and demonstrations on sustainable energy from microalgae cultures, becoming a reference also in renewable energy studies. Through four pillars, the specific objectives are consolidated, being them:

1. design and fabrication of PBRs for microalgae cultivation;
2. obtaining biodiesel (biofuels) and other bioproducts, also generating co-products and possible uses of by-products, to reduce waste;
3. mathematical modeling, exergoeconomic analysis, experimental validation, and thermodynamic optimization of the components and processes, as well as of the entire system
4. dissemination of projects, improvements of the microalgae cultivation technology, assessments of system functionality, and the potential of general replication.

The center has an infrastructure of 1,400 m^2 built, with high-tech laboratories in chemistry, biotechnology, and cultivation rooms for laboratory cultivation of microalgae. It has an industrial yard that feeds the pilot scale through airlifts, and has 5 patented PBRs, with 8 m high, occupying an area of only 10 m^2 each, totaling about 3.5 km in length (Figure 1) to produce microalgal biomass.

Figure 1. Closed Tubular PBR at NPDEAS - UFPR - Brazil - used to produced microalgae

From the microalgal biomass produced at NPDEAS, it is possible to achieve a number of goals, ranging from environmental bioremediation, promoting the conversion of CO_2 (Ho *et al,* 2011), up to to extraction of lipids, carbohydrates, and proteins, and bioproducts, such as biodiesel, bioethanol, biokerosene, within the scope of biofuels. In addition, some studies are in the initial phases and are targeting the production of nutritional additives, both for human consumption and animal feed, in addition bio stimulants for plant growth. There are also efforts addressing the generation of compounds that are called by fine chemistry, such as paints and pigments, extracted from certain microalgae, such as *H. Pluvialis*. In general, a large part of the uses of microalgae are contributing to renewable energy and sustainability.

4. Biorefinery from Microalgae

The biorefinery is a process for obtaining energy, bioproducts with high added value through the transformation of biomass, and biofuels. The concept became interesting when the effects of bioremediation promoted by it began to emerge, mainly due to the increase in global causes involving the greenhouse effect, and fossil fuel emissions increasingly contributing to global warming (Juan *et al.*, 2011).

The main feature of the biorefinery approach is to obtain several products from a matrix, without damaging or losing the by-products, another interesting point is also the simplicity of the process through the equipment used, low energy consumption, in addition to being scalable and economically viable. Thus, microalgae gain a promising scenario, as they produce several bioproducts in their production chain, such as biomass, which can be applied in various markets, including renewable sources. (González-Delgado and Kafarov, 2011).

Microalgae, when metabolized, are sources that produce carbohydrates, lipids and proteins. Its carbohydrates, when extracted, can be converted into sugars and undergo the production of biofuels such as bioethanol. While the lipid, when extracted, can be subjected to various industrial operations, and become a source of biodiesel, which in turn, on the production scale, also generates other bioproducts, such as biohydrogen, biomethane, biokerosene and even biobutanol. There is also the possibility of extracting only hydrocarbons from microalgae, which may also originate other biofuels through other biochemical routes.

Several microalgae, such as *Chlorella sp., Botryococcus braunii* and *Scenedesmus sp.* have a high concentration of lipids and carbohydrates, being commonly used for energy cogeneration, that is, for the production of biofuels, such as biodiesel, bioethanol, biohydrogen, biogas and biobutanol (Deprá *et al.* 2018; Kumar *et al.* 2021; Severo *et al.* 2021).

Concomitantly, emerging sources of microalgae energy products are developed, such as bioelectricity from microbial fuel cells based on microalgae, direct combustion of biomass for heat generation and the biogeneration of volatile organic compounds with energetic potential for use as gaseous biofuels in thermal systems (Deprá *et al.* 2018; Severo *et al.* 2018, 2020).

With this, many industries have started to invest in the biorefinery market, thus investing in biofuels, Table 2 below shows the Top 10 companies around the world that work with biofuels derived from microalgae.

Table 2. Top 10 companies around the world that work with biofuels from microalgae and their cultivation system.

Company	Location	Products	Cultivation system
Algenol	USA	Bioethanol	Raceway pond and closed and semi-closed photobioreactors
Algae Productions System	USA	Biodiesel	Closed photobioreactor
Cellana	USA	Biodiesel	Raceway pond
Culture Biosystems	USA	Biofuels	Raceway pond and closed photobioreactors
Muradel Pty Ltd.	Australia	Biofuels	Raceway pond
Origin Oils Inc.	USA	Biofuels	Ponds
Proviron	USA	Biofuels	Photobioreactors
Sea 6 Energy	India	Biofuels	Open system (offshore)
Solazyme Inc.	USA	Biodiesel	Heterotrophic bioreactor and photobioreactor
Solix Biofuels	USA	Biofuels	Photobioreactor

4.1 Biodiesel

The biofuel with greater visibility, it is the best known alternative to the use of conventional diesel (coming from fossil fuel). Biodiesel can be obtained by different production methods, where paramount importance is given to its lipid matrix. The lipid can be derived from animal, vegetable fat and even from microalgae. Microalgae are considered large producers and store lipids inside, therefore, microalgae biodiesel offers distinct advantages from conventional productions, which use very high amounts of clean water. Undeniably, microalgae biomass has more energy per weight unit than other fuels (Koberg *et al.* 2011).

Many researches reveal that microalgal biomass contain about 50% of lipids in its structure, which when extracted are subjected to a transesterification reaction, which consists of mixing alcohol with fat, in front of catalysts and chemical agents, such as acids or alkalis bases, and may also rely on biological agents to aid in the reaction, such as enzymes (Gupta and Bux 2019; Kumar *et al.* 2021).

4.2 Biohydrogen

The use of hydrogen as fuel emerged as an alternative to produce energy in a cleaner way. Hydrogen is the simplest and most abundant element in the universe. Furthermore, in its gaseous form, its heating value is extremely superior when compared to other fuels (Wanghon, 2018). Added to this, there is the fact that the burning of hydrogen gas produces only water vapor, emitting no CO_2, or other greenhouse gas, and when used in low temperature fuel cells, it results in liquid water, thus characterizing it as a friendly gas to the environment. Despite the various benefits that the use of hydrogen as a fuel provides, it presents some difficulties regarding its production, for example some of the main ways to produce this fuel is through electrolysis or the reforming of hydrocarbons, processes that demand a high energy expenditure and in the case of the reforming of hydrocarbons, there is also the emission of greenhouse gases (Jacob-Furlan *et al.,* 2020).

One of the ways to produce biohydrogen can come from microalgae. Unlike other biofuels, it does not necessarily need to be a by-product from the microalgae biomass, as it can be generated similar to the process of astaxanthin generation, when microalgae is subjected to stress through the absence of some cultivation condition that it considers mandatory for itself, thus producing hydrogen gas in response to stress, this process is called photohydrolysis. There is also direct biophotolysis, where the microalgae uses sunlight to break the water molecules existing in the medium into protons, that is, H^+ ions, also generating electrons and oxygen. (Gupta and Bux 2019; Kumar *et al.* 2021).

4.3 Biomethane and Syngas

Biomethane is a by-product derived from the anaerobic digestion of microalgal biomass. The process consists of two steps. Firstly, it derives from carbohydrates, as it consists in the fermentation of a sugar, where it is converted into alcohol through the anaerobic action of bacteria (Greses, 2020). Then, the product of the previous reaction passes to another process, where methanogenic bacteria biodegrade the previous product to produce CH_4.

Being generated from CH_4 the syngas ($CO + H_2$), also known as synthesis gas, have an important role in the world for production of other fuels, such as diesel, gasoline, ethanol and hydrogen. Although they are fuel from non-renewable sources, as syngas is from a microalgae source, they also end up becoming a sustainable chain. Syngas promote a circular chain with maximum energy use (Zabed *et al*, 2020), where the Eq. (1) happens only through the use of a catalyst, giving rise to the reaction called steam reforming, such as the chemical reaction shown below:

$$CH_{4(g)} + H_2O_{(l)} \leftrightarrow CO_{(g)} + 3\ H_{2(g)} \quad (1)$$

4.4 Bioethanol

When the lipid reserves of the microalgae are low, certainly the carbohydrates will be high. This is one of the features of microalgae to have a reserve of polymers. However, this reserve does not exceed the total concentration of 50% of the total weight of its biomass. Regardless, as microalgae with high carbohydrate stores, they are strong candidates for producing bioethanol, as they have great sources of sugar fermentation. Subjected to physical, chemical and biological processes, there is a transformation of carbohydrates into fermentable sugars. Subsequently, the yeasts consume the sugar from the carbohydrate reserve of the microalgae and thus generate bioethanol. Finally, the broth generated is drained and undergoes industrial chemical processes, in addition to being drained, pumped and directed to a distillation unit (Jambo *et al.* 2016).

An alternative for the large production of biodiesel is the genetic modification of the microalgae strain, so that it produces a higher level of carbohydrate concentration for subsequent fermentation, thus making its biomass more competitive in the market, as it has another added value linked to bioethanol production (Kumar *et al.* 2021).

4.5 Biobutanol

Another alcohol produced by the fermentation of biomass is biobutanol. It is usually obtained through fermentation, a process similar to that of obtaining bioethanol, as it also needs high carbohydrate reserves for its fermentation to be promising. One of the differences in the production of biobutanol is that it can be derived from glycerol, a residual by-product of the biodiesel production chain. When it is fermented, a 4-carbon alcohol is obtained, which makes biobutanol interesting in terms of its specific mass and chemical properties, such as low volatility when compared to bioethanol, which, being an alcohol of only 2 carbons, has less attractive y properties. Biobutanol is commonly obtained from the fermentation of *Chlorella vulgaris* biomass by the bacterium *Clostridium acetobutylicum*.

Biobutanol has another highlight, as it is in addition to being a promising biofuel for the future, which is already arousing interest to researchers, it is a powerful chemical solvent in the industry, reinforcing its main role in relation to the commercial part, standing out for its sale value. (Gupta and Bux 2019; Kumar *et al.* 2021).

5. Future perspectives with PNE 2050 Brazil and UN 2030 Agenda.

In 2015, the United Nations (UN) adopted the 2030 Agenda, in order to promote sustainable development, aiming at the eradication of poverty, as well as combating climate change and preserving the oceans and forests. It is divided into 17 topics, totaling 169 goals to be achieved, and also has themes about renewable energy as a highlight.

The most pertinent objectives and related to the themes addressed are the following: SDG 7, with regard to clean and renewable energies, enabling a change in the energy matrix to clean and renewable sources. Prior to SDG 7, there are SDG 9, industrial and infrastructure development; SDG 11, Transformation of Sustainable Cities and Communities; SDG 12 energy consumption and production responsibly; and SDG 13, which finally defends the fight against global climate change. The vast majority of the energy matrix is involved both in the means of transport and in the technological development process, which further highlights the importance of global sustainable development as a justification for the global need for the SDGs.

Brazil, in turn, prepared studies on a program called the 2030 National Energy Plan (PNE), which brings together technical notes for technological development with the premise of meeting energy demand with a long-term perspective. Faced with the same scenario, in December 2020 the so-called PNE 2050 was approved, which better prepares plans for current energy matrices and presents reports on greenhouse gases and even on consumer behavior, highlighting great importance for Brazil also in this energy segment, linking the geopolitics of the future rise of the underdeveloped country.

6. Acknowledgements

This research was carried out thanks to the technical support of the Hydraulic Machines Laboratory - LMH, and the Sustainable Energy Research and Development Center - NPDEAS, both from UFPR. To the teachers and advisors for their help and guidance in the development of this project. To the ANP Human Resources Program - PRH 12.1, FINEP management. And finally, we also thank to the Brazilian National Council of Scientific and Technological Development - (CNPq) - [projects 407198/2013-0, 403560/2013-6, 407204/2013-0, 430986/2016-5, 443823/2018-9, 313646/2020-1, 310708/2017-6, 308460/2020-0 and 446787/2020-5] for funding Scientific Initiation scholarships; to CAPES; to

Ministry of Education (MEC), Brazil - (projects 062/14 and CAPES-PRINT-UFPR-88881.311981/2018-01); and to Araucaria Foundation of Parana, Brazil (project 115/2018, no. 50.579 – PRONEX).

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