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Microalgae-based biodiesel production: Current and future scenario

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ABSTRACT

Microalgae have a clear potential to be used as a source for the production of biofuel. To utilize microalgae for this purpose, the most common systems for cultivating and harvesting them are investigated in the previous years. Since there are many species of microalgae with varying lipid accumulating characteristics, a diversity of options for the production of microalgae-based energy has been analyzed. Subsequently, the energy inputs and conditions needed for growing and harvesting the microalgae and its oil production potentials are examined. This concludes with a formulation of complete concept for producing a renewable energy carrier from microalgae, which will be used for in-depth analysis.

KEY WORDS: Biodiesel, green technology, microalgae, renewable energy, sustainable production

INTRODUCTION

For microalgae, the development of dedicated culture systems only started, in the 1950s, when algae were investigated as an alternative protein source for the increasing world population. Later, algae were researched for the interesting compounds they produce, to convert CO_2 to O_2 during space travel and for remediation of wastewater. The energy crisis, in the 1970s, initiated the research on algae as a source of renewable energy. Two types of algae: Macroalgae and microalgae came into emergence for their potential use. Morphological and physiological characteristics are very much different in both. In macroalgae, the storage compound is mainly protein and carbohydrate, whereas in microalgae it is the lipid which is stored in the form of lipid globules. The cultivation systems are very different between macroalgae (seaweed) and microalgae. Because of their small (μm) size, microalgae have to be cultivated in a system designed for that purpose (placed on land or floating on water), while seaweed can be grown directly in the open sea. Many microalgae can synthesize and accumulate a large quantity of neutral lipids up to 30-70% of their dry biomass. Theoretically, 200 barrels of microalgal oil can be produced per hectare of land (Hu *et al.*, 2008). These efforts demonstrated that many species of microalgae have properties that are desirable for biofuel production. However, there are some drawbacks that have prevented their emergence as an economically viable microalgal biofuel production system. For instance, to grow microalgae, a few relatively simple conditions have to be met, viz., light, carbon source, water, nutrients, and a suitably controlled temperature. Many different culture systems that meet these requirements have been developed over the years but meeting these conditions for scaled systems is difficult. One important prerequisite to grow microalgae commercially for energy production is the need for large scale systems which can range from very simple open air systems on- or off-shore which expose the microalgae to the environment, to highly controllable, optimized but more expensive closed systems. The necessary technology for developing profitable microalgae-based fuel generation is still in various states of development of upstream and downstream processes, and the final configuration is yet to be determined and demonstrated at the industrial scale. Consequently, continued bio-prospecting efforts and the development and engineering of selected microalgal strains are required to improve the biofuel yields.

POTENTIAL OF MICROALGAL BIODIESEL

There are several reasons that biofuel is even more viable now than at any time in the past several decades. First, oil prices are significantly higher now than they were in the past and are not likely to fall to those low levels again. Biofuel is always seen as a more attractive option whenever fuel prices rise. Therefore, research into biofuel could be more cost effective now, in an age of higher gas prices. Second, though clean energy and environment were concerned in the nineties, they are much more prominent on the nation's policy agenda in the present. Fears regarding global warming and related potential environmental catastrophes have made the government much more open to considering expensive policy options with positive environmental externalities. Since environmental concerns are being weighted with much more importance today, biofuel is much more attractive now, especially when created from a feedstock that avoids the environmental detriments of large-scale farming. Third, energy independence is more important than it was back in the nineties. This is because of the wars in Iraq and Afghanistan, a loss of progress in the Arab-Israeli conflict and an increased in the fear of terrorism. Therefore, it was necessary to make any energy policy that can make us self-sufficient. Since biofuel is entirely a domestic product, it fits these criteria quite well. Finally, the recession may be an important impetus to investment in projects such as production facilities for new types of biofuel. The governments globally are currently interested in making programs for the development of biofuel production capabilities as a way to stimulate domestic investment as well as to improve fuel generation and efficiency.

While a number of bio-feed stocks are currently being experimented for biodiesel production, microalgae have emerged as one of the most promising sources for biodiesel production. It can be grown in the presence of light, water, and CO_2 enriched air and produced oil inside their cells (Ghirardi *et al.,* 2000). Such an approach can contribute to solving two major problems: Air pollution resulting from CO_2 evolution and future energy.

CULTIVATION OF MICROALGAE

Photosynthesis is an important biochemical process, in which microalgae convert the energy of sunlight to chemical energy. This chemical energy is used to drive chemical reactions such as the formation of sugars or the fixation of nitrogen into amino acids, the building blocks for protein synthesis. Since microalgae need sunlight, carbon dioxide, and water for their growth and development, therefore, they can be cultivated in open water reservoirs (Sheehan *et al*., 1998). It has been found that the unassisted growth in the open pond is slow, owing to the lower concentration of carbon dioxide; however, when carbon dioxide concentrations are increased artificially, higher growth rates can be achieved in open ponds as well (Liam and Philip, 2009). Alternatively, microalgae could be grown in closed structures called photobioreactors, where the environment is better controlled than in open ponds (Ghirardi *et al*., 2000). Finding and isolating microalgal strains are not too difficult. However, cultivating specific strains of potential microalgae for biodiesel production could be a bit more difficult as they require high maintenance and get easily contaminated by undesirable species. While the costs of setting up and operating photobioreactors would be higher than for open ponds. The efficiency and higher oil yields from these photobioreactors could be significantly higher as well, thus offsetting the initial cost disadvantage in the medium and long run (Gao *et al*., 2012). Photobioreactors can set up to continually harvested or by harvesting a batch at a time. A batch photobioreactor is set up with nutrients and microalgal seed and allowed to grow until the batch is harvested. A continuous photobioreactor is harvested either continually or more frequently. Previously, different types of photobioreactors were used for mass cultivation of selected microalgae. These include glass or plastic tubes; tanks and plastic sleeves or bags (Mario, 2013). Each type of these photobioreactors has its own plus and minus impacts for energy inputs in terms of there usability and biomass productivity.

Research on Microalgae Cultivation

There are many commercial and academic organizations globally engaged in establishing the upstream and downstream processes for microalgae-based biodiesel production The NREL (National Renewable Energy Laboratory, Department of Energy, USA) conducted research into microalgae production at a large scale. NREL designed an open "raceway" ponds, which were stirred using a paddle wheel and had carbon dioxide bubbled through it (Sheehan *et al*., 1998). The water used for these ponds could be the wastewater, freshwater, brackish water, salt water, etc., depending on the requirement of the microalgal strains grown in it. Other countries, notably Japan, are interested in closed photobioreactor systems; however, these systems are very expensive (FAO Report, 2009).

LIPID ACCUMULATION IN MICROALGAE

There are many microalgal species studied for their suitability as oil producer across worldwide. Few of them are: *Neochloris oleoabundans* (Rismani-Yazdi *et al*., 2012), *Scenedesmus dimorphus* (Velichkova *et al*., 2013), *Euglena gracilis* (Mahapatra *et al*., 2013), *Phaeodactylum tricornutum, Pleurochrysis carterae* (Valenzuela *et al*., 2012), *Tetraselmis chui, Tetraselmis suecica, Isochrysis galbana,* and *Nannochloropsis salina* (Lim et al., 2012). *Chlorella* sp. (Fukuda *et al*., 2001), *Dunaliella* sp. (Gerpen, 2014), *Nannochloris* sp. (Ghirardi *et al*., 2000), *Parietochloris incise* (Haesman *et al*., 2000), and *Botryococcus braunii* (Johnson and Wen, 2010). Microalgae contain lipids and fatty acids as membrane components, storage products, metabolites, and sources of energy. Algal strains, diatoms, and cyanobacteria are collectively referred as microalgae and have been found to contain proportionally high levels of lipids (30-60%) (Hannon *et al*., 2010). These microalgal strains with high oil or lipid content are of great interest in the search for a sustainable feedstock for the production of biodiesel. Lipid accumulation in microalgae typically occurs during periods of environmental stress, including growth under nutrient-deficient conditions (Hannon *et al*., 2010). Biochemical studies have suggested that acetyl-CoA carboxylase (ACCase), a biotin-containing enzyme that catalyzes an early step in fatty acid biosynthesis, may be involved in the control of this lipid accumulation process (Roessler *et al*., 1994). Therefore, it may be possible to enhance lipid production rates by increasing the activity of this enzyme via genetic engineering.

Research into cloning the gene that encodes ACCase from the eukaryotic algae *Cyclotella cryptica* has been undertaken, by isolating this gene (Roessler and Ohlrogge, 1993). The research found that the amino acid sequence of ACCase deduced from this gene exhibited a high degree of similarity to the sequences of animal and yeast ACCase in the biotin carboxylase and carboxyltransferase domains but less similarity exists in the biotin carboxyl carrier protein domain. Comparison of the genomic nucleotide sequence to the sequences of cDNA clones has revealed the presence of two introns in the gene. El-Sheekh and Mostafa (2005) were reported to construct the expression vectors containing this gene and developed the algal transformation protocols to enable the overexpression of ACCase in *C. cryptica* and other algal species.

HARVESTING OF MICROALGAE

Efficient harvesting is the major challenge of commercializing microalgal biofuel. Although microalgal biomass can be "energy rich," the growth of microalgae in dilute suspension at around 0.02-0.05% dry solids poses considerable challenges in achieving a viable energy balance in microalgal biofuel process operations (John and Sonia, 2013). Additional challenges of microalgae harvesting come from the small size of microalgal cells, the similarity of the density of the microalgal cells to the growth medium, the negative surface charge on the microalgae and the microalgal growth rates which require frequent harvesting. Microalgae can be harvested by a number of methods such as sedimentation, flocculation, bio-flocculation, flotation, centrifugation, ultra-filtration, or a combination of any of these. Till date, no one method of harvesting single or in combination appears to be suited to all microalgae. The harvesting method will have a considerable influence on the design and operation of both upstream and downstream processes in an overall microalgal biofuel production process.

Recently, Liu *et al*. (2013) provided an economical, efficient, and convenient method to harvest fresh microalgae. Advantages include the capability of treating high cell biomass concentrations (>1 g/L), excellent flocculation efficiencies (≥90%), operational simplicity, low cost, and recycling of medium. It has shown the potential to overcome the hurdle of harvesting microalgae to promote the full-scale application to biofuels from microalgae.

OIL EXTRACTION FROM MICROALGAE

Many efficient processes for oil extraction from microalgal biomass are emerging. A simple process is to use a mechanical press to extract a large percentage (70-75%) of the oils out of microalgal cells (Veera *et al*., 2013). It is proposed that the remaining pulp can be mixed with cyclohexane to extract the remaining oil content. After the oil extraction from microalgal biomass, centrifuge the bio-lipid from microalgal cell debris followed by chemical separation of biodiesel. Few parameters to be considered while evaluating the ideal microalgae processor are: (a) Capacity/throughput of the system and (b) Speed/density.

KNOWLEDGE-GAPS

Microalgae technology has enormous potential not only for microalgae biofuel production but also for food, feed, renewable chemicals, and many other products that are critical for a more sustainable society. However, the research till date is relatively young and disperse; most importantly on a very small scale. To prove the viability of microalgae concepts, more information is needed on the economics of the process, e.g., optimized costs of the different inputs, market value, and market size of the outputs such as fuels and other higher value compounds produced in addition to the biofuel. In developing countries like India, if economical viability and robustness can be proven, many projects can be deployed rapidly through microcredits or similar measures. As we know that not only the economic sustainability is vital but also the environmental safety is equally important. Therefore, the important tools to quantify these aspects are actually the energy and greenhouse gas (GHG) balances. As there are many different concepts to quantify the two, therefore, generalized balances are difficult. So far, the factors that are required for a complete and consistent balance are unknown. For both the economics and environmental balances, it is critical to have dependable and reproducible, long-term figures on productivity per unit area, as well as productivity of the desired compounds as a percentage of the total biomass. Much of the currently available data is from a very small scale, extrapolated from short-term experiments under optimal conditions, or even collected by commercial sectors where the marketing benefits are considered to be high numbers. Therefore, actual operating data is being manipulated. Furthermore, it is often not clear if the total biomass is the measured unit or only the organic fraction. This is because the microalgae and seaweed (macroalgae) are grown in seawater. It is assumed that some microalgal species contain over 40% of inorganic salts. However, in the case of biodiesel, the total lipid content is usually measured and reported, not the part that is extractable and usable for biodiesel.

Other environmental effects that need to be investigated are the effects on the local ecosystem of mass cultivation of productive microalgal species. Many descriptions of microalgae project, mention the generation of carbon credits (FAO Report, 2012). Microalgae are used to adsorb CO_2 from combustion gasses. This CO_2 is not sequestered, as it becomes available again during the use of the microalgal end-product. The GHG reduction of microalgae concepts is not only the displacement of less sustainable alternatives such as fossil fuels and chemical fertilizers but also any other end product that may be displaced by microalgae-based alternative. To generate carbon credits, a baseline methodology is required to determine the prevention of exact amount of GHG emission. To the extent of our knowledge, no such baseline methodology exists, nor is under development, and furthermore, it is not possible to accurately estimate the cost of CO_2 abatement through microalgae.

CONCLUSION

Biodiesel production from microalgae is a goal that still needs much research. There is a need for strenuous research on the biosynthesis of microalgal lipids, especially triacylglycerols (TAGs), if we want to understand and manipulate microalgae for the production of biodiesel. While microalgae appear to provide the natural raw material in the form of a lipid-rich feedstock, our understanding of the details of lipid metabolism to enable manipulation of the process physiologically and genetically is emerging fast. Over 30 years ago, the Aquatic Species Programme of the US Department of Energy illustrated the potential of microalgae to provide liquid energy. To harvest the benefits of that potential will not only require critical engineering innovations and breakthroughs related to microalgal mass culture and downstream processing but also focused research on essential biological questions related to regulation of lipid metabolism. Several biological and processing challenges and opportunities area head. Available biochemical knowledge about fatty acid and TAG synthetic pathways in microalgae is still fragmentary. A critical evaluation of the relationship between the cell cycle and TAG production is needed. Genetic and metabolic engineering are likely to have the greatest impact on improving the economics of production of microalgal biodiesel. Therefore, there is a need to do the metabolic engineering through genetic manipulation for enhancing the TAG production. Microalgae biomass harvesting is considered to be an expensive and cumbersome part of the industrial production of microalgal biomass due to low cell density. There is no single best method for harvesting microalgae and reducing their water content. In existing microalgal aquaculture, the common harvesting processes are flocculation, micro-screening, and centrifugation. Most importantly, cost-effective and energy-efficient harvesting methods are required to make the whole biofuels production process economical. The concept of coupling a coal-fired power plant with a microalgae farm provides an elegant approach to recycle of the CO_2 from coal combustion into a useable liquid fuel (Philip and Darzins, 2009). The combination of wastewater treatment and microalgal CO_2 fixation provides additional economic incentives due to the saving from chemicals (nutrients) and the environment benefits (Mahapatra *et al.*, 2013; Christenson and Sims, 2011). It facilitates to remove nitrogen, phosphorus, and metal from wastewater, and in turn used as nutrient for producing microalgal biomass without using freshwater. Use of the bio-refinery concept and advances in raceway ponds engineering will further lower the cost of production. If the microalgal bio-refinery concept can be adapted to a country like ours, it could become a highly distributed source of biofuel and perhaps make us self-reliant and improve our economy many folds. In the current scenario, microalgal biomass is a key link between energy, local environment, and climate change. Therefore, further research is necessary to unlock full potentials of microalgae.

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