Journal of Innovations in Pharmaceuticals and Biological Sciences
www.jipbs.com

JIPBS

ISSN: 2349-2759

Review Article

Micro-algal lipids: A potential source of biodiesel

Parveen Kumar*[1], Pradeep Kumar Sharma[1], Pradip Kumar Sharma[1], Deepansh Sharma[2]

[1]Department of Microbiology, ChaudharyCharan Singh University, Meerut, India
[2]Dairy Microbiology Division, National Dairy Research Institute, Karnal 132 001, India

Abstract

The global demand of energy is rising day by day. Petroleum based fuels are accepted as unsustainable because of depleting supplies and they also leads to the accumulation of carbon dioxide in the environment. The oil content of various microalgae is up to 80% of their dry biomass. It is also reported that some microalgae can double their biomasses within 24 hours and the shortest doubling time during their growth is around 3.5 hours which makes microalgae an ideal renewable source for biofuel production. Photosynthesis based biofuel production summarize energy production with biological carbon dioxide fixation. Ideally they are carbon neutral, and they minimize emission of greenhouse gases. Microalgae are the group of microorganisms, which contains a huge amount of triacylglycerol inside the cellular organization. Trans-esterification process produces FAME (fatty acid methyl esters). Biodiesel can be used as a fuel for vehicles in its pure form, but it is usually used as a diesel additive to reduce levels of particulates, carbon monoxide, and hydrocarbons from diesel-powered vehicles. Microalgae, which can be either prokaryotic (cyanobacteria) or eukaryotic (green algae, diatoms) can grow rapidly as they have relatively simple cellular structure and can have high lipid percentage. They also require a least land area as compared to other biofuel crops. They are highly promising biofuel source. The production of biofuel from microalgae is a complex process. It comprises microalgae cultivation in a low cost nutrient medium, harvesting of cells from cultivation medium, cell drying &cell disruption (cells separation from the growth medium), low cost and efficient lipid extraction for biodiesel production through trans-esterification and starch hydrolysis, fermentation & distillation for bioethanol production. However, these processes are complex, technologically challenges and economically expensive. A significant challenge lies ahead for devising a viable biofuel production process.

Key words: *Microalgae, Biodiesel, Algal lipids, Transesterification, Algal fuel*

***Corresponding Author: Parveen Kumar**, Department of Microbiology, ChaudharyCharan Singh University, Meerut, India

1. Introduction

Microalgae comprise a vast group of photosynthetic organisms which has an extraordinary potential for cultivation as energy crops. Microalgae seem to be most promising renewable biofuel producer that has the potential to completely displace fossil fuels without affecting supply of foods and other crop products. Microalgae are capable of producing biofuels like other crop plants as well as these microorganisms are greenhouse gas removers. Mass cultivation of microalgal biomass can be carried out by various simple and derived methods, which shows potential for high productivity of microalgal lipids. Many species of microalgae are very high amount lipid containing and analytical approach make the quality of lipid produced by many microalgal species. In brief, many parameters including lipid content, growth rate, fatty acid composition and cultivation conditions should be considered to identify the most promising microalgae species and to maximize oil yield per acre for biofuel production. Fossil fuels, crop based biofuels, coal, nuclear power, renewable energy, natural gas, etc. are the sources of fuels from a very beginning time. But due to many disadvantages of these fuel sources, and limited reservoirs, they are not promising fuel stocks for future.

A constant rising worldwide demand of motor and power generation fuels, together with environmental concerns in terms of Green House Gases (GHG), has motivated the scientists and technologists to think about various alternate sources of energy. In recent years, a lot of thrust has been put on the search for the potential biomass feed stocks from different sources, which can be converted to liquid as well as gas fuels for energy generation. Various biomasses have been identified as alternate source of energy fuels. This biomass ranges from various kinds of bio-wastes, e.g. food wastes, municipal wastes, agricultural wastes etc. energy crops, e.g. edible as well as non-edible oilseeds; and various aquatic plants identified as bio-oil sources. In the recent years much thrust has been put on to examine the possibilities of using algae as a source of bio-oil and biogas for energy applications. Algae are basically a large and diverse group of simple, typically autotrophic organisms, ranging from unicellular to multi-cellular forms. These have the potential to produce considerably greater amounts of biomass and lipids per hectare than any kind of terrestrial biomass.

Biofuel is defined as solid, liquid or gaseous fuel obtained from lifeless or living biological material and is similar to fossil fuels, which are derived from long dead biological material. Biomass or biofuel is material derived from living organisms or certain fossil fuels. This includes plants, animals and their by-products. For example, manure, garden waste and crop residues are all sources of biomass. It is a renewable energy source based on the carbon cycle, unlike other natural resources such as petroleum, coal and nuclear fuels. The United States Department of Energy estimates that if algae fuel replaced all the petroleum fuel in the United States, it would require an area of 15,000 square miles (38,849 square kilometers), which is roughly the size of Maryland. Algae, such as *Botryococcusbraunii* and *Chlorella vulgaris* are relatively easy to grow, but the algal oil is hard to extract. There are several approaches, some of which work better than others.

Algae fuel, also called oilgae or "**Third generation biofuel"** is a biofuel from algae. Algae are low-input, high-yield feed stocks to produce biofuels. It produces 30 times more energy per acre than land crops such as soybeans. With the higher prices of fossil fuels (petroleum), there is much interest in algal culture (algae farming). One advantage of many biofuels over most other fuel types is that they are biodegradable and so relatively harmless to the environment of spilled. Cyanobacteria (BGA) contain significant quantities of lipids with a composition similar to vegetable oil. Many of these cyanobacterial fatty acids are essential component of human and animal diet and are important feed additives in aquaculture [1]. The lipids of some cyanobacteria are rich in fatty acids [2]. Lipids are oils, greasy and wax-like substances which are insoluble in water but soluble in ether, chloroform, hot alcohol or carbon disulphide. They constitute major part of stored food material in animal body and are a major structural component. The term 'lipid' was used by German biochemist 'Wilhelm Bloor' in the year 1943 for a group of fat like substances. Lipids are esters of fatty acids and alcohols that comprise a large group of structurally distinct organic compounds including fats, waxes phospholipids, glycolipids etc. Lipids are the most effective source of storage energy, function as insulators of delicate internal organs and hormones and play an important role as the structural constituents of most of the cellular membranes. They also have a vital role in tolerance of several physiological stressors in a variety of organisms including cyanobacteria.

1. Microalgae – advantageous biofuel over first and second generation biofuel

Biofuels can be produced from various sources such as jatropha, rapeseed/canola, oil palm, sunflower, mustard, soybean, cotton, and corn etc. But due to low lipid content and large land area requirement for their cultivation, and low growth rate, consumption of more quantity of nutrients and difficulty in growth in adverse environment, they are not able to fulfill the dependence of world on fuels. Algae were once considered to be 'aquatic plants' but are now classified separately because they lack true roots, stems, leaves, and embryos. While we refer to algae as feedstocks for biofuels, the definition includes all unicellular and simple multi-cellular microorganisms, including both prokaryotic microalgae, e.g. cyanobacteria (Chloroxybacteria), and eukaryotic microalgae, e.g. green algae (Chlorophyta), red algae (Rhodophyta) and diatoms (Bacillariophyta). The main advantages of microalgae derived biofuels over the first and second generation biofuels are as follows. First of all, the microalgae can be produced all year round and therefore, quantity of oil production exceeds the yield of the best oilseed crops, e.g. biodiesel yield of 58,700 l /hactare for microalgae containing only 30% oil by wt., compared with 1190 l /hactare for rapeseed or Canola [3], 1892 l/hactare for Jatropha[4], and 2590 l/hactare for Karanj (*Pongamiapinnata)*[5]. The rapid growth potential and numerous species of microalgae with oil content in the range of 20–50% dry weight of biomass is the another advantage for its choice as a potential biomass. The exponential growth rates can double their biomass in periods as short as 3.5 h [4–7]. Secondly, in spite of their growth in aqueous media, the algae need less water than terrestrial crops thus the load on freshwater sources is also reduced [8]. Due to this reason, the microalgae can also be cultivated in brackish water on non-arable land, and therefore may not incur land use change, minimizing associated environmental impacts [9], without compromising the production of food, fodder and other products derived from terrestrial crops [4]. According to Chisti[4], 1 kg of dry algal biomass utilizes about 1.83 kg of CO_2, thus the microalgae biomass production can help in bio-fixation of waste CO_2 with respect to air quality maintenance and improvement.

There is a dual potential for treatment of organic effluent from the agro-food industry for algae cultivation [10]. Apart from providing growth medium, the nutrients for its cultivation, e.g. nitrogen and phosphorus, can also be obtained from wastewater. A significant advantage to environment is that algae cultivation does not require herbicides or pesticides application [11]. In addition, these can also produce valuable co-products such as proteins and residual biomass after oil extraction, which may be used as feed or fertilizer [7], or fermented to produce bioethanol or biomethane[12]. The biochemical composition of the algal biomass can be mutated by varying growth conditions, and thus significantly boosting the oil yield [13]. Also the microalgae are capable of photo-biological production of 'bio-hydrogen' [14]. It therefore becomes rather imperative that the combination of potential biofuel production, CO_2 fixation, bio-hydrogen production, and bio-treatment of wastewater; as summarized above, accentuates the potential utilization of microalgae.

2.1. Algal fuel

Algae fuel, also called oilgae or "**Third generation biofuel"** is a biofuel from algae. Algae are low-input, high-yield feed stocks to produce biofuels. It produces 30 times more energy per acre than land crops such as soybeans. With the higher prices of fossil fuels (petroleum), there is much interest in algal culture (farming algae). One advantage of many biofuels over most other fuel types is that they are biodegradable, and so relatively harmless to the environment of spilled. The third-generation biofuels appear to be the only source of renewable biodiesel that is capable of meeting the global demand for transport fuels. Like plants, microalgae use sunlight to produce oils but they do so more efficiently than crop plants. Oil productivity of many micro algae greatly exceeds the oil productivity of the best oil producing crops. Microalgae can provide several different types of renewable biofuels. These include methane produced by anaerobic digestion of the microalgal biomass [7] biodiesel derived from microalgal oil [15, 16, 17, 18, 19] and photobiologically produced biohydrogen [20, 21, 22, 23]. It is possible to extract a number of valuable and useful by-products from the algal biomass, including fatty acid, amino acids, plant hormones, pigments, and oils for biodiesel, nutracueticals, metabolites and ß-carotene.

Microalgae are a large and diverse group of photosynthetic with a simple cellular structure, ranging from unicellular to multicellular forms; they can be found anywhere water and sunlight co-occur, including soils, ice, lakes, rivers, hot springs and ocean [24]. They have the ability to capture carbon dioxide and convert energy of sunlight to chemical energy. Algal oils, which can be used to produce biodiesel, are usually accumulated as membrane components, storage products, metabolites and sources of energy under some special conditions. Recently, microalgae have attracted considerable attention from researchers and entrepreneurs as an alternative non-food biodiesel feedstock owing to their high oil content and rapid biomass production, e.g. *Spirulina, Westiellopsis,* *Chlorella, Anabaena, Calothrix*, and some other eukaryotic algae, etc.

2.2. Merits of algal fuel over other biofuels

Algae can be preferred feedstock for high energy density, fungible liquid transportation fuels. There are several aspects of algal biofuel production that have combined to capture the interest of researchers and entrepreneurs around the world-

- Algal biomass productivity can be high per acre of cultivation.
- Algae cultivation strategies can minimize or avoid competition with arable land and nutrients used for conventional agriculture.
- Algae can utilize waste water, produced water, and saline water, thereby reducing competition for limited freshwater supplies.
- Algae can recycle carbon from CO_2-rich flue emissions from stationary sources, including power plants and other industrial emitters.
- Algal biomass is compatible with the integrated biorefinery vision of producing a variety of fuels and valuable co-products.

2.3. Best source of microalgal biofuels production

No doubt microalgae are the promising feedstock for biofuel production, these are only renewable source that potentially completely substitute fossil fuels. Many algal species are potential source for biofuel production out of all these *Botryococcusbraunii* (25-75% dry wt. biomass), *Dunaliella* sp. (17-67% dry wt.biomass) and *Schizochytium sp.* (50-77% dry wt.biomass) Are highest lipid containing microalgae which can be potential source for replacement of fossil fuels.

Microalgae species	Lipid content (% dry wt. biomass)
Ankistrodesmus sp.	*24-41*
Botryococcusbraunii	*25–75*
Chlorella emersonii	*14–57*
Chlorella protothecoides	*14–57*
Chlorella vulgaris	*5-58*
Dunaliella sp.	*17–67*
Zitzschia sp.	*45-47*
Isochrysis sp.	*7-40*
Nannochloropsis sp.	*20-56*
Neochlorisoleoabundans	*29–65*
Crypthecodiniumcohnii	*20–51*
Chlorella minutissima	*57*
Phaeodactylumtricornutum	*18–57*
Scenedesmusobliquus	*11-55*
Scenedesmusdimorphus	*16-40*
Schizochytrium sp.	*50-77*
Isochrysisgalbana	*7–40*

Table 1: Lipid content of many microalgae species [4, 25, 26, 27, 28]

3. Microalgal lipids

Lipids are high-value energy material, which are produced by microalgae more efficiently by converting sunlight energy. Thus microalgae especially cyanobacteria are very good at forming lipids and growing very fast. The lipids oils can be used as a feed stock for liquid fuels, such as biodiesel. The composition of miacroalgal fatty acids has a significant effect on the fuel properties of biodiesel produced. Several researchers reported that reducing the saturated fatty acid content of plant oil can improve the cold temperature low properties of the biodiesel derived from it because long-chain saturated fatty esters significantly increase the cloud point and the pour point of biodiesel [29]. Microalgal oils are mostly composed of four unsaturated fatty acids, namely palmitolleic (16:1), oleic (18:1), linoleic (18:2) and linolenic acid (18:3). Saturated fatty acids such as palmitic (16:0) and stearic (18:0) also present with a small proportion [30]. Some special microalgae could synthesize polyunsaturated fatty acids such as C16:4 and C18:4 in *Ankistrodesmus spp*., C18:4 and C22:6 in *Isochrysis spp.*, C16:2, C16:3 and C20:5 in *Nannochloris spp*., C16:2, C16:3 and C20:5 in *Nitzschi spp*. [31]. But biodiesel from highly unsaturated sources oxidizes more rapidly than conventional diesel, resulting in forming insoluble sediments to interfere with engine performance. Therefore, the proper percentage of saturated and unsaturated fatty acid is very important to microalgae as a biodiesel feedstock. (Figure 1)

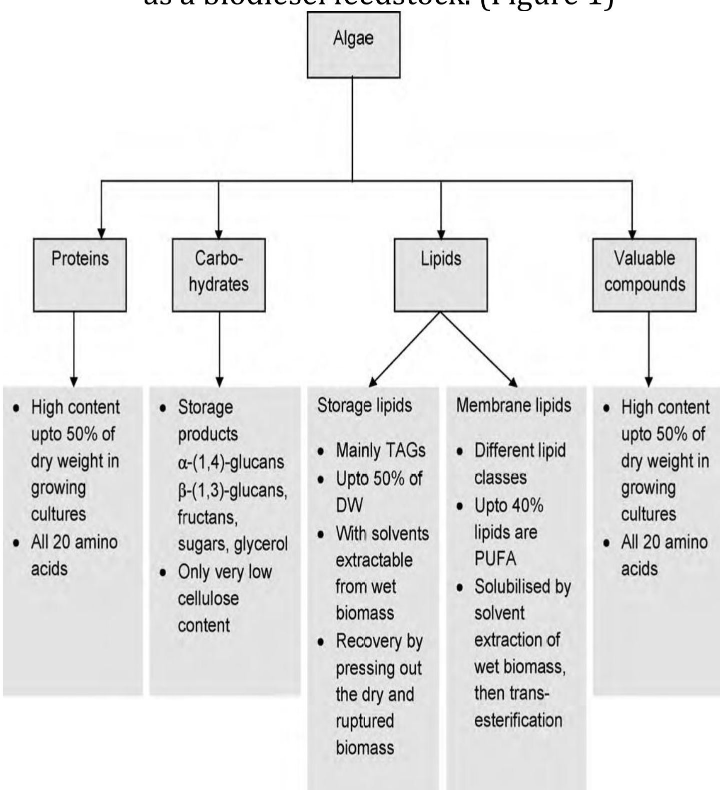

Figure 1 : Various products of microalgae [32]

3.1. Transesterification

Biodiesel consists of fatty acid methyl esters, which typically are derived from triacylglycerols (TAGs) by trans-esterification with short chain alcohols such as methanol, with glycerol as a byproduct. The current feedstocks for commercial biodiesel include waste cooking oil, animal fat and various oleaginous species such as soybean, rapeseed, corn, sunflower, peanut, *Jatropha* and oil palm [33, 34, 35]. Rapeseed oil is the predominant feedstock in European Union and soybean oil is the main contributor in the USA. However, these food-based raw materials have resulted in the debate "food vs. fuels". Since biodiesel production grew rapidly in the world, there was a dramatic increase in food prices for vegetable oils [36].

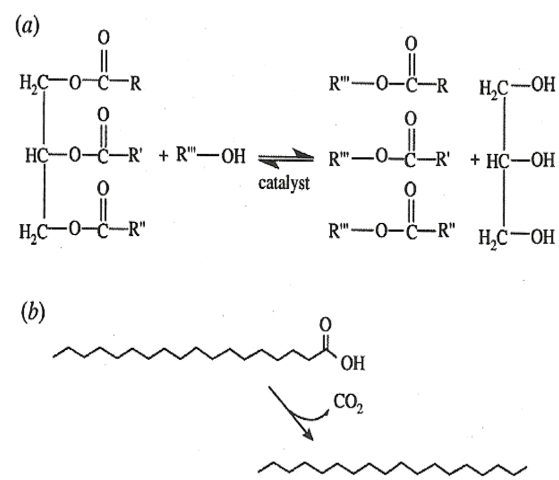

Figure 2 : Transesterification reaction of triacylglycerols with methanol, yielding methyl esters and glycerol

2. Indian prospective for micro-algal biofuel production

India, a fast growing economy facing the challenge of meeting a rapidly increasing demand for energy, ranks sixth in the world in terms of energy demand. Its economy is projected to grow at 7–8 percent over the next two decades and there will be a substantial increase in demand for oil to manage transportation and to meet various other energy needs. While India has significant reserves of coal, it is relatively poor in oil and gas resources. Due to stagnating domestic crude production, India imports approximately 72 percent of its petroleum requirement. The global annual requirement of petroleum products is approx. 17,000 MMT, out of which estimated Indian requirement is 120 MMT. Our domestic production of crude oil and natural gas will remain around 34 millon tones during 2006–07. The huge gap between demand and supply may be met only by import. The net import burden has increased from $22.6 billion (Rs. 101,963 crores) in 2004–05 to $34.1 billion (Rs. 150,557 crores) in 2005–06 and taking into account, the average prices till now during the current year, the net import bill for 2006–07 could be of the order of $43.3 billion (Rs. 190,000 crores). The increasing trends show one and half times increase annually and if the present increasing trend continues, it would be a matter of very serious concern for the country. If the corrective measures are not taken in time, the India's maximum revenue will drain towards the import of petroleum products. Bio-fuels are renewable sources of energy derived from biological raw material. Two sources of bio-fuels – ethanol and bio-diesel are gaining worldwide acceptance as one of the solutions for problems of environmental degradation, energy security, restricting imports, rural employment, agricultural economy, owing to reduced dependence on oil import; savings in foreign exchange and reduced vehicular pollution. Two major bio-fuels for the transport sector, bio-ethanol and bio-diesel, are becoming popular in many countries across the world.

Bio fuel production around the world rose by 13.8% from the previous year in 2010 with bio-ethanol holding 72% of the total share. According to BP's Statistical Review of World Energy, production is concentrated around a few countries, with Brazil and the US producing 40.924 million tons of oil equivalents amounting to 69.2% of the total global production. North America and South & Central America produced 95% of the world's ethanol in 2010 while biodiesel production is concentrated in Europe and Eurasia with 59 % of the global biodiesel produced there. In 2010, India registered the highest change in bio-fuel production from the previous year. With the addition of 0.151 million tons of oil equivalent, India registered an 85% increase in production over its 2009 production. India holds only 0.3% share of the global of production in 2010. However, this is likely to increase as India prepares for a change in its bio-fuel mandate from its current E5 (5% ethanol content in the fuel supply) to E10 (10% ethanol content in the fuel supply) and ultimately E20 by 2017.Present estimates indicate India's bio-fuel demand at 0.5 billion gallons in 2012 which will grow to 6.8 billion gallons by 2022. According to India's Planning Commission, the demand for diesel in India is five times higher than petrol. But while the ethanol industry is mature, the biodiesel industry is in its infancy. India's ambitious National Biodiesel Mission will aid in the technological research, production and trade of biodiesel in order to meet 20% of the country's diesel requirements by 2012 and drive production levels upwards for the subsequent years.

3. Conclusion

In this review, we discussed on ability of microalgal fuel to replace all fossil fuels. Its environmental benefits and the fact that it is a renewable source of energy and can remove greenhouse gases from environment, now it can be an achievable target. In order to produce low cost biofuel, today's research should be focused on cost reduction on small as well as large scale mass cultivation. Production cost can be reduced by using waste water as nutrient media. In contrast genetic and metabolic changes got less attention which can make microalgae as genetically modified microorganisms, capable of producing high oil percentage in their cell organelles. Cultivation system should be of high efficiency and low input. Photobioreactors provide a controlled environment that can be tailored to the specific demands of highly productive microalgae to attain a consistently good annual yield. Efforts should be undertaken to understand the fundamental genetic and cellular processes involved in the synthesis and regulation of potential fuel precursors from diverse species of algae. While a better understanding of the basic biology of algal growth and metabolite accumulation using modern analytical approaches will provide a wealth of hypotheses for higher oil production.

References

1. Borowitzka MA. Microalgae as source of essential fatty acids.*Austr.J.Biotechnol*. 1988;(I): 58-62.
2. Singh SC. Study on lipids of some dessication tolerant cyanobacteria. Ph.D Thesis, BanarasHindu University, Varanasi, India. 2002.
3. Schenk P, Thomas-Hall S, Stephens E, Marx U, Mussgnug J, Posten C, *et al.* Second generation biofuels: high-efficiency microalgae for biodiesel production. BioEnergy Res;(1):2008; (1):20– 43.
4. Chisti Y. Biodiesel from microalgae. BiotechnolAdv;252007; (3):294–306.
5. Lele S. Indian Green Energy Awareness Center, http://www.svlele.com/ karanj.htm.
6. Metting FB. Biodiversity and application of microalgae. J IndMicrobiol;(17):1996; (5–6):477–89.
7. Spolaore P, Joannis-Cassan C, Duran E, Isambert A. Commercial applications of microalgae. J Biosci Bioeng;1: 2006; 01(2) : 87–96.
8. Dismukes GC, Carrieri D, Bennette N, Ananyev GM, Posewitz MC. Aquatic phototrophs: efficient alternatives to land-based crops for biofuels. CurrOpinBiotechnol;19: 2008;(3):235–40.
9. Searchinger T, Heimlich R, Houghton RA, Dong F, Elobeid A, Fabiosa J, *et al.* Use of U.S. croplands for biofuels increases greenhouse gases through emissions from land-use change. Science; 319:2008; (5867) :1238–40.
10. Cantrell KB, Ducey T, Ro KS, Hunt PG. Livestock waste-to-bioenergy generation opportunities. BioresourTechnol;99: 2008;(17):7941–53.
11. Rodolfi L, Zittelli GC, Bassi N, Padovani G, Biondi N, Bonini G, *et al*. Microalgae for oil: strain selection, induction of lipid synthesis and outdoor mass cultivation in a low-cost photobioreactor Biotechnol Bioeng;102:2008; (1): 100–12.
12. Hirano A, Ueda R, Hirayama S, Ogushi Y. CO_2 fixation and ethanol production with microalgal photosynthesis and intracellular anaerobic fermentation. Energy;22: 1997;(2–3):137–42.
13. Qin J. Bio-hydrocarbons from algae—impacts of temperature, light and salinity on algae growth. Barton, Australia: Rural Industries Research and Development Corporation.2005;
14. Ghirardi ML, Zhang L, Lee JW, Flynn T, Seibert M, Greenbaum E, *et al*. Microalgae: a green source of renewable H_2. Trends Biotechnol; 2000; 18(12):506–11.
15. Roessler PG, Brown LM, Himmel ME, Baker J, Overend RP, Dunahay TG, Heacox DA, Jarvis EE, Schneider JC, Talbot SG &Zeiler KG. Genetic engineering approaches for enhanced production of biodiesel fuel from microalgae. In Enzymatic Conversion of Biomass for Fuels Production, *eds. Am. Chem. Soc*., 1994; 256-270.

16. Hoda H. senousy, GordanW. Beaks and Ethan Hack J Phycol (40):2004;412-423.
17. Sheehan J, Dunahay T, Benemann J, Roessler PA. Look Back at the U.S. Departmentof Energy's Aquatic Species Program-Biodiesel from Algae; National Renewable Energy Laboratory: Golden, CO, USA.1998
18. Banerjee A, Sharma R, Chisti Y and Banerjee UC. *BotryococusBraunii*: A renewable source of hydrocarbons and other chemicals. C*rit Rev Biotechnol.,* 2002;(22):245-279.
19. Gravirilescu M, Chisti Y. Biotechnology- a sustainable alternative for chemical industry (23):2005; 471-499.
20. Ghirardi ML, Zhang L, Lee JW, Flynn T, Seibert M, Greenbaum E, *et al*. Microalgae: a green source of renewable H_2. Trends in Biotechnology; 2000; 18 (12) 506-11.
21. Akkerman I, Janssen M, Rocha J, Wijffels RH. Photobiological hydrogen production,Photochemicalefficiency and bioreacter design. Interternational journal of Hydrogen energy; 2002; 27. (11): 1195-208.
22. Melis A. Green alga hydrogen production: progress, challenges and prospectes. Interternational journal of Hydrogen energy; 2002; 127 (11-12): 1217-28.
23. Kapdan IK, Kargi F. A review on biohydrogen production from waste material.Enzyme and Microbial Technology. 38; 2006; 569-582.
24. Parker MS, Mock T, Armbrust EV. Genomic insights into marine microalgae. Annu. *Rev. Genet.,* 2008; 42: 619-645.
25. Li Y, Horsman M, Wang B, Wu N, Lan CQ. Effects of nitrogen sources on cell growth and lipid accumulation of green alga *Neochlorisoleoabundans*. Appl. Microbiol. Biotechnol.2008; 81: 629-636.
26. Mata TM, Martins AA, Caetano NS. Microalgae for biodiesel production and other applications: A review. Renew. Sustain. Energy. 2009; Rev. doi:10.1016/j.rser.2009.07.020.
27. Um BH, Kim YS. Review: A chance for korea to advance algal-biodiesel technology. *J. Ind. Eng. Chem.,* 2009; 15: 1-7.
28. Sialve B, Bernet N, Bernard O. Anaerobic digestion of microalgae as a necessary step to make microalgal biodiesel sustainable. Boitechnol. Adv.,2009; 27: 409-416.
29. Serdari A, Lois E &Stournas S. Impact of esters of mono- and dicarboxylic acids on diesel fuel quality. *Ind. Eng. Chem. Res.,*1999; (38): 3543-3548.
30. Meng X, Yang JM, Xu X, Zhang L, Nie QJ & Xian M. Biodiesel production from oleaginous microorganisms. *Renew. Energy.,* 2009; 34: (1): 1-5.
31. Thomas WH,Tornabene TG, Weissman J. Screening for lipidyielding microalgae: activities for 1983. SERI/STR-231-2207. 1984; p. 31
32. Schmid-Straiger U. Algae biorefinery–Concept. National German Workshopon Biorefineries, 15 September 2009, Worms.
33. Barnwal BK, Sharma MP. Prospects of biodiesel production from vegetables oils in India.*Renew Sustain Energy Rev.*, 2005; (9): 363-378.
34. Felizardo P, Correia MJN, Raposo I, Mendes JF, Berkemeier R &Bordado JM. Production of biodiesel from waste frying oil. *Waste Manage.,* 2006; (26): 487-493.
35. Vasudevan PT &Birggs M. Biodiesel production-current state of the art and challenges. *J. In. Microbiol. Biotechnol.*2008; (35): 421-430.
36. Chakravorty U, Hubett M-H, Nøstbakken L. Fuel versus food. *Annu. Rev. Resour. Econ.*, 2009;1: 645-663