Renewable Energy 69 (2014) 114–122

Contents lists available at ScienceDirect

Renewable Energy

journal homepage: www.elsevier.com/locate/renene

Review

Evolution retrospective for alternative fuels: First to fourth generation

Kasturi Dutta, Achlesh Daverey, Jih-Gaw Lin*

Institute of Environmental Engineering, National Chiao Tung University, 1001 University Road, Hsinchu City 30010, Taiwan

ARTICLE INFO

Article history:
Received 27 May 2013
Accepted 24 February 2014
Available online 3 April 2014

Keywords:
Alternative biofuels
Greenhouse gas
Biodiesel
Algae
Metabolic engineering

ABSTRACT

The ever increasing worldwide demand for liquid and gaseous fuels along with overwhelming environmental concerns for greenhouse gas emissions have driven scientists and technologists to consider different alternative energy sources. In past decades, several biomass sources have been identified with increasing potential to be used as new alternative sources of energy - the “Biofuels”. The evolution of biofuels is classified into four different generations.

In this article an overview of the systematic evolution of different biofuel generations with their advantages and disadvantages has been presented. The advancements in technology, reduction in greenhouse gas emission and assessment of commercial production cost of each generation of biofuel have also been highlighted. Finally this review provides an outlook for a better future generation biofuel.

1. Introduction

Inescapable growth of the global population, which is projected to exceed 9 billion by 2050, will raise the average calorie intake pushing the productivity from already scarce arable land to its limit. The energy demand in developing nations are expected to increase by 84%, and nearly one-third of this additional fuel possibly come from alternative renewable sources – such as biofuels [1]. In the last few decades major research emphasis has been directed towards sustainable and economical sources of biofuel as evident from large number of research publications worldwide, particularly from Europe and USA (Fig. 1a). The year wide distribution of research articles published on biofuels has exponentially increased over the past 20 years (Fig. 1b). This clearly depicts the growing concern to have alternative biofuel energy resources.

The paradigm in biofuel research and developments has considerably been shifted toward alternative and efficient biomass source. Starting from the production of biodiesel from of edible or nonedible seed-oils to algae metabolic engineering, this series of evolution are classified into four generations. The major feedstocks for first generation biofuels (corn, wheat, barley and sugarcane) are the sources of food, thus may result in a food-fuel competition. It was reported that though only 2% of world’s arable land had been used to grow biomass feedstock [2] for first generation biofuel production, it had significant contribution toward increased commodity prices for food and animal feeds. However, direct or indirect impact of biofuels on food price hike remains inconclusive in literature or media. The production of biofuels from agricultural and forest waste or non-food crop could resolve the crisis. Second generation biofuel from these lignocellulosic feedstocks including by-products (cereal straw, sugarcane bagasse, forest residues), wastes (organic components of municipal solid wastes), and dedicated feedstocks (purposely-grown vegetative grasses, short rotation forests and other energy crops) would also need land in competition with food and fiber production. However, energy yields (in terms of GJ/ha) from these crops are likely to be higher than those of first-generation biofuels crops or their products [3].

The search for alternative sources continued further to reduce the food cropland competition until using algae - as a sustainable and rich source of biofuel which is known as third generation biofuel. Algae do not compete with food or other crops and can be cultivated in shallow lagoons or raceway ponds on marginal land or closed ponds. Moreover, algal biofuel feedstocks can be produced throughout the year unless it is limited by light irradiation, and the oil yield can even exceed that of the best oilseed crops. For example, yield of biodiesel from algae (58,700 L/ha) containing only 30% oil by weight, is much higher when compared with rapeseed or canola (1190 L/ha) [4], Jatropha (1892 L/ha) [5], and Karanj (*Pongamia pinnata*) (2590 L/ha) [6]. However, the biofuels from above sources have limitations in terms of ecological footprint, economic performance, dependence on environment (sunlight), geographical location (latitude) and thus insufficient to replace fossil fuels.

---

* Corresponding author. Tel.: +886 35722681; fax: +886 35725958.
E-mail address: jglin@mail.nctu.edu.tw (J.-G. Lin).

http://dx.doi.org/10.1016/j.renene.2014.02.044
0960-1481/

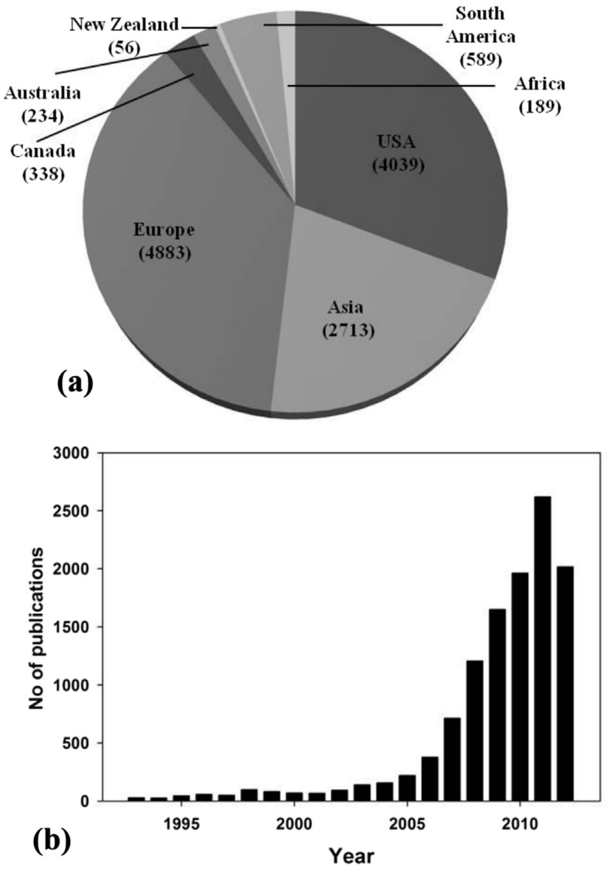

Fig. 1. Number of published paper related to biofuel (a) geographical area wise and (b) annually (based on a literature survey by using the ISI Web of Knowledge on April 2013).

Metabolic engineering of algae for production of biofuel is considered as fourth generation of biofuel and has great potential in providing sustainable and clean energy [7].

The advantages and limitations of different generations of biofuel are given in Table 1. Fig. 2 shows a comparative view on research performed on different generations of biofuel in past years.

Though the idea of first generation biofuel had emerged a long time ago, it gained significant attention in early 90's. However, the majority of research work has been carried out in between 2006 and 2010. Whereas fourth generation biofuel research has set in from that period only. Based on published research articles, it appears that in last two years third generation biofuel has received the maximum emphasis.

The aim of this paper is to provide the evolutionary path of biofuel research in terms of feedstock development and technological advancement to obtain sustainable and economically feasible replacement of fossil fuel. The review presents the cost effectiveness, GHG mitigation and energy efficiency of each biofuel generation compared to fossil fuel. Altogether the paper describes about each generation of biofuel with its merits and demerits, and how the limitations of each biofuel generation have been overcome by its successor.

2. Biomass production for different generations of biofuels

Biofuels are classified in four different generations depending on their biomass feedstock. Following sections discuss about the biomass feedstock for different generation biofuels and their suitability to be used as replacement of fossil fuel. Table 2 shows the feedstocks and the end products of different generations' biofuels.

2.1. First generation biofuel feedstock

As discussed earlier, first generation biofuel feedstocks mainly comprised of oilseed, sugarcane and other oil containing food and animal feed crops. First generation bioethanol is mainly produced from sugar containing plants or cereal (grain) crops. Till date the largest volume of biofuel is produced in the form of ethanol, 80% of which has come from corn and sugarcane. Hayashida et al. [8] obtained about 20% (v/v) ethanol conversion from raw ground corn using a mutant of *Aspergillus awamori* var. *kawachi*. Rolz and de Leon [9] studied the ethanol production from sugarcane at different maturity levels. They observed better ethanol yields after 300–325 days of planting. Vegetable oils are also used after a range of conversion to fatty acid methyl or ethyl esters. Nabi and his co-workers [10] obtained 77% biodiesel yield with 20% methanol in

Table 1
Pros and cons of alternative fuel options.

Generation	Pros	Cons
First	GHG savings Simple and low cost conversion technology	Low yield Cause food crisis as a large portion arable land required for growing crops
Second	GHG savings Utilize food wastes as feed-stock No food crop competition Use of non-arable land for growing few energy crop	Costly pretreatment of lignocellulosic feedstock highly advanced technology need to be developed for cost effective conversion of biomass to fuel
Third	Easy to cultivate algae Higher growth rate No food crop competition Versatility: can use wastewater, seawater	More energy consumption for cultivation of algae (for mixing, filtration, centrifugation etc), low lipid content or biomass contamination problem in open pond system High cost of photo-bioreactor
Fourth	High yield with high lipid containing algae More CO_2 capture ability High production rate	Initial investment is high research is at its primary stage

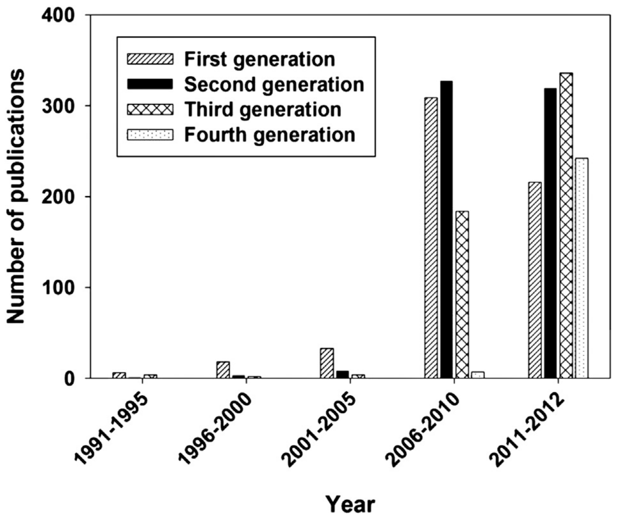

Fig. 2. Number of published paper related to different generations of biofuel in last 20 years (based on a literature survey by using the ISI Web of Knowledge).

Table 2
Different generations of biofuel: major source, process and their examples.

Generation	Feedstocks	Processing technology	Examples of biofuel	Reference
First	Edible oil seeds, food crops, animal fats	Esterification and transesterification of oils and fermentation of sugars, thermochemical process	Biodiesel, bioethanol, biobutanaol	[46,67]
Second	Nonedible oil seeds, waste cooking oil, Ligno-cellulosic feedstock materials: cereal straw, sugarcane bagasse, forest residues	Physical, chemical, biological pretreatment of feedstock and fermentation, thermochemical process	Bioethanol, biobutanol, biodiesel, syngas	[2,3,18]
Third	Algae	Algae cultivation, harvesting, oil extraction, transesterification, or fermentation, or thermochemical process	Biodiesel, bioethanol, biobutanaol, syngas, biohydrogen, methane	[6,68]
Fourth	Algae and other microbes	Metabolic engineering of algae with increases carbon entrapment ability, cultivation, harvesting, fermentation, or oil extraction, transesterification, or thermochemical process	Same as in 3rd generation	[29,73]

presence of 0.5% sodium hydroxide from cottonseed oil. A reduction in carbon monoxide (CO), particulate matter and smoke emissions in exhaust were experienced for all biodiesel mixtures. A slight increase in NO_x emission was observed for cotton-seed biodiesel mixtures. Efforts were also made to evaluate the usability of vegetable oils directly in diesel engines. However, vegetable oils were deemed unsuitable leaving heavy deposition of wax and gum in the engines [11]. Another promising source of biofuel could be vegetable oil sludge- a major by-product of vegetable oil factories [12]. The cracking reaction of vegetable oil sludge results in products like dry gas, liquefied petroleum gas (LPG), gasoline, light cycle oil (LCO) and heavy cycle oil (HCO) similar to those of petroleum cracking process.

2.2. *Second generation biofuel feedstock*

About 80% cost of biodiesel is accounted for oil feedstock [2]. Bioethanol production from low cost crops, forest residue, wood process waste and organic portion of municipal waste has been classified as second generation biofuel [2,3]. Because of its high octane number, low cetane number and high heat of vaporization, bioethanol blend is capable of replacing gasoline [13]. Bioethanol production using these materials does not require any additional land and therefore would not have any impact on food and fiber crop production. Lignocellulosic biomass is abundant in nature, though only small portion of it could be utilized. Theoretically these sources of biomass can supply energy of 100 EJ per year [2]. Vegetative grasses like *Miscanthus*, switchgrass and short rotation forest species (Eucalyptus, Poplars, Robinia) are becoming increasingly popular, as these energy crops can be grown in marginal and degraded lands, which are not otherwise suitable for food/fiber crop production [2]. Chen et al. [14] studied ethanol production from differently pre-treated xylose/cellulose fraction of corn cobs by saccharification and/or fermentation with *Candida shahatae* and *Saccharomyces cerevisiae*. They observed that 40–70% of hemicellulose and 72–90% of cellulose in corn cobs could be converted to ethanol using different fractionation methods. Recently, use of kapok fiber, pineapple waste, waste papers and coffee residue waste for bioethanol production has been reported [15–18]. Bioethanol production from agricultural waste can also be coupled with biogas production. Kaparaju et al. [19] investigated the production of bioethanol, biohydrogen and methane from wheat straw within a biorefinery framework. The biodiesel produced from raw Jatropha and Karanj oil in blends with diesel were capable of power generation in a 7.5-kVA diesel engine generator [20]. Deshpande et al. [21] studied production of biogas (which may serve as energy source for cooking) from de-oiled seedcake of Mahua (*Madhuca indica*) and Hingan (*Balanites aegyaptiaca*). Genetic modification can be a useful tool to develop low input fast growing energy crops with lesser requirements of insecticides, fertilizers and water, thus reducing the overall cost of biofuel production. The genetically modified wheat and barley have been shown to contain more hydrolyzable biomass without compromising their compositions and sacrificing their agronomic performance [22]. Recently apart from ethanol, advanced biofuels such as isopropanol, butanol, isobutanol and fernesol are gaining more attractions because of their high energy density, being low hygroscopic and less corrosive to pipelines during transportation [23–25]. Metabolic engineering of biosynthetic pathways leading to production of these alcohols can improve productivity. The overexpressions of aldehyde/alcohol dehydrogenase in *Clostridium tyrobutyricum* not only enhanced the butanol production of about 27–30% but also its tolerance towards butanol [24]. Modification of amino acid biosynthetic pathway in *Escherichia coli* also resulted in higher production of 1-butanol and 1-propanol [26].

2.3. *Third and fourth generation biofuel feedstock*

Recently algae have received a significant interest as alternative biofuel feedstock because of their higher photosynthesis and fast growth rate as compared to any terrestrial plant. Algae may contain up to 70% of lipid on a dry weight basis [27] and can grow in liquid medium utilizing different wastewater streams (saline/brackish water/coastal seawater) resulting in reduced freshwater demand [28]. Recent research activities have been focused on the search for ideal combination of algal species with high lipid content and their optimum growth conditions. Several algal species such as *Botryococcus braunni*, *Chaetocero scalcitrans*, several *Chlorella* species, *Isochrysis galbana*, *Nanochloropsis*, *Schizochytrium limacinum* and *Scenedesmus* species have been studied as potential sources of biofuel [5,6,29]. Among these, the highest average lipid content and biomass was obtained in *Chlorella* but has low triglyceride content [6,29]. Whereas some algal species like *Botryococcus braunii, Nannochloropsis* and *Schizochytrium* sp. can produce 25–75%, 31–68% and 50–77% of triglycerides on dry cell weight basis, respectively, though the yield of biomass is low in each case [5,6,29]. It remains a common observation that fast growing algae (such as *Spirulina*) have low oil content whereas high lipid containing algae are slow growing organisms. Therefore, identification of correct species with high biomass as well as high lipid content is necessary for commercialization of algal biofuel. Type of cultivation (phototrophic and heterotrophic) also affects the biomass and lipid yield in same microalgal strain [5,6]. Scientists are looking for proper cultivation method for these species which will lead to the maximization of lipid contents to make it more cost-effective and sustainable source of biofuel [6]. In this context genetic modification/metabolic engineering could be promising alternative to increase the lipid content and biomass yield of algae [6]. The pathways for lipid anabolism and catabolism are investigated to identify and modify key enzymes of these pathways [30]. The inactivation of ADP-glucose pyrophosphorylase in a *Chlamydomonas* starchless mutant led to a 10-fold increase in triacylglycerol (TAG), suggesting that shunting of photosynthetic carbon partitioning from starch to TAG synthesis may represent a more effective strategy than direct manipulation of the lipid synthesis pathway to overproduce TAG [31]. Lan and Liao [32] showed that a modification in CoA-dependent 1-butanol production pathway into a cyanobacterium, *Synechococcus elongatus* can produce butanol from CO_2 directly.

3. Technology used in different generations of biofuel production

First generation biofuel (bioethanol) production, as shown in Fig. 3 starts with harvesting the crops, extraction of sugar and its dilution to about 20% solution, if necessary, and addition of yeasts to convert into ethanol. A two-step distillation process involving other solvent (such as cyclohexane), produces the desired dry ethanol, which is the most suitable for existing distribution and usage patterns for petrol. The biodiesel from oil is produced by trans-esterification reaction with chemical catalyst (acid/alkali) or enzyme [33,34].

The produced biodiesels generally are subjected to distillation process to remove byproducts like glycerol produced during the reaction. Distillation process can be avoided by proper and careful decanting of the lower layer and water washing (or dry absorbent washing) of the biodiesel product after transesterification. However, distillation is necessary to reduce mainly the monoglycerols that may present in trace concentrations after water washing. Lipases are becoming much popular bio-catalysts for the production of alkyl fatty acid esters. Some of the lipases catalyze trans-esterification of triglyceride, converting them to methyl esters; and other catalyze esterification of the fatty acids in presence of alcohol. The use of enzymatic pretreatment for conversion of fatty acid into biodiesel is being considered but yet not commercialized till date [34] due to the high energy costs and slow reaction rate. Moreover, the yields of methyl esters are typically less than 99.7%, which is the minimum requirement for fuel-grade biodiesel [34].

The basic steps for production of second generation biofuel such as cellulosic ethanol and butanol include pre-treatment, saccharification, fermentation and distillation (Fig. 3). Pretreatment is necessary for separation of cellulose and hemicellulose from lignin, followed by enzymatic hydrolysis (also known as saccharification) into their constituent simple sugars, fermentation and distillation. Second generation biochemical alcohol production through thermochemical biomass conversion with either gasification or pyrolysis occurs at much higher temperature and pressure than those in biochemical systems. Although, the gasification is more cost-intensive and requires larger scale for the best economics, it finally produces cleaner fuel to be used directly in engines [35]. Gasification based processes can produce variety of biofuels including Fisher-Tropsch liquids (FTL), dimethyl ether (DME) and various alcohols [35,36]. French and Czernick [37] reported three types of biomass feedstocks; cellulose, lignin and wood pyrolysis in quartz boats in presence of different catalysts namely, commercial zeolites, laboratory prepared ZSM-5 catalysts modified by substituting Al or hydrogen with different metals (Co, Fe, Ni, Ce, Ga, Cu, Na), X and Y zeolites, and different silica and alumina materials at temperature ranging from 400 °C to 600 °C. The pyrolysis process of biomass conversion to fuel is still facing challenges like low yield and low quality of bio-oil with higher contaminants. In this context, Yin [38] reported that microwave heating could enhance bio-oil production using pyrolysis.

Third and fourth generation biofuels include common steps of cultivation of microalgae, harvesting, extraction and biomass conversion (Fig. 4). Algae cultivation involves a two-stage culture process. The first stage is development of cell biomass during zoospore settlement and the second stage is an enhancement in

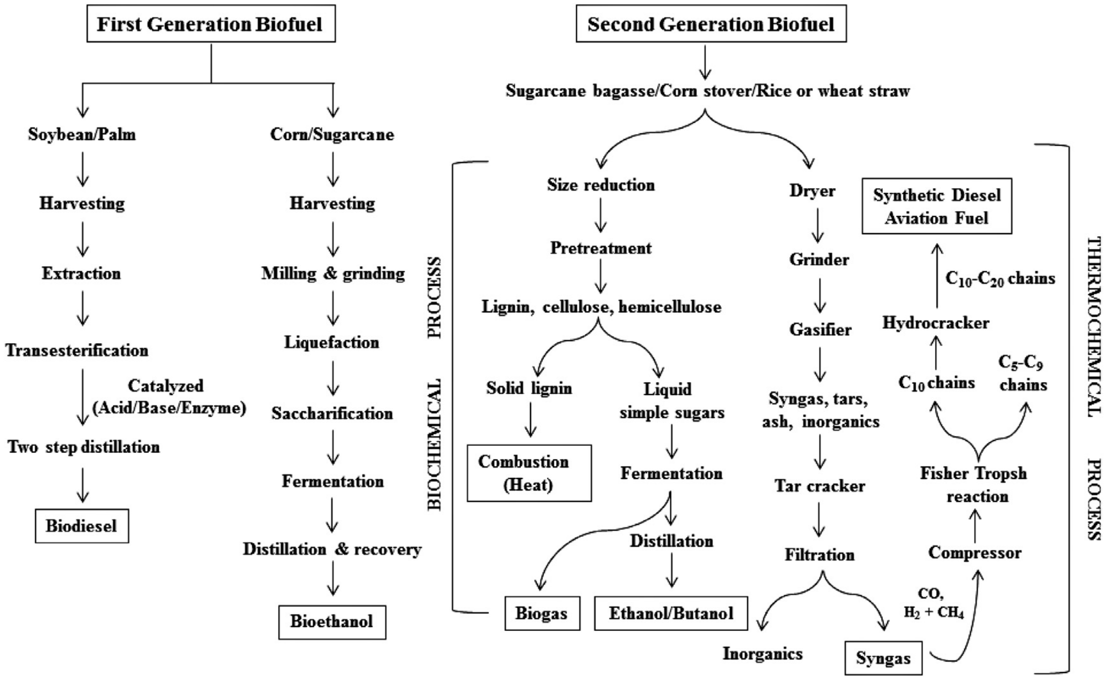

Fig. 3. Technologies involved for production of first and second generation of biofuel.

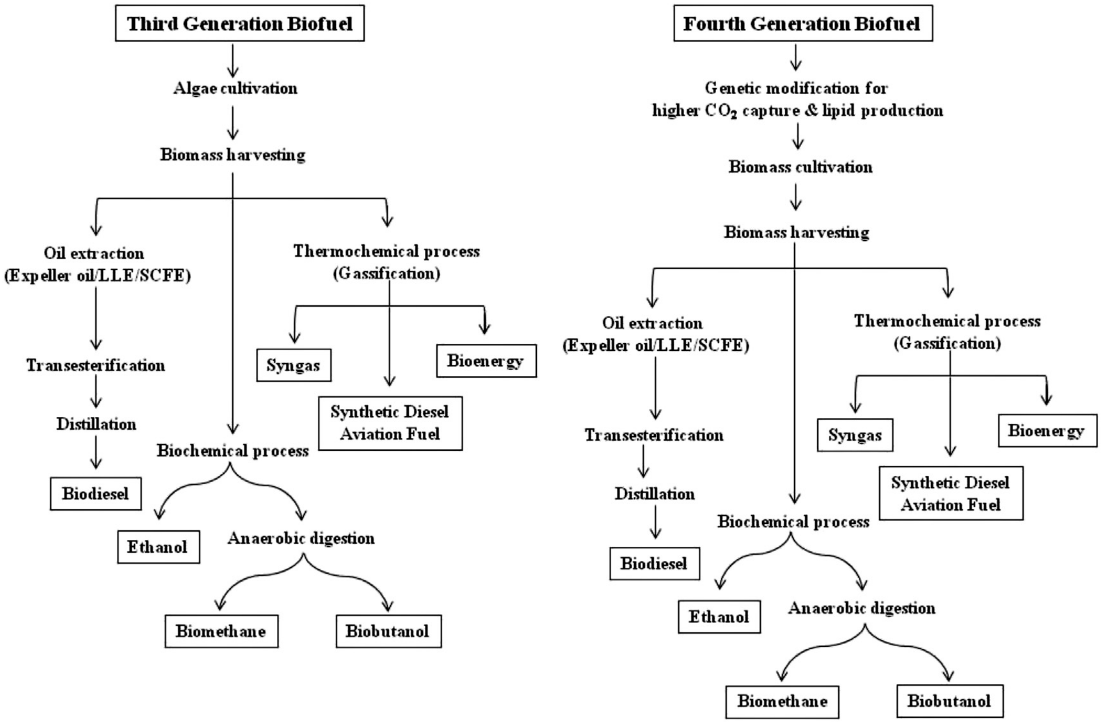

Fig. 4. Technologies involved for production of third and fourth generation of biofuel.

lipid content [27]. Microalgae can be cultured either in an open pond system or using photobioreactor. Initially, open pond cultivation system was more acceptable in terms of commercial point of view as its set-up is easier than photobioreactors. Several companies such as Aurora Biofuel, Green Fuel Technologies, Petro Algae and Seambiotic have reported algae cultivation in open ponds [6]. Recently, algae cultivation in photobioreactors are drawing more attention, as it is easy to control, less prone to contamination and can achieve high productivity.

Harvesting microalgae usually involves flocculation followed by filtration, centrifugation, sedimentation or flotation by ultrasound techniques. Some of these techniques are already in use by several companies whereas some are still under research [6]. Flocculants are needed to aggregate small algal cells to ease the separation process. Several organic (chitosan–polyacrylamide) and inorganic flocculants (combinations of salts and metals such as ferric chloride or alum) are available. Depending on the microalgal cell size and anticipated further separation process (filtration or centrifugation), the appropriate flocculant is selected. Guar gum is a good flocculating agent for small size algae (*Chlorella* sp. CB4 and *Chlamydomonas* sp. CRP7) with negatively charged surface [39].

Magnafloc LT-25, Drewfloc 447, Flocudex CS/5000, Flocusol CM/78, Chemifloc CV/300 and Chitosan were used as flocculants by several researchers for harvesting algae [40,41]. After concentrating the biomass, oil is extracted with or without dewatering process by thermochemical liquefaction, ultrasonic techniques and solvent aided methods. The microalgae can be converted to biofuels by either biochemical or thermochemical conversion process, like previous generation biofuels. Biodiesel, biogas and bioethanol are produced through biochemical conversion process, whereas bio-oil and syngas are produced through thermochemical conversion process. The biodiesel and bio-oil production are affected by lipid content of the algae, however others (bioethanol, biogas and syngas) are not. Therefore, depending on required type of biofuel, microalgae can be selected for cultivation. Such as *Schizochytrium* sp. and *Haematococcus pluvialis* are more suitable for biodiesel production as they are well-known lipid producers. Similarly, *Botryococcus braunii* can be used for bio-hydrogen production as it is well-known hydrogen producer. While, the potential methane producers are *Laminaria* sp., *Gracilaria* sp., *Macrocystis* and *Ulva* sp., and the potential ethanol producers are *Chlamydomonas perigranulata* [42,43] and *Clostridium saccharoperbutylacetonicum* N1–4 [44].

4. Environmental impact through different biofuel generations

The greenhouse gas (GHG) emission from biofuel is not only dependent on the gas coming out from burning fuel but also from the combinational effect of GHG emission at different supply stages such as production of feedstock biomass (fuel used in agriculture, N_2O soil emission from N-fertilizer and residues), transportation to the industrial conversion unit, the industrial unit (a crucial issue is the methodology that is used to include co-products from conversion) and distribution. Three GHGs mostly studied in recent past are CO_2, CH_4, and N_2O, which are converted to CO_2-equivalent by the global warming potential (GWP) recommended by Intergovernmental Panel on Climate Change (IPCC) [45,46]. For fossil fuel, the net GHG emission includes the emissions from mining/extraction, transport, conversion to primary energy carrier, distribution and end use.

4.1. Effect of direct and indirect land use change

The GHG emissions from direct and indirect changes in land use and from changes in (above- and below ground) biomass are well studied [47–50]. Direct land use change occurs when bioenergy crops are cultivated on land that was previously not used for cropland or farming. This includes grasslands, forests, set aside land or degraded land. Indirect land-use change occurs when energy crop production shifts previous activity of the land. Wicke et al. [50] showed that for production of palm oil biodiesel, the land use change is the most critical factor of net GHG emissions, which could have been reduced by 60% if degraded land could have been used instead of converted natural rain forest or peatland. Land use change helps in carbon sequestration in soil to broaden the GHG mitigation by biofuel. As per several reports, conversion of cropland to grassland for growing switchgrass typically increases soil carbon at rates of 0.2–1.0 t carbon/ha per year for several decades [48,49]. Another study carried out by Zan et al. [51] on the carbon storage by three crops – corn, switchgrass and willow showed that corn had higher level above-ground carbon whereas switchgrass had significantly higher root carbon levels below 30 cm of ground.

4.2. Life cycle assessment of biofuel composition and sources

The ethanol engines release less carbon monoxide compared with gasohol (gasoline/ethanol blends) engines and thereby minimize GHG emission [52]. A number of reports are available on reduction in net GHG emission from lignocellulosic ethanol [48,53,54] in comparison to fossil fuel. Spatari et al. [54] reported 57% and 65% reductions in GHG emission for E85-fuel derived from switchgrass ethanol and corn stover ethanol, respectively compared to gasoline. Morey et al. [53] showed that corn stover as a source of fuel for heat and power applications, reduced life-cycle GHG emissions by factors of approximately 8 and 14 compared to natural gas and coal, respectively.

The net GHG emissions vary with biofuel source and are associated with large quantities of by-products formed during biofuel production, so life cycle assessment (LCA) of biofuel production from different sources is necessary to estimate net GHG emissions. The life cycle GHG emissions from different sources of biofuel are shown in Fig. 5 [48,53,55]. The co-products from biofuel can be included in the LCA. LCA studies on lignocellulosic ethanol from switchgrass and corn stover in Canada, using Cradle-to-gate modules for feed process showed that corn stover is more attractive than switchgrass in terms of net GHG emission. A saving of 33% GHG emissions was reported from corn stover-derived ethanol (330 g of CO_2 equivalent/L) compared to switchgrass-derived ethanol (489 g of CO_2 equivalent/L) [54]. Fig. 5 shows that the GHG emission is minimum for corn stover ethanol [53] followed by algal biodiesel [55].

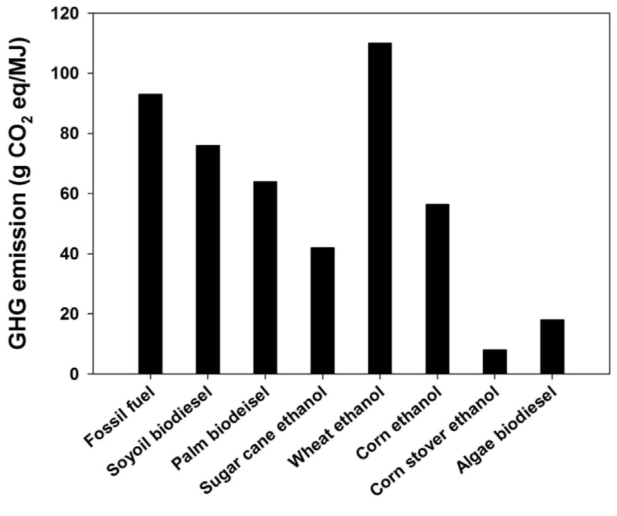

Fig. 5. Life cycle of GHG emission from different sources of biofuel [48,53,55].

Stephenson et al. (2010) [56] reported that a savings of 70–90% in GHG emission was possible with ethanol production from willow, grown in idle land using allocation method of system expansion when compared to that with fossil-derived gasoline. Levelton [57] compared the GHG emissions with E85 made from different feedstocks like corn, switchgrass, corn stover, wheat straw and hay. The authors observed 71% reduction in GHG emission from switchgrass followed by hay (68.3%), corn stover (61%), wheat straw (57%) and corn (38%). MacLean and Spatari [58] concluded that the overall GHG emission from lignocellulosic ethanol including GHG emissions from the production of process chemicals and enzymes used for pretreatment of lignocellulose biomass, is expected to be significantly less than that of corn ethanol and crude oil-derived gasoline/diesel. In a proposed LCA model of ethanol conversion from municipal solid waste, Kalogo et al. [59] showed its better performance in terms of net energy and GHG savings as compared to corn ethanol or cellulosic biomass-ethanol. In case of third and fourth generation biofuels a reduction in GHG emission can be obtained by recycling the CO_2 produced in fermentation process as carbon source for algae cultivation [6]. Collet et al. [60] studied the life cycle of biogas production from the algae *Chlorella vulgaris* and compared the same with first generation biodiesels. They emphasized that high productivity of algae enabled the recycling of CO_2 present in the flue gas. The algae also had the capacity to carry out anaerobic digestion directly, to produce methane and recycle nutrients (N, P and K). Their results suggested that the production of methane from algae can be directly correlated and can mitigate the electric consumption, and further improvements could be realized by decreasing the mixing costs and circulation between different production steps, or by improving the efficiency of the anaerobic process under controlled conditions [60]. In an interesting study, Stephenson et al. [61] suggested algal cultivation (such as *C. vulgaris*) in typical raceway ponds of depth ~0.3 m would be significantly more environmentally sustainable than fossil fuel and other-generation biofuels with future productivity target of 40 tons lipids/ha per year. They also argued that the biodiesel produced from algae cultivated in raceway ponds would have ~80% lower GWP than fossil-derived diesel. The fossil energy requirements and GWP of each of the algal processing steps were found to be significantly lower than that for the cultivation [28].

4.3. Other environmental concerns

Apart from the two main environmental concerns discussed above, biofuel production can directly or indirectly affect water and nutrients resources. Regardless of biofuel generation process, cultivation of biomass feedstock requires water and nutrients. Instead of freshwater, use of seawater or wastewater for biomass cultivation could be a possible alternative. Seawater is widely available but its high salinity limits the use for growing several organisms. First and second generation biofuel feedstocks (plants and crops) are difficult to grow in raw wastewater. However algae, as discussed earlier, can grow by utilizing the nutrients present in wastewater streams and seawater, reducing freshwater demand. Thus algae cultivation provides an integrated system for wastewater treatment or seawater utilization with biofuel production [62].

4.4. Net energy production from the biofuel

The net energy ratio (NER) – MJ of energy produced/MJ consumed, for any biofuel is very important parameter as it indicates the commercial feasibility of the process. The NER from various biofuel sources are presented in Table 3. NER value less than 1 suggested that the process is not commercially feasible. Fore et al.

Table 3
Net Energy Ratio (NER) of different biofuels using different LCA models.

Biofuel source	NER (MJ/MJ)	Model used for data calculation	Reference
Fossil fuel	5.26	The GREET 1.8c model	[55]
Canola biodiesel	1.78	Energy Life Cycle Analysis (ELCA) of small-scale on-farm production	[63]
Soybean biodiesel	2.05	Energy Life Cycle Analysis (ELCA) of small-scale on-farm production	[63]
Rapeseed biodiesel	2.10	CED, PNAS, CONCAWE	[74]
Corn ethanol	0.88–1.22	Energy and Resources Group (ERG) Biofuel Analysis Meta-Model (EBAMM)	[64]
Soyabean biodiesel	2.55	GREET model	[75]
Switchgrass ethanol	1.02–2.62	Energy accounting method	[76]
Cellulosic ethanol	6.70–8.23	Energy and Resources Group (ERG) Biofuel Analysis Meta-Model (EBAMM)	[64]
Switchgrass ethanol	5.79	EBAMM model	[77]
Algae biodiesel	1.1	The GREET 1.8c model	[55]

[63] studied NER of small-scale on-farm production of canola and soybean biodiesel. They observed that soybean biodiesel was energetically more efficient than canola biodiesel due to less nitrogen fertilizer requirement. However, canola was more productive feedstock due to its higher oil content than soybean. Batan et al. [55] compared the net energy use and GHG emissions of algae (*Nanochloropsis*), soybean biofuel and fossil fuel, and observed that the algae biodiesel was 5% better in terms of net GHG emissions compared to soybean biodiesel. In addition, they also observed the NER of the algae biodiesel was 43% lower than the NER of the soybean biodiesel. From Table 3 it can be observed that NER of second generation bioethanol is comparable with the NER of fossil fuel. Farrel et al. [64] obtained highest NER for cellulosic ethanol. Due to the lower NER values (<1), Jorquera et al. [65] commented that horizontal tubular photobioreactors (PBRs) and flat-bed bioreactors to be commercially unfit.

5. Cost effectiveness of biofuel from different generation

The production cost of biofuel is the value of feedstocks over time, plus the calculated conversion cost including capital cost, chemicals and enzymes, energy, operational and maintenance cost, minus the co-product value over time. Among all of these, the production cost is extremely sensitive to biofuel feedstock costs. With the exception of ethanol from sugarcane in Brazil, production costs of all the first generation biofuels in every country are essentially subsidized [34,45]. Simply, higher costs of edible crops make first generation biofuels (excluding Brazil) more expensive. Fast growing demand of feedstocks for biofuel production, has dramatically increased prices for some feedstocks, such as corn in the USA. In this context second generation biofuels are derived from the low cost feedstocks [34]. The costs of different generations of biofuel compared to conventional fuel as projected in different studies are shown in Table 4.

Table 4
Cost of biofuel from different sources compared with conventional fuels.

Type of fuel/biofuel	Price ($/Gallon)	Reference
Gasoline	3.37	[78]
Diesel	3.46	[78]
CNG[a]	2.13	[78]
Ethanol (E85)	3.14	[78]
Biodiesel (B20)	3.95	[78]
Biodiesel (B99/100)	4.2	[78]
Corn ethanol	2.12	[72]
Switchgrass ethanol	4.53	[72]
Miscanthus ethanol	2.74	[72]
Corn stover ethanol	2.62	[66]
Algae biodiesel	9–40	[6]

[a] Conventional natural gas.

The minimum cost was projected for corn ethanol [66] while the maximum was for algal biodiesel. Larson [34] reported that the capital cost per unit of production capacity decrease with increasing plant size for relatively large plants. Usually it is seen that this cost reduction is sufficient to compensate the increased biomass costs that arise from longer average transportation distances associated with the larger scale of production. The large scale economies are more significant for thermochemical process rather than biochemical conversion, but for both cases challenges need to be faced for commercial production [34]. Ajanovic and Hass [67] reported that under current policy conditions mainly exemption from excise taxes, the economic prospects of first generation biofuels in Europe are rather promising, but the major problems of this generation biofuels are lack of land availability for feedstocks and the modest ecological performance. The commercial production of first and second generation biodiesel is practiced in many countries (Table 5).

Kovacevic and Wesseler [68] compared the cost effectiveness of algae and rapeseed biodiesel. Biomass production costs dominate for both algae and rapeseed biodiesel. Carbon supply, harvesting and water supply are major factors of algal biomass production cost. Though the land cost is low for algae biomass production, infrastructure and mixing costs are higher. However, in case of algae, the high cost of energy recovery counteracts large part of the benefit. Impact of food prices and GHG emissions are the most prominent costs in case of the rapeseed biodiesel. Browne et al. [69] studied the cost efficiency of biomethane production from three different sources namely organic fraction of municipal solid waste (OFMSW), slaughter house waste (SHW) and grass and slurry. Among the three sources, OFMSW is the cheapest one (€ 0.36/L and € 1.35/L diesel equivalent with and without gate fee, respectively, including value added tax (VAT) of 21%), followed by SHW (€0.65/L diesel equivalent), and grass and slurry (€1.40/L diesel equivalent (lde)). Though still more costly, biomethane produced from grass and slurry is within the price range of petroleum derived transport fuels at the service stations (diesel and petrol from €1.38 to 1.45/L in February 2011). Using biomethane is advantageous over liquid fuel, as a saving of about 75% of CO_2 emissions can be realized [69]. As on Jan 2007, estimated by IEA [70] (International Energy agency) the cost of sugarcane ethanol in Brazil is $0.30/L gasoline equivalent (lge) and the cost of ethanol from maize, sugar beet and wheat are in range of $0.6–$0.8/lge excluding subsidies whereas the cost of lignocellulosic ethanol is around $1.0/lge at the pilot scale. Currently animal fat is the cheapest ($0.4–$0.5/lde) source for biodiesel while the cost for traditional transesterification of vegetable oil is at present around $0.6–$0.8/lde. Further cost reductions (of $0.1–$0.3/lde) from economies of scale are expected for new processes. The cost of BTL (Biomass to Liquid) diesel from lignocellulose is more than $0.9/lde (feedstock $3.6/GJ), with a potential reduction to $0.7–$0.8/lde [70]. According to the recent report (2013) by IEA [71] the costs of cane ethanol, conventional biodiesel, advanced ethanol and biodiesel are ~$0.6/lge, ~$1/lge and ~$1.1/lge, respectively. This showed that in spite of significant efforts, the cost of biofuel has not significantly reduced in these years (2007–2013). An improved economy of conversion process may enable biofuels to compete with fossil fuel in long-run [71]. Khanna et al.

Table 5
Biofuels at different commercializing level.

Generation	Type of biofuel	Countries producing	Commercial stage	Reference
1st generation	Corn ethanol	US	Commercialized	[2]
1st generation	Sugarcane ethanol	Brazil	Commercialized	[2]
1st generation	Biodiesel (rapeseed, soyabean)	European countries, (Germany, Belgium, France, Netherland, Polanetc)	Commercialized	[2]
1st generation	Palm oil biodiesel	Malaysia, Indonesia	Commercialized	[2]
2nd generation	Cellulosic ethanol	US	Under research level	[2]
3rd generation	Algal biodiesel	New Zealand, Spain	Under research level	[6]

[72] gave a more optimistic estimation of second generation biofuels (switchgrass, *Miscanthus* and corn stover ethanol) costs which are comparable or lower than conventional fuel (Table 4) including co-products credit. On the basis of several available reports, Singh and Gu [6] estimated the costs of algal oils and algal biodiesel production between \$9–\$25 and \$15–\$40 per gallon in ponds and photobioreactors (PBRs), respectively. This high cost is primarily due to complex composite algae production systems. Singh and Gu [6] suggested a reduction in the number of steps in algal biofuel production as well as co-production of some more valuable fraction would significantly reduce the algal biofuel cost.

6. Conclusions

In future, biofuel will definitely have an important role in meeting the world's energy need. In this review article, four generations of biofuel are reviewed in terms of their biomass feedstocks, conversion technologies, environmental impacts and economic performance. Each generation of biofuel has come with its own set of advantages and challenges. To meet the growing demand of energy the uninterrupted supply of raw material is the primary requirement. For a country to be self-sufficient in raw oil supply to frame a biorefinery, the major biofuel feedstock needs to be produced within the country without compromising food supply. The availability and production of biofuel feedstock depend on several factors like geographical location, the economic condition of population and food-fuel demand. This makes the first generation biofuel unfit for replacing fossil fuel as it arouses the controversy of food-fuel competition. In this context third and fourth generations of biofuel are more suitable as they do not create such food-fuel competition. Another most important key to make economically and environmentally sustainable biofuel is the advancement in technology for biofuel production. For profitable commercial production of biofuel, cost effective and high yield conversion systems need to be established. The improvement in conversion yield may also be obtained by employing metabolic engineering tools. Metabolic engineering may be used to modify the existing biological pathways to improve biofuel both quantitatively and qualitatively. This tool also holds an important role either in the modification of feedstock or improving the microbes to obtain greater conversion. More researches are required to accomplish high yield and cost-effective conversion processes. For GHG mitigation second and third generations of biofuel have been proved to be superior to first generation biofuel. Further improvement in GHG mitigation can be envisaged from fourth generation of biofuel.

In terms of economic performance, at present first generation biofuel is projected as the most cost effective fuel. However, production capacity of first generation biofuel is limited to certain countries only as its production is highly land intensive. Whereas in case of second and third generation biofuel, the current costs are still high, mainly because of the high investment cost and low conversion efficiencies of feedstock into biofuel. Further optimization and improvements of conversion technology can make second and third generations bioethanol and biodiesel production more promising and economic. Overall the present review summarizes that the future biofuel may be a combination of some or all of the four generations of biofuel rather than only one generation.

References

[1] Graham-Rowe D. Beyond food versus fuel. Nature 2011;474:S6–8.

[2] OECD/IEA. From 1st to 2nd generations of biofuel technologies, overview of current industry and RD&D activities; 2008.

[3] Sims REH, Mabee W, Saddler JN, Taylor M. An overview of second generation biofuel technologies. Bioresour Technol 2010;101:1570–80.

[4] Schenk P, Thomas-Hall S, Stephens E, Marx U, Mussgnug J, Posten C. Second generation biofuels: high-efficiency microalgae for biodiesel production. Bioenergy Res 2008;1(1):20–43.

[5] Chisti Y. Biodiesel from microalgae. Biotechnol Adv 2007;25(3):294–306.

[6] Singh J, Gu S. Commercialization potential of microalgae for biofuels production. Renew Sust Energ Rev 2010;14:2596–610.

[7] Lu J, Sheahan C, Fu P. Metabolic engineering of algae for fourth generation biofuels production. Energy Environ Sci 2011;4:2451–66.

[8] Hayashida S, Ohta K, Flor PQ, Na N. High concentration-ethanol fermentation of raw ground corn. Agric Biol Chem 1982;46(7):1947–50.

[9] Rolz C, de León R. Ethanol fermentation from sugarcane at different maturities. Ind Crop Prod 2011;33(2):333–7.

[10] Nabi MN, Rahman MM, Akhter MS. Biodiesel from cotton seed oil and its effect on engine performance and exhaust emissions. Appl Therm Eng 2009;29:2265–70.

[11] Singh SP, Singh D. Biodiesel production through the use of different sources and characterization of oils and their esters as the substitute of diesel: a review. Renew Sust Energ Rev 2010;14:200–16.

[12] Nam LTH, Vinh TQ, Loan NTT, Tho VDS, Yang XY, Su BL. Preparation of biofuels by catalytic cracking reaction of vegetable oil sludge. Fuel 2011;90:1069–75.

[13] Balat M, Balat H, Oz C. Progress in bioethanol processing. Prog Energy Combust Sci 2008;34(5):551–73.

[14] Chen Y, Dong B, Qin W, Xiao D. Xylose and cellulose fractionation from corncob with three different strategies and separate fermentation of them to bioethanol. Bioresour Technol 2010;101(18):6994–9.

[15] Choi IS, Wi SG, Kim SB, Bae HJ. Conversion of coffee residue waste into bioethanol with using popping pretreatment. Bioresour Technol 2012;125:132–7.

[16] Dubey AK, Gupta PK, Garg N, Naithani S. Bioethanol production from waste paper acid pretreated hydrolyzate with xylose fermenting *Pichia stipites*. Carbohydr Polym 2012;88(3):825–9.

[17] Ruangviriyachai C, Niwaswong C, Kosaikanon N, Chanthai S, Chaimart P. Pineapple peel waste for bioethanol production. J Biotechnol 2010;150S:S10.

[18] Tye YY, Lee KT, Abdullah WNW, Leh CP. Potential of *Ceiba pentandra* (L.) Gaertn. (kapok fiber) as a resource for second generation bioethanol: effect of various simple pretreatment methods on sugar production. Bioresour Technol 2012;116:536–9.

[19] Kaparaju P, Serrano M, Thomsen AB, Kongjan P, Angelidaki I. Bioethanol, biohydrogen and biogas production from wheat straw in a biorefinery concept. Bioresour Technol 2009;100(9):2562–8.

[20] Kalbande SR, More GR, Nadre RG. Biodiesel production from non-edible oils of Jatropha and karanj for utilization in electrical generator. Bioenergy Res 2008;1(2):170–8.

[21] Deshpande NV, Kale NW, Deshmukh SJ. A study on biogas generation from Mahua (*Madhuca indica*) and Hingan (*Balanites aegyaptiaca*) oil seedcake. Energy Sustain Dev 2012;16(3):363–7.

[22] Li Z, Liu Y, Liao W, Chen S, Zemetra RS. Bioethanol production using genetically modified and mutant wheat and barley straws. Biomass Bioenergy 2011;35(1):542–8.

[23] Chen WH, Chen YC, Lin JG. Evaluation of biobutanol production from non-pretreated rice straw hydrolysate under non-sterile environmental conditions. Bioresour Technol 2013;135:262–8.

[24] Yua M, Zhanga Y, Tangb IC, Yanga ST. Metabolic engineering of *Clostridium tyrobutyricum* for n-butanol production. Metab Eng 2011;13(4):373–82.

[25] Zhang F, Rodriguez S, Keasling JD. Metabolic engineering of microbial pathways for advanced biofuels production. Curr Opin Biotechnol 2011;22: 775–83.

[26] Shen CR, Liao JC. Metabolic engineering of Escherichia coli for 1-butanol and 1-propanol production via the keto-acid pathways. Metab Eng 2008;10(6): 312–20.

[27] Suali E, Sarbatly R. Conversion of microalgae to biofuel. Renew Sust Energ Rev 2012;16:4316–42.

[28] Gouveia L. Microalgae as a feedstock for biofuelsIn Springer briefs in microbiology; 2011. http://dx.doi.org/10.1007/978-3-642-17997-6-1.

[29] Rodolfi L, Zittelli GC, Bassi N, Padovani G, Biondi N, Bonini G. Microalgae for oil: strain selection, induction of lipid synthesis and outdoor mass cultivation in a low-cost photobioreactor. Biotechnol Bioeng 2008;102(1):100–12.

[30] Radakovits R, Jinkerson RE, Darzins A, Posewitz MC. Genetic engineering of algae for enhanced biofuel production. Eukaryot Cell 2010;9:486–501.

[31] Li Y, Han D, Hu G, Dauvillee D, Sommerfeld M, Ball S, et al. *Chlamydomonas* starchless mutant defective in ADP-glucose pyrophosphorylase hyper-accumulates triacylglycerol. Metab Eng 2010;12(4):387–91.

[32] Lan EI, Liao JC. Metabolic engineering of cyanobacteria for 1-butanol production from carbon dioxide. Metab Eng 2011;13(4):353–63.

[33] Dutta K, Sen S, Dasu VV. Production, characterization and applications of microbial cutinases. Process Biochem 2009;44:127–34.

[34] Van Gerpen J, Shanks B, Pruszko R, Clements D, Knothe G. Biodiesel production technology August 2002–January 2004, National Renewable Energy Laboratory, subcontractor report.

[35] Larson ED. Biofuel production technologies: status, prospects and implications for trade and development. New York: United Nations Conference on Trade and Development; 2007. UNCTAD/DITC/TED/2007/10.

[36] FitzPatrick M, Champagne P, Cunningham MF, Whitney RA. A biorefinery processing perspective: treatment of lignocellulosic materials for the production of value-added products. Bioresour Technol 2010;101:8915–22.

[37] French R, Czernik S. Catalytic pyrolysis of biomass for biofuels production. Fuel Process Technol 2010;91:25–32.

[38] Yin C. Microwave-assisted pyrolysis of biomass for liquid biofuels production. Bioresour Technol 2012;120:273–84.

[39] Banerjee C, Ghosh S, Sen G, Mishra S, Shukla P, Bandopadhyay R. Study of algal biomass harvesting using cationic guar gum from the natural plant source as flocculant. Carbohydr Polym 2013;92(1):675–81.

[40] de Godos I, Guzman HO, Soto R, García-Encina PA, Becares E, Muñoz R, et al. Coagulation/flocculation-based removal of algal–bacterial biomass from piggery wastewater treatment. Bioresour Technol 2011;102(2):923–7.

[41] Schlesinger A, Eisenstadt D, Bar-Gil A, Carmely H, Einbinder S, Gressel J. Inexpensive non-toxic flocculation of microalgae contradicts theories; overcoming a major hurdle to bulk algal production. Biotechnol Adv 2012;30(5): 1023–30.

[42] Hon-Nami K. A unique feature of hydrogen recovery in endogenous starch-to-alcohol fermentation of the marine microalga, *Chlamydomonas perigranulata*. Appl Biochem Biotechnol 2006;131:808–28.

[43] Harun R, Singh M, Forde GM, Danquah MK. Bioprocess engineering of microalgae to produce a variety of consumer products. Renew Sust Energ Rev 2010;14:1037–47.

[44] Ellis JT, Hengge NN, Sims RC, Miller CD. Acetone, butanol, and ethanol production from wastewater algae. Bioresour Technol 2012;111:491–5.

[45] Climate change. Mitigation of climate change contribution of working group III to the fourth assessment report of the intergovernmental panel on climate change. Cambridge, UK: Intergovernmental Panel on Climate Change; 2007.http://www.ipcc.ch/ipccreports/index.htm [accessed 28.07.10].

[46] Rodriguez MFC, Nebra S. Assessing GHG emissions, ecological footprint, and water linkage for different fuels. Environ Sci Technol 2010;44:9252–7.

[47] Hoefnagels R, Smeets E, Faaij A. Greenhouse gas footprints of different biofuel production systems. Renew Sust Energ Rev 2010;14:1661–94.

[48] Cherubini F, Bird ND, Cowie A, Jungmeier G, Schlamadinger B, Woess-Gallasch S. Energy- and greenhouse gas-based LCA of biofuel and bioenergy systems: key issues, ranges and recommendations. Resour Conserv Recycl 2009;53:434–47.

[49] Conant RT, Paustian K, Elliot ET. Grassland management and conversion into grassland: effects on soil carbon. Ecol Appl 2001;11:343–55.

[50] Wicke B, Dornburg V, Junginger M, Faaij A. Different palm oil production systems for energy purposes and their greenhouse gas implications. Biomass Bioenergy 2008;32:1322–37.

[51] Zan CS, Fyles JW, Girouard P, Samson RA. Carbon sequestration in perennial bioenergy, annual corn and uncultivated systems in Southern Quebec. Agric Ecosyst Environ 2001;86:135–44.

[52] Macedo IC, Leal MRLV, Da Silva JEAR. Assessment of greenhouse gas emissions in the production and use of fuel ethanol in Brazil. Brazil: Secretariat of the Environment of the State of Sao Paulo; 2004. pp. 32–7.

[53] Morey RV, Kaliyan N, Tiffany DG, Schmidt DR. A corn stover supply logistics system. Appl Eng Agr 2010;26(3):455–61.

[54] Spatari S, Zhang Y, Maclean HL. Life cycle assessment of switchgrass- and corn stover-derived ethanol-fueled automobiles. Environ Sci Technol 2005;39: 9750–8.

[55] Batan L, Quinn J, Willson B, Bradley T. Net energy and greenhouse gas emission evaluation of biodiesel derived from microalgae. Environ Sci Technol 2010;44:7975–80.

[56] Stephenson AL, Dupree P, Scott SA, Dennis JS. The environmental and economic sustainability of potential bioethanol from willow in the UK. Bioresour Technol 2010;101:9612–23.

[57] Levelton. Assessment of net emissions of greenhouse gases from ethanol blended gasolines in Canada: lignocellulosic feedstocks. Levelton Engineering, Ltd. and (S&T) 2 Consultants; 2000. Report R-2000-2.

[58] MacLean HL, Spatari S. The contribution of enzymes and process chemicals to the life cycle of ethanol. Environ Res Lett 2009;4:014001. http://dx.doi.org/10.1088/1748-9326/4/1/014001.

[59] Kalogo Y, Habibi S, Maclean HL, Joshi SV. Environmental implications of municipal solid waste-derived ethanol. Environ Sci Technol 2007;41:35–41.

[60] Collet P, Hélias-Arnaud A, Lardon L, Ras M, Goy RA, Steyer JP. Life-cycle assessment of microalgae culture coupled to biogas production. Bioresour Technol 2011;102:207–14.

[61] Stephenson AL, Kazamia E, Dennis JS, Howe CJ, Scott SA, Smith AG. Life-cycle assessment of potential algal biodiesel production in the United Kingdom: a comparison of raceways and air-lift tubular bioreactors. Energy Fuels 2010;24:4062–77.

[62] Dahiya A, Todd J, McInnis A. Wastewater treatment integrated with algae production for biofuel. In: Gordon R, Seckbach J, editors. The Science of algal fuels. Cellular Origin, life in Extreme Habitats and Astrobiology, 447–466; 2012. http://dx.doi.org/10.1007/978-94-007-5110-1_24.

[63] Fore SR, Porter P, Lazarus W. Net energy balance of small-scale on-farm biodiesel production from canola and soybean. Biomass Bioenergy 2011;35: 2234–44.

[64] Farrell AE, Plevin RJ, Turner BT, Jones AD, O'Hare M, Kammen DM. Ethanol can contribute to energy and environmental goals. Science 2006;311:506–8.

[65] Jorquera O, Kiperstok A, Sales EA, Embiruçu M, Ghirardi ML. Comparative energy life-cycle analyses of microalgal biomass production in open ponds and photobioreactors. Bioresour Technol 2010;101:1406–13.

[66] Khanna M, Dhungana B. Economics of alternative feedstocks in corn–based ethanol in Illinois and the U.S.: a report from Department of Agricultural and Consumer Economics, University of Illinois. Urbana: University of Illinois; 2007.

[67] Ajanovic A, Haas R. Economic challenges for the future relevance of biofuels in transport in EU countries. Energy 2010;35:3340–8.

[68] Kovacevic V, Wesseler J. Cost-effectiveness analysis of algae energy production in the EU. Energy Policy 2010;38:5749–57.

[69] Browne J, Nizamia AS, Thamsiriroja T, Murphya JD. Assessing the cost of biofuel production with increasing penetration of the transport fuel market: a case study of gaseous biomethane in Ireland. Renew Sust Energ Rev 2011;15: 4537–47.

[70] IEA energy technology essentials, biofuel production; January 2007.

[71] IEA. Tracking clean energy progress 2013. IEA input to the clean energy ministerial; 2013.

[72] Khanna M, Dhungana B, Clifton-Brown J. Costs of producing switchgrass and miscanthus for bioenergy in Illinois. Biomass Bioenergy 2008;32(6):482–93.

[73] Ajanovic A, Haas R. The future relevance of alternative energy carriers in Austria. In: 5th International Ege Energy Symposium and Exhibition (IEESE-5) 27-30 June. Denizli, Turkey: Pamukkale University; 2010.

[74] Kalnes TN, Koers KP, Marker T, Shonnard DR. A technoeconomic and environmental life cycle comparison of green diesel to biodiesel and syndiesel. Environ Prog Sustain Energy 2009;28:111–20.

[75] Pradhan A, Shrestha DS, Van Gerpen J, Duffield J. The energy balance of soybean oil biodiesel production: a review of past studies. Am Soc Agric Biol Eng 2008;51:185–94.

[76] Felix E, Tilley DR. Integrated energy, environmental and financial analysis of ethanol production from cellulosic switchgrass. Energy 2009;34:410–36.

[77] Schmer MR, Vogel KP, Mitchell RB, Perrin RK. Net energy of cellulosic ethanol from switchgrass. Proc Natl Acad Sci 2008;105:464–9.

[78] Clean cities alternative fuel price report. US Department of Energy; Jan 2012.