Review Paper

Meeting the U.S. renewable fuel standard: a comparison of biofuel pathways
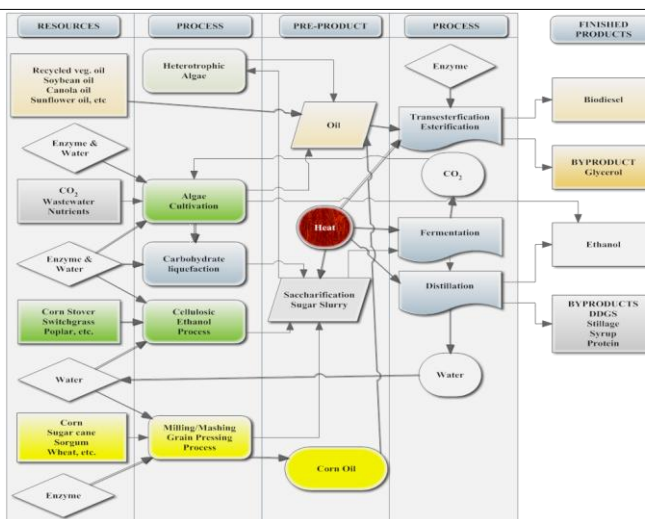
Marc Y. Menetrez

United State Environmental Protection Agency, Office of Research and Development, National Risk Management Research Laboratory, Air Pollution Prevention and Control Division, Research Triangle Park, NC 27711, USA.

HIGHLIGHTS

➢ *The year 2014 will see a significant increase in the U.S. production of cellulosic ethanol to a total of 104 Mgal/yr (394 x10³ m³/yr) when three new plants are brought under full operation.*

➢ *The accomplished increase in U.S. cellulosic ethanol production although significant, will remain far below the mandated 2014 Renewable Fuel Standard of 1.75 Bgal/yr (6.6 x10⁶ m³/yr).*

➢ *Potential pathways for producing biofuels from algae were evaluated and compared for their feedstock and footprint demands, as well as productivity potential.*

➢ *The U.S. commercial production of renewable fuels is increasingly behind levels mandated by the Renewable Fuel Standard. Algae based fuels could be developed to help fill this growing gap.*

GRAPHICAL ABSTRACT

ARTICLE INFO

ABSTRACT

The production of renewable energy is undergoing rapid development. Ethanol primarily derived from corn and biodiesel made from recycled cooking oil and agricultural grains are established sources of renewable transportation fuel. Cellulosic ethanol production is increasing substantially, but at a rate below expectations. If future renewable fuel projections are to be accomplished, additional sources will be needed. Ideally, these sources should be independent of competing feedstock use such as food grains, and require a minimal footprint. Although the uses of algae seem promising, a number of demonstrations have not been economically successful in today's market. This paper identifies efforts being conducted on ethanol and biodiesel production and how algae might contribute to the production of biofuel in the United States. Additionally, the feedstock and land requirements of existing biofuel pathways are compared and discussed.

* Corresponding author at: Tel.: +1 919 541 7981
E-mail address: menetrez.marc@epa.gov

1. Introduction

The 2007 Energy Independence and Security Act (EISA) (H.R. 6) and the 2005 federal renewable fuel standard (RFS) stipulates that, by 2022, the United States must produce 15 billion gallons per year (Bgal/yr) or 56.8 million cubic meters ($x10^6$ m³/yr) of corn-based ethanol (CBE), 16 Bgal/yr (60.6 Mm³/yr) of cellulosic biofuels, 1 Bgal/yr ($3.8 x10^6$ m³/yr) of biodiesel, and 4 Bgal/yr ($15.1 x10^6$ m³/yr) of advanced biofuels (other than corn-based ethanol) (Environmental News Service, 2011; Public Law 110–140, 2007; RFS Renewable Fuels Association, 2012). Achieving a total production goal of 36 Bgal/yr ($136 x10^6$ m³/yr) of renewable fuel by 2022 is a substantial challenge, largely due to the established RFS requirement for cellulosic ethanol (see Table 1). However, U.S. biofuel production is substantial and growing.

Table 1.
Renewable Fuel Projections (Public Law 110–140, 2007; Thompson et al., 2010; Environmental News Service, 2011; RFS Renewable Fuels Association, 2012).

Year	Corn-based Ethanol (CBE) Bgal ($x10^6$ m³/yr)	Cellulosic Ethanol Bgal ($x10^6$ m³/yr)	Biodiesel [a] Bgal ($x10^6$ m³/yr)	Advanced Biofuel Bgal ($x10^6$ m³/yr)	Total RFS Bgal ($x10^6$ m³/yr)	Total biofuel (minus CBE) Bgal ($x10^6$ m³/yr)
2013	13.8 (52.2)	1.0 (3.8)	1.0 (3.8)	0.75 (2.8)	16.6 (62.6)	2.8 (10.4)
2014	14.4 (54.5)	1.8 (6.6)	1.0 (3.8)	1.0 (3.8)	18.2 (68.7)	3.8 (14.2)
2015	15.0 (56.8)	3.0 (11.4)	1.0 (3.8)	1.5 (5.7)	20.5 (77.6)	5.5 (20.8)
2016	15.0 (56.8)	4.2 (15.9)	1.0 (3.8)	2.0 (7.6)	22.3 (84.2)	7.3 (27.4)
2017	15.0 (56.8)	5.5 (20.8)	1.0 (3.8)	2.5 (9.5)	24.0 (90.8)	9.0 (34.1)
2018	15.0 (56.8)	7.0 (26.5)	1.0 (3.8)	3.0 (11.4)	26.0 (98.4)	11.0 (41.6)
2019	15.0 (56.8)	8.5 (32.2)	1.0 (3.8)	3.5 (13.2)	28.0 (106.0)	13.0 (49.2)
2020	15.0 (56.8)	10.5 (39.7)	1.0 (3.8)	3.5 (13.2)	30.0 (113.6)	15.0 (56.8)
2021	15.0 (56.8)	13.5 (51.1)	1.0 (3.8)	3.5 (13.2)	33.0 (124.9)	18.0 (66.1)
2022	15.0 (56.8)	16.0 (60.6)	1.0 (3.8)	4.0 (15.1)	36.0 (136.3)	21.0 (79.5)

[a] EISA is not specific on biodiesel after 2012.

The RFS projected production of multiple renewable fuels listed in Table 1 establishes guidance in consideration of environmental consequences while advocating fuel production practices with lower associated green-house gas (GHG) emission.

2. Meeting the RFS challenge for cellulosic ethanol - current to future production

As of 2014, the production of CBE (from more than 200 ethanol plants) was 15 Bgal/yr ($56.8 x10^6$ m³/yr) (equal to the 2022 RFS) (Parker, 2012a; Ethanol Producers Digest, 2013). The majority of this ethanol (or approximately 13.3 Bgal [$50.3 x10^6$ m³/yr]) was used for the 10% additive to gasoline (Parker, 2012a). Biodiesel production, largely from yellow grease (recycled vegetable oil), soy bean oil, and canola oil, was 1.0 Bgal/yr ($3.8 x10^6$ m³/yr) (equal to the 2022 RFS), from more than 200 biodiesel plants (U.S. Biodiesel Digest, 2011; U.S. Energy Information Administration, 2012a; Biodiesel Industry Directory, 2013). Cellulosic ethanol plant production capacity is currently 20 million gallons per year ($75.7 x10^3$ m³/yr), as listed in Table 2. Thus, production is already at or above the 2022 RFS levels for biodiesel and CBE biofuels (U.S. Energy Information Administration, 2012a; Ethanol Producers Digest, 2013).

As of early 2014, there were twelve commercial cellulosic ethanol plants in operation, and they have a combined capacity of producing a total of 20 Mgal/yr ($75.7 x10^3$ m³/yr), (listed in Table 2), more than triple the 2013 capacity of 6.25 Mgal/yr ($23.7 x10^3$ m³/yr) (Ethanol Producers Digest, 2013). As listed in Table 2, the Indian River Bioenergy Plant has the capability of producing 8 Mgal/yr ($30.3 x10^3$ m³/yr) (as well as 5 megawatts of electric power), and the Fiberight Blairstown Plant can produce 6 Mgal/yr ($22.7 x10^3$ m³/yr) however, the other ten facilities are demonstration pilot plants, with a production limit of less than 1.4 Mgal/yr ($5.3 x10^3$ m³/yr) (Shaffer, 2012; Ethanol Producers Digest, 2013). These demonstration projects have proven that their technology is successful. The RFS challenge (listed in Table 1) to

produce 1.75Bgal/yr ($6.6 x10^6$ m³/yr) of cellulosic ethanol in 2014 is unlikely, and the possibility of meeting future RFS demands seems increasingly distant.

Five major cellulosic ethanol plants, listed in Table 2, are currently under construction that when in operation will produce in excess of 104 Mgal/yr ($393.7x10^3$ m³/yr). The Poet and Royal plant in Emmetsburg, Iowa is referred to as 'Project Liberty' and is located near other Poet CBE plants (Shaffer, 2012). The plant was constructed at a cost of $250 million, will produce 20 to 28 Mgal/yr (75.7 to $106 x10^3$ m³/yr) from corn-stover, and is expected to be operational in 2014 (Shaffer, 2012). Poet is an established ethanol manufacturer that owns and operates 27 CBE plants in the U.S. (Shaffer, 2012). The successful operation of the Project Liberty plant and the established availability of corn stover could lead to further expansion and additional plants. (Poet is also currently extracting 250,000 t/yr [$227 x10^6$ kg/yr] or 68 Mgal/yr [$257 x10^3$ m³/yr], of corn oil from 25 equipped CBE plants [Biodiesel Magazine, 2013; Ethanol Producers Digest, 2013].)

As mentioned, in addition to the Project Liberty plant that will be completed this year, there are four other major projects underway, i.e. Abengoa Bioenergy's cellulosic ethanol plant using corn stover in Kansas; Blue Fire Renewables, LLC.'s plant, in Mississippi (that will use wood waste); Dupont Danisco's plant, in Iowa that will use corn stover; and Enerkem Alberta's plant, in Alberta Canada that will use municipal solid waste (Shaffer, 2012; Ethanol Producers Digest, 2013). Table 2 lists the commercial cellulosic ethanol plants and their stages of construction/operation. There are three stages: (1) "Existing Plants", consisting of nine existing plants in intermittent operation with a combined capacity of 20 Mgal/yr ($75.7x10^3$ m³/yr); (2) "Under Construction" consisting of nine plants with a combined capacity of 104 Mgal/yr ($394 x10^3$ m³/yr); and; (3) "Proposed Plants" consisting of 16 proposed plants with a combined capacity of 368 Mgal/yr ($1.4 x10^6$ m³/yr) (Shaffer, 2012; Ethanol Producers Digest, 2013). It should be understood that the existing plants are demonstration plants, many of which may not maintain production on a continuous basis since their purpose may not be to produce ethanol economically. These small scale plants are used to study the details of the process, demonstrate the feasibility of the technology and conduct process optimization studies. An acceptable level of proof of the viability and sustainability of a major cellulosic ethanol plant (producing a minimum of 10 Mgal/yr [$37.9x10^3$ m³]) entails the successful operation of a year or more, with an acceptable level of profit.

Based on data compiled by the U.S. Energy Information Administration and reported in Bloomberg News, the cellulosic ethanol industries will increase production significantly in 2014 due to refinery startups (Shaffer, 2012; U.S. Energy Information Administration, 2012a; Ethanol Producers Digest, 2013). The 'Ethanol Producers Digest' has estimated that the 2013 level of 20 Mgal/yr ($75.7x10^3$ m³/yr) will increase to a total of 104 Mgal/yr ($394 x10^3$ m³/yr) when plants that already are under construction are brought into operation (Shaffer, 2012; Ethanol Producers Digest, 2013). Even if these optimistic predictions for cellulosic ethanol production are achieved, production still will remain far below the 2014 RFS of 1.75 Bgal/yr ($6.6 x10^6$ m³/yr) (Herndon, 2012).

The total capacity of all three stages of cellulosic ethanol plants listed in Table 2 (Existing 20 MMgal/yr ($75.7x10^3$ m³/yr), Under Construction 104 Mgal/yr ($394 x10^3$ m³/yr), and Proposed Plants 368 Mgal/yr ($1.4 x10^6$ m³/yr]) would yield an estimated maximum production rate total of 492 Mgal/yr ($1.9 x10^6$ m³/yr). As a theoretical exercise, the production rate of 492 Mgal/yr ($1.9 x10^6$ m³/yr) of cellulosic ethanol if attained by 2017, would greatly improve U.S. capacity, however, it would make up only 11% of the 5.5 Bgal/yr ($20.8 x10^6$ m³/yr), which is specified in the 2017 RFS listed in Table 1. Additionally, until an established cellulosic ethanol plant has been fully operational, production costs, operational and maintenance requirements, and environmental impacts will not be known (Herndon, 2012; Shaffer, 2012; Ethanol Producers Digest, 2013).

The cellulosic ethanol production requirement of the RFS presents a great commercial challenge. This is largely due to the fact that the development of a new industry based on cutting-edge research is a complex matter, and the associated timelines are difficult to predict. Additionally, the cost of an average size cellulosic ethanol plant of 20 to 25 Mgal/yr (75.7 $x10^3$ to $94.6x10^3$ m³/yr), can range from $200 to $250 million, a daunting investment (Menetrez, 2012). Still, the RFS production standard represents an important national goal that encourages the development of additional sources of renewable ethanol and biodiesel. While the cellulosic ethanol industry

Table 2.
Renewable Fuel Projections (Public Law 110–140, 2007; Thompson et al., 2010; Environmental News Service, 2011; RFS Renewable Fuels Association, 2012).

Plants	Location	Feedstock		Capacity Mgal/yr (x10⁶m³/yr)
Existing Plants				
American Process Inc/Alpena Biorefinery	Alpena MI	Wood Sugars		0.8 (3.0)
BP Biofuels Demonstration Plant,	Jennings LA	Energy Grasses		1.4 (5.3)
DupontDanisco Cellulosic Ethanol LLC	Vonore TN	Corn Stover, Switchgrass		0.3 (1.0)
Enerkem Inc.	Westbury QC	Treated Wood		1.0 (3.8)
Fiberight Demonstration Plant	Lawrenceville VA	MSW		0.5 (1.9)
Fiberight Blairstown LLC	Blairstown IA	MSW		6.0 (22.7)
ICM Inc. Pilot Integrated Cellulosic Bio	St. Joseph MO	Corn fiber, switchgrass		0.3 (1.2)
Indian River Bioenergy Center	Vero Beach FL	Veg., Agric Waste, MSW		8.0 (30.3)
Iogen Inc.	Ottawa ON	Straw		0.5 (2.0)
Mascoma Corporation	Rome NY	Woody Biomass		0.2 (0.8)
Western Biomass Energy, LLC	Upton WY	Cellulosic		0.5 (1.9)
ZeaChem Boardman Biorefinery LLC	Boardman OR	Poplar		0.3 (1.0)
			Total	19.8 (74.8)
Plants Under Construction				
Abengoa Bioenergy Biomass	Hugoton KS	Corn Stover, Switchgrass		25.0 (94.6)
American Process Inc. Demonstration Plant	Thomaston GA	Sugarcane bagasse, wood		0.3 (1.1)
Blue Fire Renewable LLC	Fulton MS	Wood Waste		19.0 (71.9)
DupontDanisco Cellulosic Ethanol LLC	Nevada IA	Corn Stover		30.0 (113.6)
Enerkem Alberta Biofuels LP	Edmonton AB	Sorted MSW		10.0 (37.9)
Freedom Pines Biorefinery	Soperton GA	Woody Biomass		2.0 (7.6)
Poet-DSM Advanced Biofuels LLC	Emmetsburg IA	Corn Stover		20.0 (75.7)
Quad County Cellulosic Ethanol Plant	Galva IA	Corn Fiber		2.0 (7.6)
Woodland Biofuels Inc	SarniaON Canada	Wood Waste		0.5 (2.0)
			Total	108.8 (411.9)
Proposed Plants				
Advanced Biofuels Corp	Moses Lake WA	Cellulose		6 (22.7)
Agresti LLC	Pikeville KY	MSW		20 (75.7)
Atlantic Ethanol Inc.	Providence RI	Wood Waste		10 (37.9)
Canergy LLC	Brawley CA	Energy cane		25 (94.6)
Chemtex International Inc., Project Alpha	Clinton NC	Energy Grasses		20 (75.7)
Enerkern Mississippi Biofuels LLC	Pontotoc MS	RDF, Wood residue		10 (37.9)
Enerkern Green Field	Varennes QC	RDF, C&D debris		10 (37.9)
Fulcrum BioEnergyInc.Sierra Biofuels	McCarran NV	RDF		10 (37.9)
Mascoma Corporation	Drayton Valley AB	Hardwood		20 (75.7)
Mascoma Corp/Frontier Renewable Res.	Kinross MI	Hardwood		20 (75.7)
Mendota Bioenergy LLC	Tranquility CA	Energy beets		1 (3.8)
Nipawin Biomass Ethanol Co-operative	Nipawin SK	Waste Wood, Straw		26 (98.4)
Sunset Ethanol Inc	Fernley NV	Switchgrass, Sorgum		5 (18.9)
The Green Fuel Association Bieber II	Bieber CA	Switchgrass		40 (151.4)
The Green Fuel Association Corning II	Corning CA	Switchgrass		40 (151.4)
The Green Fuel Association Dorris II	Dorris CA	Switchgrass		40 (151.4)
Woodland Biofuels Inc.	Newton Falls NY	Wood Waste		20 (75.7)
World Ethanol Institute, LLC	Lenox GA	Paulownia		20 (75.7)
ZeaChem Boardman Biorefinery LLC	Boardman OR	Poplar, Straw		25 (94.6)
			Total	368 (1393)

establishes itself, the RFS goal illustrated in Table 1, continues to climb with each year. The development of additional renewable fuel sources seems necessary to achieve the 2022 RFS goal of producing a total of 36 Bgal/yr (136 x10⁶ m³/yr) of renewable fuel.

3. Biofuel production pathways

The following discussion of biofuel pathways illustrated in Figure 1, compares the parallel nature of biofuel production. The extraction of oils and sugars from a large variety of feedstocks can be utilized for final product development. The production of biofuel can be accomplished using different organisms and processes while producing a range of end products. Out of the many variables, process scenarios were chosen to serve as examples of possible pathways for the production of biofuel (Fig. 1). The pathways chosen represent ideal, but realistic alternatives. The generation of ethanol was diagramed by autotrophic algae pathways, taking into consideration both direct generation and indirect generation through the conversion of carbohydrates (Andersen and Andersen, 2006; Shen et al., 2009).

The generation of biodiesel was diagramed by way of autotrophic and heterotrophic algae as well as agricultural grain oils and recycled vegetable oils (Chisti, 2007; Gouveia et al., 2009; Meng et al., 2009).

The generation of biofuels is often referred to as first or second (also known as advanced) generation. First generation biofuels are derived from the sugars or vegetable oils from grown crops, such as corn, sugar cane or rapeseed oil, palm oil. These feedstocks can easily be converted into ethanol or biodiesel. However, second generation biofuels are made from cellulosic feedstock or woody crops, agricultural or municipal waste which are comparatively more involved to convert into sugars or oils. Once extracted, these sugars and oils can also be converted into ethanol and biodiesel, as depicted in Figure 1.

Biofuels, such as biodiesel, ethanol, and various petroleum products, can be produced by a large variety of biologically-dependent processes (Menetrez, 2012). Both natural and genetically modified organisms (GMOs) (algae, bacteria, fungi, and yeast) have been used to generate biofuels directly or indirectly by producing biofuel intermediate products such as oil,
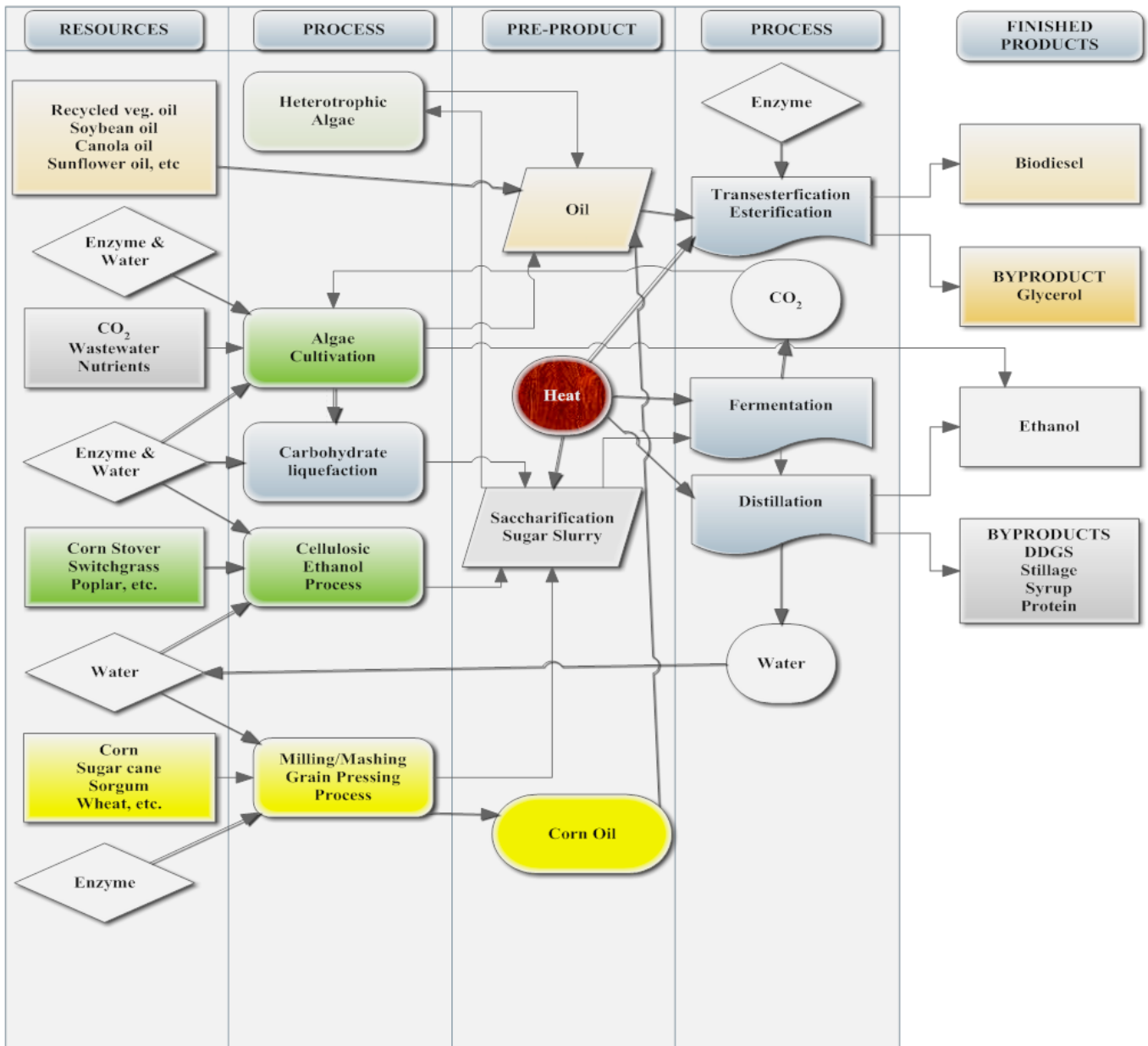
Fig.1. Biofuel Pathways (conventional bioethanol, cellulosic bioethanol, autotrophic and heterotrophic algae)

.

carbohydrates, and sugars or enzymes (Briggs, 2004; Hu et al., 2006; Becker, 2007; Li et al., 2007; Adams, et al., 2008; Evangelista et al., 2008; Graham et al., 2008; Hu et al., 2008; Lundquist et al., 2010; González- Fernández et al., 2011; Venkata Mohan et al., 2011; Menetrez, 2012).

The composition of many dry mass plant feedstocks (corn kernels, corn stover, switchgrass, poplar hybrid) commonly used for biofuel generation can vary (Table 3). Carbohydrates are either soluble sugars (made of glucose, sucrose, or fructose), or storage carbohydrates (composed of starch or fructans) (Sukenik et al., 1991; Burkholder, 1998; American Heritage, 2005; Dien et al., 2006; Dien and Bothast, 2009; Gao et al., 2010; U.S. Department of Agriculture, 2011; U.S. Department of Agriculture, 2012).

Table 3.
Constituents of plant feedstock biomass by % of dry matter (Dien et al., 2006; Dien and Bothast, 2009).

Constituent	Corn kernels	Corn stover	Switchgrass	Poplar hybrid
Either Ext. (nonpolar)	4.6	4.6	1.0	4.2
Protein	9.1	4.0	3.2	1.2
Starch	72.0	0.0	3.9	0.0
Cellulose	2.0	36.0	28.3	42.4
Hemicellulose	3.6	23.4	24.5	19.0
Klasonlignon	trace	18.6	15.4	25.7
Ash	1.5	12.5	5.4	1.8

4. Ethanol pathway

As apposes to ethanol derived from a petrochemical process, biologically derived ethanol sometimes referred to as bioethanol (the word ethanol is used in this paper), is a renewable fuel. Ethanol is an alcohol obtained from the fermentation of sugars and starches or by chemical synthesis. All four of the feedstocks shown as column headings in Table 4 can be processed to yield simple sugars and ultimately ethanol. Ethanol is used as a solvent, disinfectant, and as an additive to or replacement for petroleum-based fuels (American Heritage, 2005). Ethanol made from any sources (such as ethylene, algae, or cellulose) is identical to ethanol from other sources, such as corn starch, sugar, or sugarcane. An advantage that ethanol has over petroleum sources is that it can be produced from diverse renewable raw materials that are abundant. As mentioned above, U.S. ethanol production is currently at 15 Bgal/yr (56.8 x10^6 m^3/yr) from corn, and the U.S. Department of Energy (DOE) and the U.S. Department of Agriculture (USDA) are supporting the development of ethanol production from cellulosic feedstocks (plant materials, such as wood and switchgrass) (Demain et al., 2005; Perlack et al., 2005; U.S. Dept. Of Agric., 2000, 2005, 2006; U.S. Dept. Of Energy, 2004, 2007a, 2007b; Donner et al., 2008; Farrell and Morris, 2008; National Research Council, 2008; Tannura et al., 2008).

Ethanol production in the U.S. relies almost completely on sugar-platform feedstocks in the form of corn, and it consumes approximately one-third of the U.S. corn harvest, of which one-third is converted into distillers grains with solubles (DDGS) (Monceaux, 2011; Schroeder, 2003). The DDGS protein content (20 to 30%) has increased by a factor of three from the original corn grain and is a valuable byproduct ($208-327/ton [$0.29-0.36/kg] when dehydrated to 100% DW) used for animal feed (Schroeder, 2003; Monceaux, 2011; Broderick, 2013).

As of June 2012 the price of one bushel of corn (kernels) was $6.04/bu, ($171.40/m^3) and the price of ethanol was $2.14/gal ($0.57/L) (Parker, 2012a, b). At the yield of 2.32 gal/bu (17.4 L/m^3) (low) to 2.93 gal/bu (21.9 L/m^3) (high) the conversion of one bushel (0.035m^3) of corn will yield from $4.95 to $6.26 worth of ethanol at a cost per gallon (3.79 L) of $2.06 to $2.60 (Haefele and Ross, 2009; Parker, 2012b). The feedstock to product price straddles the break-even point, ranging from a loss of $0.08/gal ($0.02/L) to a profit of $0.46/gal ($0.12/L). The actual cost of production will vary for each plant, dependent on staff, operating and maintenance cost and financial commitments which are specific for the plant, but would be expected to raise cost and lower profit. Additionally, the renewable identification number (RIN) (see RIN section below) value to the manufacturer will vary and is also variable, but could add substantially to profits. Actual profitability would be affected by all factors which are beyond the capability of this paper. A number of examples of corn and ethanol prices are included in Table 4, to exemplify the variability and marginal profitability of the corn based ethanol industry. The trend of decreasing corn prices from Feb. to Aug. 2013 has created a favorable economic climate for ethanol producers despite the fluctuation in ethanol prices.

The sale of DDGS, and corn oil (used for biodiesel) can account for 25% additional revenues (3 to 4 cents/gal (0.8 to 1 cents/L) ethanol assuming a 0.3 lb/bu (4.8gm/m^3) corn press yield) (Emberland, 2013). Incentives such as tax subsidies, and the revenues from the RIN mechanism created by the 2007 Energy Independence and Security Act can also improve profitability (RIN values are discussed in more detail below, which as of May 2013 had a value of $0.79, approximately three to four times the profit margin) (Parker, 2013c, d). The profitability of producing corn-based ethanol is subject to the dictates of market volatility. The prices of corn, ethanol, and petroleum based products change daily, as do the opening and closing of ethanol manufacturing plants (Gaub, 2013; Parker 2013a). These factors create a difficult and unstable position for ethanol manufacturers that are saddled with enormous investments, long-term commitments, and ever increasing competition for feedstocks.

The trend from 2012 to 2014 as shown in Table 4, has seen corn prices decrease from $8.07 to $3.30 per bu ($0.23 to $0.09/L) and ethanol prices raise from $2.14 to $3.57 per gal ($0.57 to $0.94/L), and produced an increase in profits from near zero to between $2.45 to $2.15 per gallon ($0.65 to $0.57/L). During this same period, petroleum prices have decreased to approximately $91 per barrel and correspondingly, gasoline has decreased to less than the price of ethanol (Marketwatch, 2014). Over the two year period decreasing gasoline prices have absorbed what was initially the blending of ethanol from a less-expensive constituent to a cost addition. However temporary, this exemplifies the nature of fluctuating market pressures.

Climate change has brought about higher temperatures, droughts and floods, all of which contribute to lower yields of corn and higher prices. The marginal success of ethanol production from corn is likely to continue to vary with time, which emphasizes the need for using co-production techniques, such as using cellulosic corn stover and grass to produce ethanol. It also makes it clear why it is important to locate feed-lots that use DDGS near corn-based ethanol plants.

In addition to using corn to produce ethanol, a large variety of other types of biomass can be used, including cellulosic biomass, which has already been mentioned, and feedstocks that can be used to produce ethanol. Feedstocks that are high in carbohydrates (starches and sugars) are barley, cassava, sugar cane, sugar beet, sorghum, bagasse, grain, potatoes, sweet potatoes, sunflowers, fruit, molasses, and wheat. These feedstocks can be processed (using naturally occurring amylase enzymes to hydrolyze carbohydrates) to yield simple sugars (similar to other sugar platform processes). Subsequently, *Saccharomyces cerevisiae* (Brewer's yeast) can be used to convert these sugars to ethanol (Monceaux, 2009). In addition, many types of algae produce carbohydrates that could contribute significantly to the production of ethanol (Chisti, 2007; Mabee et al., 2011).

Carbohydrate containing feedstocks are milled and liquified into a mash or slurry, or they are pressed to yield a liquid (syrup) that has a high sugar content, and this syrup is cooked until it is gelatinous, after which it undergoes enzymatic hydrolyesis using the enzyme glucoamylase (Monceaux, 2009). The yeast *S. cerevisiae* is added to this sugar rich

Table 4.
Corn feedstock to ethanol profit/loss ($/bu or $/m^3) (Haefele and Ross, 2009).

Date	Corn Price $/bu ($/m^3)	Ethanol Price $/gal ($/L)	Feedstock to Ethanol High to Low Yield $/bu ($/m^3)	Range of Profit/Loss High to Low Yield $/bu ($/m^3)
6/2012	6.04 (171.40)	2.14 (0.57)	2.06 to 2.60 (58.46 to 73.78)	0.08 to -0.46 (2.27 to -13.05) [a]
8/2012	8.07 (229.00)	2.63 (0.70)	2.75 to 3.48 (78.04 to 98.75)	-0.12 to -0.85 (-3.41 to -24.12) [b]
1/2013	6.86 (194.67)	2.21 (0.58)	2.34 to 2.96 (66.40 to 84.00)	-0.13 to -0.75 (-3.69 to -21.28) [c]
2/2013	6.90 (195.80)	2.36 (0.62)	2.35 to 2.97 (66.69 to 84.28)	0.01 to -0.61 (0.28 to -17.31) [d]
4/2013	6.59 (187.00)	2.42 (0.64)	2.25 to 2.84 (63.85 to 80.59)	0.17 to -0.42 (4.82 to -11.92) [e]
5/2013	6.37 (180.76)	2.52 (0.67)	2.17 to 2.75 (61.58 to 78.04)	0.35 to -0.23 (9.93 to -34.90) [f]
7/2013	4.89 (138.76)	2.23 (0.60)	1.67 to 2.11 (47.39 to 59.88)	0.56 to 0.12 (15.89 to 3.41) [g]
8/2013	4.69 (133.09)	2.18 (0.58)	1.60 to 2.02 (45.40 to 57.32)	0.58 to 0.16 (16.46 to 4.54) [h]
9/2014	3.30 (93.64)	3.57 (0.94)	1.12 to 1.42 (31.78 to 40.30)	2.45 to 2.15 (69.52 to 61.01) [i]

[a] Parker,2012b   [b] Parker,2012c   [c] Parker, 2013a   [d] Parker, 2013b   [e] Parker, 2013c   [f] Parker, 2013d

[g] Parker, 2013e   [h] Parker, 2013f   [i] AAA Fuel Gauge Report, 2014

mixture, and the fermentation process produces ethanol and carbon dioxide (CO_2). The fermented mash contains 10 to 20 vol% ethanol and is heated to 82 to 84 ºC in a distillation process (Actual vaporization points of the constituents of the process are; methanol [wood alcohol], 64 ºC; ethanol 78 ºC; water 100 ºC) (Monceaux, 2009). Distillation of the mash and the following distillations strip the ethanol into a condensate that contains 95 vol % ethanol. Further heating of the remaining solids evaporates water from the mixture and produces either stillage, DDGS, or a syrup-like solution, depending on the original feedstock (Monceaux, 2009). A comparison of alternative feedstocks by unit mass (such as ton per unit ton) that is required to produce the same unit of mass of ethanol (i.e., a ton equivalent is approximately 304 gal [1,150 L] of ethanol) is provided in Table 5, along with the amounts of input water and output byproducts.

An ethanol production strategy should take into account feedstock availability, feedstock cost, processing cost, ethanol yield, process efficiency, and the sales prices of ethanol and byproducts of the process. Implicit to each stage is the cost of transportation, i.e., transporting corn to the plant and transporting ethanol and DDGS out of the plant. Transportation costs are significant, and they must be taken into account during the planning stage when making decisions concerning the location of the plant. One example of minimizing transportation costs is the location of animal feedlots near corn based ethanol plants to accommodate the large amounts of DDGS. The cost of producing feedstock and its availability are of great importance to the long-term sustainability of the plant.

Securing a stable, long-term supply of cellulosic feedstock or algae biomass feedstock is also of eminent importance and is uniquely difficult. The growth requirements for the production of feedstock can be effected by many factors, such as climate, seasonal variations and environmental limitations, which can limit or preclude growth in some areas or make it economically infeasible to transport the product. The importance of acquiring a dependable supply of feedstock is emphasized by the cost required to construct a plant, which was $250 million in the case of the Project Liberty plant (Shaffer, 2012).

5. Cellulose to ethanol pathway

Milling and pretreatment (milling, including steam explosion, pH adjustment and enzymatic hydrolysis saccharification) of cellulosic, hemicellulosic and lignocellulosic feedstocks, such as those in Table 4, can generate sugars. Feedstocks that have demonstrated the ability to produce various sugars from cellulose are corn stover, switchgrass, miscanthus, straw, hemp, cotton, and kena. In addition, ethanol can be produced from a variety of cellulosic waste materials, such as paper and cardboard municipal waste and agricultural and wood products (Dien and Bothast, 2011; Mabee et al., 2011; Monceaux, 2009). These feedstocks can be processed using naturally occurring enzymes derived either from biological interaction directly with the substrate feedstock or from a biological process that has harvested the enzyme and made it available for the cellulosic process. This processing yields simple sugars, i.e., the six carbon sugars D-glucose, fructose, and sucrose, and five carbon sugar D-xylose (Monceaux, 2009; Dien and Bothast, 2011). Then the slurry that results from the processing contains the sugars that are required to undergo fermentation with *S. cerevisiae* in a manner that is similar to that of other biologically induced processes (Olsson and Hahn-Hägerdal, 1996; Palmqvist and Hahn-Hägerdal, 2000; De Maagd et al., 2001; Kuiper, 2001; Letourneau, 2003; High, 2004; Iogen Technology, 2005; U.S. EPA, 2008).

Several processes commonly use both natural organisms and varieties of GMOs such as the fungus *Trichoderma reesei* for producing commercial cellulases (cellulose specific enzymes), which are used to convert cellulosic biomass to sugar (Palmqvist and Hahn-Hägerdal, 2000; U.S. EPA, 2008; Dien and Bothast, 2009). Other biologically dependent cellulosic processes use various types of fungi and bacteria (including GMO varieties) to produce cellulase, xylanase, and hemicellulase enzymes for the feedstocks listed in Table 4 to produce fermentable sugars, which may then be used to produce cellulosic ethanol (Palmqvist and Hahn-Hägerdal, 2000; U.S. Environmental Protection Agency, 2008; U.S. Department of Agriculture, 2013). After the process has yielded five and/or six carbon sugars, processing continues through yeast fermentation and distillation, similar to any other carbohydrate rich feedstock, to generate ethanol.

Generating ethanol from cellulosic feedstocks involves processes that are similar to, but uniquely different from generating ethanol from other feedstocks. For example, converting corn to sugar is inherently different from converting corn stover to sugar, however, the conversion of sugar to ethanol is similar to that in other processes. A comparison of feedstocks and their resource and footprint requirements for ethanol production are discussed below and listed in Tables 5 and 6.

Table 5.
Ethanol production process – mass of input feedstock required per unit mass of ethanol product produced and equivalent mass of byproduct output (i.e. ton of feedstock/ton of ethanol) (Monceaux, 2009).

Feedstock Alternatives	Input Materials Feedstock and H_2O	Input Water input/unit ethanol	Output DDGS DDGS and stillage	Output Stillage output/unit ethanol
Corn kernels (maize)	3.08	0.98	0.99	-
Suger cane juice	14.41	0.0	-	11.98 (biomass)
Wheat	3.34	1.81	1.32	-
Barley	4.03	2.89	2.0	-
Rye	3.72	2.57	1.7	-
Grain sorghum	3.05	1.64	1.04	-
Cassava chips	2.77	5.94	-	6.07 (land application)
Potatoes	10.80	8.71 (recycle)	1.25 (cake)	-
Sugar beet syrup	3.42	0.0	0.49 (syrup)	-

Cellulosic processes must deal with the mass and volume of the feedstock which are larger than most commonly used feedstocks such as corn. As listed in Table 6, one ton of ethanol (303.8 gal or 1,150 L) would require three tons of corn, or up to 10 tons (9,072 kg) of corn stover (U.S. Department of Energy, 2007b; Dien and Bothast, 2009; Monceaux, 2011). Process estimations predict that a 20 Mgal/yr (75,708 m^3) plant using energy grasses, such as switchgrass and miscanthus, will require approximately 300,000 to 600,000 tons per year (272.2 x10^6 to 544.3 x10^6 kg) of feedstock delivered by 100 trucks per day. The location of the plant will impact the mean truck-route for the incoming feedstock, but transportation costs remain largely unknown, and they include the cost of transporting the outgoing waste solids, which are likely to contain large quantities of lignin. Lignin is commonly used for heating, or converted to liquid fuel by thermochemical processing (Dien and Bothast, 2009; Pedroso et al., 2011).

Techniques for producing ethanol from cellulosic materials are currently being applied to industrial scale plants. The techniques that use corn stover, such as the Project Liberty plant, depend indirectly on the continued success of the corn ethanol industry. Although this industry is likely to continue, it should be recognized that this dependence brings with it inherent instabilities due to varying economic condition. There is little doubt that the success of the cellulosic ethanol industry is vital to the future of renewable energy (Menetrez, 2010). However, until industrial plants, such as the Project Liberty plant, have established a history of proven success, the viability of this industry will remain in question. Therefore, it also is necessary to explore the potential of other forms of ethanol production.

6. Algae ethanol pathway

Development of the algae biofuel industry has the potential to generate a variety of fuels. Commercial facilities that produce algae exist worldwide, and they are used predominantly for manufacturing food, cosmetics, and health-related products, not biofuel. Commercial enterprises have invested heavily and established industrial-scale, micro-algae farms that have the potential to bring algae to a stage similar to that of cellulosic ethanol (Menetrez, 2012; Milledge, 2001). These commercial processes use a variety of technologies, produce many different products, and usually are located in the lower latitudes of the U.S., where temperature, climate, and solar irradiance are favorable for the growth of algae.

Table 6.
Ethanol production – mass of feedstock input required per equal ton mass of ethanol produced (303.8 gal [1150 L] @ $2.63/gal [$0.69/L] = $800.00), and acres/ton ($m^2$/kg) C_2H_5OH.

Feedstock Alternatives and Ethanol Yield	Intake feedstocks (mass/mass)	Footprint required [acres/ton (m^2/kg)]
Corn kernels (154 bu/acre or 1.34 m^3/km^2 maize, 39.4 bu/ton or 1.5 L/kg)	3.08 [b]	0.75 (3.35) [g]
Wheat (70 bu/acre, or 609 L/km^2, 62 lb/bu or 0.8 kg/L)	3.34 [b]	1.80 (8.03) [h]
Barley (72 bu/acre, or 626 L/km^2, 48 lb/bu or 0.6 kg/L)	4.03 [b]	2.33 (10.39) [i]
Rye (12 bu/acre, or 104 L/km^2, 56 lb/bu or 0.7 kg/L)	3.72 [b]	4.74 (21.14) [j]
Potatoes (401 bu/acre, or 3,488L/km^2, 52 lb/bu or 0.7 kg/L)	10.80 [b]	1.04 (4.64) [j]
Sugar beet (SB) root (30 ton SB/acre, or 6.7 kg/m^2)	12.323 [c]	0.379 (1.69) [c]
Sugar cane (SC) stalk (36 ton/acre SC or 8.1kg/m^2)	13.54 [c]	0.372 (1.66) [c]
Grain sorghum (1.83 ton/acre or 0.4 kg/m^2)	3.05 [b]	1.67 (7.45) [j]
Sweet sorghum stalk (39 ton/acre SSS or 8.7 kg/m^2)*	21.30 [c]	0.548 (2.44) [c]
Switchgrass (4.6 ton/acre, or 1.0 kg/m^2 non-irrigated)	5-10 [d]	0.3-1 (1.34-4.46) [d,k,l]
Corn Stover (80 gal/ton, 0.16 L/kg byproduct)	5-10 [d]	0**
Microalgae (30% yield DW by PBR, 23.85 ton/acre, or 5.3 kg/m^2carbohydrate, byproduct) [a]	2.5 [e,f]	0***
Microalgae (7,000gal/acre or 6,546 L/km^2 by PBR)	*in situ*	0.044 (0.195) [m]

*Based on two crops per year   **Byproduct of corn kernels   ***Byproduct of microalgae oil production

[a] Chisti, 2007   [b] Monceaux, 2009   [c] Amorim, 2009   [d] Dien and Bothast, 2009   [e] Kim et al., 2011   [f] Lee et al., 2011

[g] Haefele and Ross, 2009   [h] Kansas Wheat Harvest Reports, 2012   [i] USDA, 2012   [j] USDA, 2011   [k] Pedroso et. al., 2011   [l] Mononoa et al., 2012   [m] Algenol, 2013

Algae can be used to produce ethanol and biodiesel in ponds that are open to the atmosphere [i.e. shallow ponds or tanks that can be circular or parallel raceway ponds (PRPs)] or in closed photobioreactors (PBRs). Most of the cultivation of algae is done in PRPs because of their low construction and operating costs (Briggs, 2004; Li et al., 2007; Venkata Mohan et al., 2007a,b Evangelista et al., 2008; Graham et al., 2008; González- Fernández et al., 2011). PBRs are contained, closely controlled systems in which an ideal environment is maintained to ensure high and stable production levels of algae (Chisti, 2007; Hu et al., 2006; 2008). Algae require water, efficient exposure to light, carbon dioxide, optimal temperature, culture density, appropriate pH levels, and a reasonable mixing regime. The most significant advantage that closed PBRs have over open PRPs is the ability to eliminate the introduction of unwanted microorganisms. Controlling contamination is necessary to achieve a stable, optimum culture and maximum yield. PBRs are often utilized for growing pure inoculant populations of microalgae in the early stages of the process for supplying large open, raceway, paddle wheel mixed pond PRPs where high growth rate cultivation can occur (Rosenberg et al., 2008; Lundquist, 2010).

Forms of cyanobacteria, commonly known as blue-green algae (including GMO varieties such as those harbouring genes from *Zymomonas mobilis*), can manufacture ethanol within the microalgae cell and (lacking true cell membranes) diffuse it into the culture media and headspace. Using a unique autotrophic PRP, CO_2 and inorganic nutrients such as those found in wastewater, ethanol can be synthesized, concentrated and recovered directly from the PRP (Badger, 2002; Demain et al., 2005; U.S. DOE, 2008; Algenol, 2011; Snow and Smith, 2012). Algenol Biofuels in cooperation with Dow Chemical Company, the Linde Group, the National Renewable Energy Laboratory (NREL), Georgia Institute of Technology, and Membrane Technology and Research, Inc., is in the process of developing an operational pilot-scale plant that uses this technology (Deng et al., 1999; Algenol, 2011).

Algae can produce substantial concentrations of lipids which can be used to produce biodiesel, jet-fuel and other petroleum products; as well as carbohydrates which can be processed into ethanol; and proteins (such as *Spirulina maxima* which is composed of 60-71% protein content) with high nutritional quality that commonly are used for human and animal consumption (Dien et al., 2006). Microalgal preparations are commonly marketed as health food, cosmetics, and animal feed (Dien et al., 2006; Becker, 2007; Adams et al., 2009; Dien et al., 2009). The potential value of algae's co-products is evident when comparing the protein content to that of other biofuel feedstocks such as corn kernels, which contains 9.1% protein (used as distillers grains with solubles or DDGS), corn stover which contains 4% protein, switchgrass which contains 3.2% protein, and a hybrid Poplar at 1.2% protein (Dien et al., 2006; Dien et al., 2009).

7. Algae biodiesel pathway

Biodiesel is a renewable fuel that is currently produced from various feedstocks, such as, soybean oil (90% of current production), recycled vegetable oil, sunflower oil, canola oil, cottonseed oil, animal fats, and lipids produced by algae (Chisti, 2007; Gouveia et al., 2009; Meng et al., 2009). Lipids are long carbon-chain molecules that serve as a structural component of the membrane of the algal cell. The lipid production of algal species varies from 20% to 80% DW. In addition, temperature, solar irradiance, the species-specific speed of growth and process time, and the manipulation of nutrients supplied per stage of growth can all affect lipid yield (Becker, 2007; Adams, et al., 2008; Gouveia, et al., 2009; Wageningen University, 2011). Becker (2007) listed the lipid, carbohydrate and protein constituents of 17 common algal species, and they are presented in Table 3. Algae that belong to several different families possess the ability to produce and accumulate a large fraction of their dry mass as lipids. As listed in Table 3, autotrophic lipid production from the algae *Botryococcus braunii* can produce 86% DW of lipid (oil) that can be separated and converted to biodiesel (Chisti, 2007; Gouveia et al., 2009; Meng et al., 2009; Chisti, 2013).

Biodiesel is defined as a mono-alkyl ester of a long-chain fatty acid that conforms to the American Society for Testing and Materials (ASTM) D6751 specifications for use in diesel engines (ASTM D6751 – 12, 2013). After extraction and separation, algae oil is processed into biodiesel by either of two process methods. In the classical method, the oil is mixed with an alcohol (usually methanol) and a basic catalyst (usually potassium hydroxide (KOH), or sodium ethanolate (CH_3COONa) and heated to approximately 70 °C (at 20 psi) for several hours in a process called transesterification. The products are biodiesel (67%) and glycerol (33%) (Chisti, 2007; Coppola et al., 2009; Gouveia et al., 2009; Meng et al., 2009).

In the second method, the oil and alcohol are mixed with the enzyme lipase, which can be produced from a number of organisms, such as the fungus *Metarhizium anisopliae, Aspergillus oryzae* (and *A. niger*) or a varieties of Gram-negative bacteria, i.e. *Chromobacterium viscosum,* causing transesterification at room temperature (Adachi et al, 2011; Fiametti et al., 2011; Foley et al., 2011; Talukder, 2011). Energy consumption for producing biodiesel is reduced in the enzyme method, and the process is made more energy efficient and less sensitive to process problems (Lam and Lee, 2011). In addition, this process has other advantages such as; creating a high-grade glycerin byproduct with improved value, reduced water consumption (less water washing), and reduced methanol consumption. There are no caustic catalysts used, no soap formed and no ion exchange, or adsorbents are used (Piedmont Biofuels, 2013). The enzyme cost of $0.15/gal ($0.04/L) of processed biodiesel is currently outweighed by the positive attributes of the process (Piedmont Biofuels, 2013; AAA Daily Fuel Gauge Report, 2014).

As of 2012, biodiesel manufacturers in North Carolina were paying the sources of yellow grease, such as restaurants, as much as $1.50/gal ($0.40/L) (U.S. Energy Information Administration, 2012a; Piedmont Biofuels, 2013). Yellow grease is the preferred, economical choice, although limited in quantity, and corn oil, soybean oil and canola oil are additional feedstocks being used (U.S. Energy Information Administration. 2012a,b,c; Piedmont Biofuels, 2013). If algae derived oil is to be economically successful, it must be produced and supplied at a price approaching $1.50/gal ($0.40/L). With the inclusion of transportation costs (which can vary greatly) and overall processing costs a gallon of B100 biodiesel can be produced for approximately $3.00 to $3.10 ($0.79 to $0.82/L) (Piedmont Biofuels, 2013). Comparing this price to petroleum-based diesel (of approximately $4.00/gal or $1.06/L) indicates an approximate profit of $1.00/gal ($0.26/L).

The profitable nature of using yellow grease for biodiesel has allowed small plants to survive, but their production of biodiesel is limited by the supply of recycled cooking oil. There also are major biodiesel plants in operation, such as the Louis Dreyfus Agricultural Industries plant in Claypool, IN, which produces 80 Mgal/yr (0.3×10^6 m^3) from soybean oil (Biodiesel Industry Directory, 2013). A biodiesel plant under construction by Archer Daniels Midland in Lloydminster, AB, Canada, will produce 265 Mgal/yr (1.0×10^6 m^3) from canola oil (Biodiesel Industry Directory, 2013). When the Lloydminster plant becomes operational, it will yield more than double the output of any other plant. This scale of operation will ensure that overall profitability is sustainable even though the profit per gallon will be less due to the high cost of the feedstock oil. It is not clear whether the plant will satisfy the process-dependent RFA requirement for advanced biofuel of 50% GHG emission reduction. The RFA qualification could affect RIN applicability and further marginalize profits.

(303.8 gal or 1.15×10^3 L) produced, also produced 0.96 tons (871 kg) of CO_2 as a byproduct of fermentation, almost a one to one ratio by weight (Monceaux, 2009). Presently in the U.S., the CO_2 produced from the annual production of 15 Bgal (50 Mton or 45.4×10^6 kg) of corn based ethanol is vented to the atmosphere or 48 Mton (43.5×10^9 kg) of CO_2, a known greenhouse gas (GHG). As depicted in Figure 1, CO_2 should be viewed as a resource, not a waste product. Producing biofuel from growing algae with the use of CO_2 is an example of a complete resource utilization, renewable fuel generation and GHG emission reduction. If algae cultivation is located to utilize industrial sources of CO_2, heat, and nutrients could improve growth and decrease costs. Locating algae cultivation sites near industrial sources of wastewater, CO_2, and waste heat could add value to the process itself. Algae oil may never be competitively priced at approximately $1.50/gal ($0.40/L) (Menetrez, 2012). However, if linked to a CO_2 emission mitigation price to offset the algae cultivation cost a multi-product mindset as depicted in Figure 1, can be achieved.

CO_2 injection from industrial sources such as coal-burning power plants has been demonstrated to increase the yield of algae while decreasing atmospheric emissions (Bhatnagar and Bhatnagar, 2001; Benemann et al., 2003; Chisti, 2006; Woertz et al., 2009; Bhatnagar et al., 2010; Weissman and Benemann, 2011).

Wastewater (agricultural or human), waste heat and CO_2 (from biological fermentation or power plant flue-gas) should be viewed as resources for growing algae (Venkata Mohan et al., 2007b; Venkata Mohan et al., 2008). Contributing to the capture and conversion of CO_2 into a marketable commodity may be the most important asset of algae cultivation, regardless of the economic value assigned that commodity. All products of lipids and carbohydrates for biofuels and protein for animal or human consumption

Table 7.
Biodiesel production – mass of feedstock input required per equal mass of biodiesel produced (assuming 33% loss of oil to glycerol, a density of 7.6 lb/gal [0.9 kg/L] oil, 350 gal [1,300 L] of oil equivalent) (Chisti, 2007; Jatropha for Biodiesel Figures, 2012; NC State University, 2012).

Feedstock Alternatives and Biodiesel Yield	Intake feedstocks (mass/mass)	Footprint required [acres/ton (m^2/kg)]
Corn kernels (maize, 3.8 ton/acre, or 0.85 kg/m^2, 18.4 gal/acre oil, or 17.2 L/km^2) [a]	72.3	19.00 (84.7)
Soybeans (1.32 ton/acre, or 0.30 kg/m^2, 66 gal/acre oil, or 61.72L/km^2) [c]	9.69	7.34 (32.7)
Canola (1.71 ton/acre, or 0.38kg/m^2, 150 gal/acre oil or 140 L/km^2) [c]	4.71	2.75 (12.3)
Jatropha (2.8 ton/acre, 0.63kg/m^2, 1.012 ton/acre oil, or 230 kg/m^2) [b]	4.9	1.73 (7.7)
Microalgae (70% yield DW by PBR, 14,636 gal/acre, or 13,588L/km^2) [a]	2.14	0.024 (0.1)
Microalgae (30% yield DW by PBR, 6,276 gal/acre, or 5,869 L/km^2) [a]	5.0	0.056 (0.3)
Microalgae (25% yield DW by PBR, 2,100 gal/acre or 1,964 L/km^2) [d]	14.9	0.166 (0.74)
Microalgae (45% yield DW by PRP, 12-15 ton/acre oil, 2,724-3,405 kg/m^2) [e]	10.5	0.1 (0.4)
Recycled vegetable.oil (recycled yellow grease)	1.5	0

[a] Chisti, 2007   [b] Jatropha For Biodiesel Figures, 2012   [c] NC State University, 2012   [d] Lundquist et al., 2010   [e] Chisti, 2013

In addition, similar to corn-based products, questions remain regarding the use of feedstocks that could be used for human consumption, such as soybeans and canola. The area of land required (or footprint) to generate a given quantity of biofuel feedstock is provided in Tables 6 and 7.

Questions remain regarding the balance of resource inputs and outputs which can only be answered after the commercial success of biofuel production facilities has been demonstrated. In order to achieve commercial success it will be necessary for algae farms to take advantage of every available resource to enhance growth potential, while simultaneously recycling the byproducts of the processes. This process interdependency is addressed separately for its symbiotic nature.

8. Industrial process symbiosis

Symbiotic processes currently can be found in the biofuel industry. The production of ethanol from corn also produces corn stover, which is being used to produce cellulosic ethanol. Ethanol manufacturers, such as Poet, also are generating corn oil and using the inedible fraction to produce biodiesel. In 2012, Poet extracted 250,000 tons of corn oil from 25 of its plants, which is equivalent to 68 Mgal (0.26×10^6 m^3) (Biodiesel Magazine, 2014). This example demonstrates how the harvest of corn can be used to generate bioethanol from carbohydrates (starches/sugars), cellulosic and bioethanol from corn stover, biodiesel from corn oil, as well as DDGS for animal feed.

An example of a potential benefit from the ethanol manufacturing process is the recycling of CO_2 from fermentation (see Figure 1). Every ton of ethanol

should be utilized.

9. Resource balance

Resources that are universally required to develop feedstocks into renewable fuels are nutrients, water, sunlight, land, mechanical processing (harvesting, washing, milling, etc.), heat (cooking, fermentation, distilation, etc.), and mechanical transport (feedstock to plant, process and product to market) energy. Tables 6 and 7 provide examples of sources of feedstocks that can be used to produce renewable fuel. It is however difficult to generalize resource inputs quantitatively due to process differences, recycle and byproduct utilization, such as corn stover for ethanol generation (which comes from the same harvest as grain-corn feedstock used to generate ethanol) or algae products/byproducts of oil for biodiesel and ethanol. The resources that were calculated are tons of feedstock and acres required per ton of ethanol (Table 6) or biodiesel (Table 7).

The land required to grow any feedstocks can be a significant resource investment. A comparison of the land area required for a feedstock is a useful indicator of the energy productivity of that pathway. For example, Table 6 lists the amount of land required to grow enough sugar cane (0.372 acres or 1.5 km^2) to produce a ton of ethanol (303.8 gal or 1.15×10^3 L), which is half of that required for corn (0.75 acres or 3.0 km^2) (Monceaux, 2009). However, the growth requirements and land attributes differ (such as land value and arability), as does the availability of feedstock. Irrespective of the inherent differences, land area, as a resource input, provides a valuable comparison of

renewable fuels. Tables 6 and 7 were developed to approximate the land resource requirement that was sufficient to produce enough feedstock to produce one ton of fuel (ethanol in Table 6: biodiesel in Table 7). Each intake of feedstock material or area of land listed in Tables 6 and 7 can produce a ton of ethanol or biodiesel respectively (Kansas Wheat Report 2012; USA Biodiesel Prices.com, 2012; U.S. Energy Information Administration, 2012b). Commodity prices of corn, ethanol, biodiesel, petroleum, and petroleum-based diesel are constantly changing based on global economic conditions and forces.

As mentioned above, every ton of corn based ethanol (303.8 gal or 1.15 $\times 10^3$ L) produced, also produced 0.96 tons (871 kg) of CO_2 as a byproduct of fermentation (Monceaux, 2009). The U.S. production of 15 Bgal (50 Mton or 45.4 $\times 10^6$ kg) of corn based ethanol produced an average 48 Mton (43.5 $\times 10^6$ kg) of CO_2. The biomass of microalgae contains approximately 50% carbon, which is obtained from the atmosphere or other sources of CO_2. The injection of the additional carbon in the form of CO_2 has demonstrated an increase in algae yield while sequestering the carbon (Benemann, 2003; Weissman and Benemann, 2003; Chisti, 2006; Menetrez, 2012). Theoretically, applying the yearly rate of CO_2 production to growing algae could produce 26 Mtons (23.6 $\times 10^6$ kg) of algae biomass (assuming 1.83 tons CO_2 per ton algae biomass), which could be converted into biofuel, such as; 10 Mtons (9.1 x 10^6 kg) of algae crude oil, or 6.5 Mgal (24,603 m^3) of biodiesel (assuming a 40% DW yield, and 95% separation recovery) (Christi, 2007). A substantial goal of sustainable biofuel generation (considering the present biodiesel production level is 1Bgal/yr or 3.8 $\times 10^6$ m^3), and a substantial displacement of a GHG which is currently being vented to the atmosphere. There are multiple advantages to both the CO_2 supplier (for resource recycling) and the algae/biofuel industry (for growth enhancement by CO_2 sequestration). This algae pathway could be promoted through RIN additions for both sides to offset additional costs.

In order to achieve a cost-efficient biofuel process, using waste products, such as wastewater or CO_2 emissions, and using land that has marginal agricultural potential should be priorities. It is understood that the availability of land, CO_2 and nutrient rich wastewater can be a difficult set of assets to find. However, it is important that every attempt at economic advantages be made, especially in the initial phase of industrial start-up. In addition, the use of byproducts and recycled materials should be an immediate or planned priority. The footprint of byproducts such as corn stover or algae carbohydrate (after lipid extraction) was listed as zero due to it not being a primary or virgin product. In each of these examples the footprint was listed for the primary product corn kernels and algae oil for biodiesel production (see Table 7). Eventually, the economic feasibility will be decided by the site plan (combining wastewater, CO_2, and land resources) the process plan, and operational success (including transporting resource inputs and outputs, and containing potential contaminants).

Table 6 lists two sources of ethanol produced from microalgae. The first example utilizes PBR technology as a byproduct after the lipid fraction has been removed, while the second uses a PRP system. Although PRP algae cultivation is the economical choice for biodiesel production purposes, PBR cultivation is required for the direct production of algae to ethanol as currently employed by Algenol. PBR technology is often used in hybrid PBR/PRP systems for biodiesel production. PBR systems are also used in pilot scale CO_2 capture systems. Often considered to be "niche systems" the importance of carbon capture could increase its future importance beyond biofuel production.

A form of marine algae (*Laminaria japonica*) contains hydrolysate solids comprised of up to 31.0% mannitol and 7.0% glucose. GMO *Escherichia coli* (KO11) can be used to convert the mannitol and glucose to ethanol, producing about 0.4 g of ethanol per gram of carbohydrate (Kim et al., 2011). Another form of marine algae, *Chlorella vulgaris* was used by Lee et al. (2011) to produce ethanol with a pretreatment of GMO *E. coli* (strains W3110, and SJL2526), achieving 0.4 g ethanol/g biomass (Lee et al., 2011). These are two examples of unique processes that utilize marine algae with high sugar contents in combination with GMO strains of *E. coli* that are able to produce ethanol at a high efficiency. Although feedstock availability may be a limiting factor, these processes illustrate an innovative approach to using many forms of algae.

Numerous species of microalgae can be used to produce lipids, proteins and carbohydrates. One such algae, *Prymnesium parvum*, produces approximately equal portions of lipids, proteins and carbohydrates, i.e., about

33% of each. For the purpose of generating the numbers listed in Tables 6 and 7, the yield from algae cultivation would agree with that measured from the growth of *P. parvum*. In this example, the primary product was lipids, while carbohydrates were listed as a secondary byproduct. Actual yields will depend on the specific microalgae and the conditions of the process. Recycled vegetable oil is listed as having a footprint of zero (similar to the byproducts listed in Table 6) due to it not being a primary and virgin product. The production of one ton of biodiesel (263.0 gal [995 L] at a density of 7.6 lb/gal [0.9 kg/L]) would require 2,666 lb (1,209 kg) of oil (350 gal [1,300 L] of oil, assuming a loss of approximately 33% to glycerol).

The versatility of algae for producing lipids, carbohydrates, and proteins will be needed to create multiple products in multiple markets to satisfy economic considerations successfully. Currently, biotechnology firms and the algae industry are focused on producing relatively low volumes of high-value products, such as pharmaceuticals or nutritional supplements. These same industries must refocus on high volumes of biofuel production at low competitive prices, as well as utilizing byproducts, such as protein for DDGS and carbohydrates for ethanol (Gladue and Maxey, 1994; Unkefer et al., 2004; Foley et al., 2011; Mononoa et al., 2012).

The cultivation of algae requires nutrients that can be accessed by using various wastewater streams, including water from the same recycled streams. It also requires heat for temperature control, which can be obtained from sunlight (when clouds are absent, and sunlight is abundant), industrial waste heat, or geothermal sources. In addition, algae can be utilized simultaneously for both the lipids (to produce biodiesel) and carbohydrates (to produce ethanol).

10. Renewable identification number (RIN)

The renewable identification number (RIN) mechanism created by the 2007 Energy Independence and Security Act documents the production of biofuel at refineries by registering every gallon (3.79 L) of biofuel that is produced. The assigned RIN value also reflects the energy content of the biofuel (Table 8). Corn based ethanol is issued a RIN of 1 which as of May 2013 had a value of $0.79, and by July 17 2017 had risen to $1.43, much greater than the profit margin (Haefele et al., 2009; Parker, 2012a,b,c; Emberland, 2013; Parker, 2013a,b,c,d,e).

One gallon of biodiesel derived directly from oil from grown and pressed feedstock (such as soybean-based biodiesel) or recycled cooking oil (yellow grease) is issued a RIN of 1.5, and as of July 17 2013 had a RIN value of $1.30 (Parker, 2013b). Advanced biofuels, such as cellulosic ethanol (produced from biomass such as corn stover or switchgrass), are issued a RIN of 2.5 (Weihrauch, 2007; Weisner, 2009). The EPA sets a yearly quota which dictates the amount of biofuel blended into petroleum based transportation fuel. Refiners, importers and blenders must comply to their EPA assigned RFS quota. The biofuel manufacturer registers every gallon produced with the EPA at the time of sale, transfer, or export/import for the purpose of ensuring that the biofuel is actually blended into motor fuel. The ethanol or biodiesel company issues the RIN for a month's production of biofuel, and this RIN is reported to EPA using a unique, 38 character number (Weihrauch, 2007). The RIN values are transferred with the biofuel to the purchaser (refiners, exporter, importers, and blenders of the fuel). Eventually, the bioethanol is blended into gasoline (E10, E15, E25 E85, E100), and the biodiesel is blended into petrodiesel (B2, B5, B10, B20, B100) (Weihrauch, 2007; Weisner, 2009).

Table 8.
Energy content of transportation fuels (Weihrauch, 2007; Consumer Energy Report, 2013; Zfacts, 2013).

Fuel	Btu/gal (Btu/L)	Etoh /Fuel Ratio	RIN
Ethanol (E100)	76,000 (2,008)	1	1
Biodiesel (B100)	118,296 (3,125)	0.655 Etoh/Gasoline	1.5
Gasoline	116,090 (3,067)	0.642 Etoh/Biodiesel	0.0
Cellulosic Ethanol	76,000 (2,008)	1	2.5

During economic periods in which blending biofuel is unprofitable, the RINs can be separated from the fuel and used as a commodity on the open market (Weihrauch, 2007; Weisner, 2009). During those years in which

more ethanol is produced than is set by the RFS, RINs can accumulate for up to one year, as noted by the expiration dates. Blenders with excess RINs can sell them to other blenders to be used as the market dictates (Weihrauch, 2007; Weisner, 2009; Wall Street Journal, 2013).

From manufacturing to blending and consumption, a RIN documents the compliance of the biofuel blenders with the RFS mandates of 2005 (Thompson et al., 2010). The buying, selling, and trading (swapping) of RINs in the marketplace is a complex undertaking, because it is influenced by market trends, agricultural fluctuations and regional needs (Weihrauch, 2007; Weisner, 2009). An example of the volatility of this system is seen in the three month variation in the corn grain RIN (D6), changing from $1.44 on July 16, 2013 to $0.675 on August 9, 2013, a 76.5 cent decrease (Platts McGraw Hill Financial, 2013). As described above, the economic position held by the biofuel industry is one of marginal profitability. The RIN system could lend a degree of flexibility to survive unfavorable economic periods, but market influences have demonstrated the vulnerability of the present system (Emery, 2012). A more complete discussion of the RIN system is beyond the scope of this paper.

11. Land requirement

The efficiency of algae to produce ethanol and biodiesel listed in Tables 6 and 7 were used to calculate the area of land required per billion gallons of biofuel (see Table 9). Depending on the efficiency of the process and overall yield, each billion gallons of ethanol would require a commitment of 175 miles^2 (453 km^2) to produce. Similarly, each billion gallons of biodiesel would require from 107 to 741 miles^2 (277 to 1,919 km^2) to produce (a wide range of variation). This range represents the collective area which would be dedicated to algae cultivation for the primary purpose of biofuel production. Although the dedication of this much land area to algae cultivation is significant, it is less than any other feedstock by at least an order of magnitude.

cellulosic ethanol plants in operation have a combined capacity of 19.75 Mgal/yr (74.8 x10^3 m^3/yr) (listed in Table 2), more than triple the 2013 capacity of 6.25 Mgal/yr (23.7 x10^3 m^3/yr), but far below the 2014 RFS of 1.75 Bgal/yr (6.6 x10^6 m^3/yr). Additionally, if 2014 is as successful as anticipated, the Abengoa Bioenergy plant (25 Mgal/yr [94.6 x10^3 m^3]), Poet-DSM plant (20 Mgal/yr [75,708 m^3]) and Dupont Danesco plant (30 Mgal/yr [113.6 x10^3 m^3]) will start production. With the addition of these three cellulosic ethanol plants the total capacity will exceed 100 Mgal/yr (378.5 x10^3 m^3), approximately five times the 2014 capacity, another major increase which falls far short of the RFS.

Many efforts are being made in the U.S. to develop cellulosic ethanol as an integral contributor to biofuels. Although numerous cellulosic ethanol plants are anticipated to initiate operation, the actual gap between available commercial supply and the RFS demand is widening. The 2022 RFS of 36 Bgal (136 x10^6 m^3) of total biofuel is a difficult goal to achieve, specifically due to the timeline necessary for the development of the cellulosic ethanol industry. It is for this reason that additional advantages should be created which promote other sources of transportation fuels, among them, algae biofuel which are highly flexible in process requirements and versatile in product output. Examples of advantages to promote algae biofuel production are: (1) Incentives to carbon capture processes by algae cultivation through the use of increased RIN values; and (2) Similarly, increased RIN values to algae derived biofuels produced through wastewater reclamation.

Potential pathways for producing biofuels from algae were evaluated for their feedstock and footprint demands. The present focus of the existing algae industries is on producing low volumes of high-value products for pharmaceuticals or nutritional supplements. There also are many developmental efforts backed by significant financial resources that are focused on the production of large volumes of biofuel products. However, little information regarding the present status of these process operations has been made available.

Table 9.
Microalgae Ethanol/Biodiesel production footprint, acres/Bgal (km^2/m^3) from Tables 6 and 7.

	Acres/ton (m^2/kg)	Acres/Bgal (km^2/m^3)
	Ethanol	**Ethanol**
Microalgae (30% yield DW by PBR, byproduct) [a]	0.0*	0.0*
Microalgae (7,000gal/acre, or 6,547 L/km^2 by PBR) [b]	0.044(48.4)	142,800 (152.8)
	Biodiesel	**Biodiesel**
Microalgae (70% yield DW by PBR, 14,636 gal/acre, or 13,588 L/km^2) [a]	0.024 (26.4)	68,570 (73.4)
Microalgae (30% yield DW by PBR, 6,276 gal/acre, or 5,869 L/km^2) [a]	0.056 (61.6)	160,000 (171.2)
Microalgae (25% yield DW by PBR, 2,100 gal/acre, or 1,964 L/km^2) [c]	0.166(182.6)	474,286 (507.5)
Microalgae (45% yield DW by PRP, 12-15 ton/acre oil, or 2,724-3,405 km/m^2) [c]	0.1 (11.0)	285,714 (305.7)

* Recycled Byproduct   [a] Chisti, 2007   [b] Algenol Biofuel, 2011   [c] Chisti, 2013

Actual sites may vary in size, efficiency, capacity and purpose. An example of a dual purpose algae production facility might have a primary purpose of using algae to reduce CO$_2$ emissions produced by coal-fired power plants. In addition to emissions from the combustion of coal, natural gas, and petroleum products, carbon capture to removal CO$_2$ with algae cultivation can be applied to many additional commercial sources such cement manufacturing and biological fermentation. In these examples reducing CO$_2$ emissions is the primary purpose, while the production of biofuels is integral but secondary.

12. Conclusion

Currently, biofuel production in the U.S. is limited to approximately 15 Bgal/yr (56.8 x10^6 m^3/yr) of corn, based ethanol and 1 Bgal/yr (3.8 x10^6 m^3/yr) of biodiesel (derived mainly from recycled vegetable oil). Feedstock limitations, i.e., sources of oil, are expected to limit the expansion of biodiesel production. Efforts are being made to quickly develop a cellulosic ethanol industry in the U.S. However, the development of this new (and largely unproven) industry with high investment requirements ($200 to $250 million for a plant that produces 20-25 Mgal/yr [75.7 x10^3 -94.6 x10^3 m^3]) and large resource needs is not likely to occur rapidly. As of early 2014, the industrial

The RFS is a guidance tool for the commercial production of a new generation of renewable biofuels (biodiesel, bioethanol, and petroleum). These sustainable energy sources contribute to our energy needs much like other established technologies such as solar, wind, and geothermal sources. Currently, the development of cellulosic and algae feedstocks for biofuel production has great potential to help diminish our dependence on petroleum-based fuels. The versatility of algae to produce multiple products in multiple markets is unique. The established focus of the algae industry to produce low volumes of high-value products (for pharmaceuticals or nutritional supplements) is evolving. There are many efforts backed by significant financial resources that are focused on the production of large volumes of biofuel products. The biofuel industry is young and growing quickly, it is hoped that this paper can provide a basis for the development of guidance that will assist this industry in growing in an environmentally friendly manner.

References

AAA Daily Fuel Gauge Report, 2014. Available at http://fuelgaugereport.aaa.com. (accessed on 28 November 2014).

Adachi, D., Hama, S., Numata, T., Nakashima, K., Ogino, C., Fukuda, H., Kondo, A., 2011. Development of an *Aspergillus oryzae* whole-cell

biocatalyst coexpressing triglyceride and partial glyceride lipases for biodiesel production. Bioresour. Technol. 102, 6723-6729.

Adams, J.M., Gallagher, J.A., Donnison, I.S., 2009. Fermentation study on Saccharina latissima for bioethanol production considering variable pre-treatments. J. Appl. Phycol. 21, 569-574.

Algenol Biofuels, 2011. Available at http://www.algenolbiofuels.com. (accessed on 28 November 2014).

Amorim, H.V., Basso, L.C., and Lopes, M.L. 2009. Sugar cane juice and molasses, beet molasses and sweet sorghum: Composition and usage, in: Ingledew, W.M., The Alcohol Textbook: A reference for the beverage, fuel, and industrial alcohol industries. Lallemand Ethanol Technology. 1, 39-46.

American Heritage, 2005. Science Dictionary, copyright by Houghton Mifflin Company, Published by Houghton Mifflin Company. pp. 217.

Andersen, T., Andersen, F.O., 2006. Effects of CO_2 concentration on growth of filamentous algae and Littorella uniflora in a Danish softwater lake. Aquat. Bot. 84, 267-271.

ASTM D6751 – 12, 2013. Standard Specification for Biodiesel Fuel Blend Stock (B100) for Middle Distillate Fuels. Available at http://www.astm.org/Standards/D6751.htm. (accessed on 28 November 2014).

Badger, P.C., 2002. Ethanol from cellulose: A general review, in: Trends in New Crops and New Uses, ed. J. Janick and A. Whipkey, ASHS Press, Alexandria, VA, 17–21.

Badger, P.C., 2002. Ethanol from cellulose: A general review. Trends in new crops and new uses. 17-21.

Becker, E.W., 2007. Micro-algae as a source of protein. Biotechnol. Adv. 25, 207–210.

Benemann, J., Pedroni, P.M., Davison, J., Beckert, H., Bergman, P., 2003. Technology Roadmap for Biofixation of CO_2 and Greenhouse Gas Abatement with Microalgae, Second National Conference on Carbon Sequestration. Available at http://www.netl.doe.gov/publications/proceedings/03/carbon-seq/PDFs/017.pdf. (accessed on 28 November 2014).

Bhatnagar, A., Bhatnagar, M., 2001. Strategies to employ algae and cyanobacteria for wastewater remediation, in: Maheshwari, D.K., Dubey, R.C. (Eds.), Innovative Approaches in Microbiology. Bishen Singh Mahendra Pal Singh, Dehra Dun, India, 379-403.

Bhatnagar, A., Bhatnagar, M., Chinnasamy, S., Das, K.C., 2010. Chlorella minutissima - a promising fuel alga for cultivation in municipal wastewaters. Appl. Biochem. Biotechnol. 161, 523-536.

Biodiesel Industry Directory, 2013. Available at http://store.bbiinternational.com/2012-Biodiesel-Industry-Directory-P9.aspx. (accessed on 28 November 2014).

Biodiesel Magazine, 2013. Twenty-five poet plants extract enough corn oil for 68 Mgal/yr of biodiesel, Poet, Available at http://www.biodieselmagazine.com/articles/8912/25-poet-plants-extract-enough-corn-oil-for-68-mmgy-of-biodiesel. (accessed on 28 November 2014).

Biomass Magazine, 2014. The Year is Here, visited 2014. Available at http://www.biomassmagazine.com/articles/10086/the-year-is-here. (accessed on 28 November 2014).

Briggs, M., 2004. Widescale Biodiesel Production from Algae. University of New Hampshire.

Broderick, S., 2013. DDGS export demand driving prices for now. Ethanol Producers Magazine.

Burkholder, J.M., 1998. Implications of harmful microalgae and heterotrophic dinoflagellates in management of sustainable marine fisheries. Ecol. Appl. 8(sp1), S37-S62.

Chisti, Y., 2006. Microalgae as sustainable cell factories. Environ. Eng. Manage. J. 5, 261-274.

Chisti, Y., 2007. Biodiesel from microalgae. Biotechnol. Adv. 25, 294-306.

Chisti, Y., 2013. Raceways-based production of algal crude oil. Green. 3(3-4), 195-216.

Consumer Energy Report, 2013. Available at http://www.energytrendsinsider.com/2006/03/28/biodiesel-king-of-alternative-fuels. (accessed on 28 November 2014).

Coppola, F., Simoncini, E., Pulselli, R.M., Brebbia, C.A., Tiezzi, E., 2009. Bioethanol potentials from marine residual biomass: an Emergy evaluation. Ecosyst. Sustain. Dev. VII. 379-387.

Crickmore, N., 1998. Revision of the nomenclature for the Bacillus thuringiensis pesticidal crystal proteins. Microbiol. Mol. Biol. Rev. 62, 807-813.

De Maagd, R.A., Bravo, A. and Crickmore, N., 2001. How *Bacillus Thuringiensis* Has Evolved Specific Toxins to Colonize the Insect World. Trends Genet. 17, 193-199.

Demain, L.D., Newcomb, M., and Wu, J.H., 2005. Cellulase, Clostridia and Ethanol. Microbiol.Mol. Biol. Rev. 69(1), 124-154.

Deng, M.D., Coleman, J.R., 1999. Ethanol synthesis by genetic engineering in cyanobacteria, Appl. Environ. Microbiol., 65(2), 523-528.

Dien, B.S., Bothast, R.J., 2009. A primer for lignocellulose biochemical conversion to fuel ethanol, in: Ingledew, W.M. (Eds.), The Alcohol Textbook. Nottingham University Press, Nottingham, U.K. Available at http://www1.eere.energy.gov/biomass/feedstock_databases.html. (accessed on 28 November 2014).

Dien, B.S., Jungb, H-J.G., Vogelc, K.P., Caslerd, M.D., Lamb, J.F.S., Itena, L., Mitchellc, R.B., and Sarathc, G., 2006. Chemical composition and response to dilute-acid pretreatment and enzymatic saccharification of alfalfa, reed canarygrass, and switchgrass. Biomass Bioenergy. 30, 880-891.

Donner, S.D., and Kucharik, C.J., 2008. Corn-based ethanol production compromises goal of reducing nitrogen export by the Mississippi River. Proc. Natl. Acad. Sci. U.S.A. 105(11), 4513-4518.

Emberland, P., 2013. Corn oil adds significant profitability to ethanol. Ethanol Producers Magazine, Benchmarking Business Analyst, Christianson and Associates.

Emery, T., 2012. Fraud Case Shows Holes in Exchange of Fuel Credits. The New York Times. Available at http://www.nytimes.com/2012/07/05/us/biofuel-fraud-case-shows-weak-spots-in-energy-credit-program.html. (accessed on 28 November 2014).

Environmental News Service, 2011. Cellulosic Ethanol Production Far Behind Renewable Fuel Standard, Washington, DC, (ENS) - The United States is not likely to reach cellulosic ethanol production mandates. Available at www.ens-newswire.com/ens/oct2011/2011-10-11-093.html. (accessed on 28 November 2014).

Ethanol Producers Digest, 2013. Ethanol Producers Magazine. BBI International, Grand Forks, North Dakota. Available at www.ethanolroducer.com. (accessed on 28 November 2014).

Evangelista, V., Barsanti, L., Frassanito, A.M., Passarelli, V., Gualieri, P., 2008. Algal Toxins: Nature, Occurrence, Effect and Detection Proceedings of the NATO Advanced Study Institute on Sensor Systems for Biological Threats: The Algal Toxins Case. Pisa, Italy, ISBN: 978-1-4020-8479-9.

Farrell, J., and Morris, D., 2008. Rural Power, Community-Scaled Renewable Energy and Rural Economic Development, New Rules Project. Minneapolis, MN. Available at http://www.newrules.org/de/ruralpower.pdf. (accessed on 28 November 2014).

Fiametti, K.G., Sychoski, M.M., De Cesaro, A., Furigo, A., Bretanha, L.C., Pereira, C.M.P., Treichel, H., de Oliveira, D., Oliveira, J.V., 2011. Ultrasound irradiation promoted efficient solvent-free lipase-catalyzed production of mono- and diacylglycerols from olive oil. Ultrason. Sonochem. 18(5), 981-987.

Foley, P.M., Beach, E.S., Zimmerman, J.B., 2011. Algae as a source of renewable chemicals: opportunities and challenges. Green Chem. 13(6), 1399-1405.

Gao, C., Zhai, Y., Ding, Y., Wu, Q., 2010. Application of sweet sorghum for biodiesel production by heterotrophic microalga *Chlorella protothecoides*. *Appl. Energy*. 87, 756-761.

Gaub, A., 2013. Ethanol company slashes staff, production comes to halt, Casa Grande Dispatch. Available at http://www.trivalleycentral.com/news_premium/ethanol-co-slashes-staff-production-comes-to-halt/article_ee5f041e-6edd-11e2-b0ed-001a4bcf887a.html. (accessed on 28 November 2014).

Gladue, R.M., Maxey, J.E., 1994. Microalgal feeds for aquaculture. J. Appl. Phycol. 6(2), 131-141.

González-Fernández, C., Molinuevo-Salces, B., García-González, M.C., 2011. Nitrogen transformations under different conditions in open ponds by means of microalgae–bacteria consortium treating pig slurry. Bioresour. Technol. 102(2), 960-966.

Gouveia, L., Marques, A.E., da Silva, T.L., Reis, A., 2009. Neochloris oleabundans UTEX #1185: a suitable renewable lipid source for biofuel production. J. Ind. Microbiol. Biotechnol. 36(6), 821-826.

Graham, L.E., Graham, J.E., Wilcox, L.W., 2008. Algae, Benjamin-Cummings Publishing, Menlo Park, CA, 2nd ed., ISBN: 0321559657.

Haefele, D.M., and Ross, A.J., 2009. Corn: Genetics, composition and quality, in: Ingledew W.M., The Alcohol Textbook. Lallemand Ethanol Technology.

Herndon, A., 2012. Cellulosic Biofuel to Surge in 2013 as First Plants Open, Bloomberg. Available at http://www.bloomberg.com/news/print/2012-12-11/cellulosic-biofuel-to-surge-in-2013-as-first-plants-open.html. (accessed on 28 November 2014).

High, S.M., Cohen, M.B., Shu, Q.Y. and Altosaar, I., 2004. Achieving successful deployment of Bt rice. Trends Plant Sci. 9(6), 286-292.

Hu, Q., Sommerfeld, M., Jarvis, E., Ghirardi, M.. Posewitz, M., Seibert, M., Darzins, A., 2008. Microalgal triacylglycerols as feedstocks for biofuel production: perspectives and advances. Plant J. 54(4), 621-639.

Hu, Q., Zhang, C., Sommerfeld, M., 2006. Biodiesel from algae: lessons learned over the past 60 years and future perspectives. Annual Meeting of the Phycological Society of America, Juneau, AK, July 7-12, pp. 40-41.

Iogen Technology, 2005. Makes it Possible (Process Overview), Iogen. Available at http://www.iogen.ca/cellulose_ethanol/what_is_ethanol/process.htm. (accessed on 28 November 2014).

Jatrophia for Biofuel, 2012. Figures, Look at the financial costs of commercial Jatrophia growing for Biodiesel. Available on http://www.reuk.co.uk/Jatropha-for-Biodiesel-Figures.htm. (accessed on 28 November 2014).

Kansas Wheat Harvest Reports, 2012. Kansas Wheat.

Kim, N.J., Li, H., Jung, K., Chang, H.N., Lee, P.C., 2011. Ethanol production from marine algal hydrolysates using Escherichia coli KO11. Bioresour. Technol. 102(16), 7466-7469.

Kuiper, H.A., Kleter, G.A., Noteborn, H.P., Kok, E.J., 2001. Assessment of the food safety issues related to genetically modified foods. Plant J. 27(6), 503-528.

Lam, M.K., Lee, K.T., 2011. Mixed methanol–ethanol technology to produce greener biodiesel from waste cooking oil: a breakthrough for $SO_4^{2-}/SnO_2–SiO_2$ catalyst. Fuel Process. Technol. 92(8). 1639-1645.

Lee, S., Oh, Y., Kim, D., Kwon, D., Lee, C., Lee, J., 2011. Converting Carbohydrates Extracted from Marine Algae into Ethanol Using Various Ethanolic Escherichia coli Strains. Appl. Biochem. Biotechnol. 164(6), 878-888.

Letourneau, D.K., Robinson, G.S. and Hagen, J.A., 2003. *Bt* crops: predicting effects of escaped transgenes on the fitness of wild plants and their herbivores. Environ. Biosaf. Res. 2(04), 219-246.

Li, X, Xu, H., Wu, Q., 2007. Large-scale biodiesel production from microalga Chlorella prothothecoides through heterotrophic cultivation in bioreactors. Biotechnol. Bioeng. 98(4), 764-771.

Lundquist, T.J., Woertz, L.C., Quinn, N.W.T., and Benemann, J.R., 2010. A Realistic Technology and Engineering Assessment of Algae Biofuel Production. Energy Biosci. Inst. 1.

Mabee, W.E., McFarlane, P.N., and Saddler, J.N., 2011. Biomass availability for lignocellulosic ethanol production. Biomass Bioenergy. 35(11), 4519-4529.

Marketwatch, 2014. Available at http://www.marketwatch.com/story/oil-rebounds-on-positive-china-manufacturing-data-2014-09-23-1103542. (accessed on 28 November 2014).

McAloon, A., Taylor, F., Yee, W., Ibsen, K., Wooley, R., 2000. Determining the cost of producing ethanol from corn starch and lignocellulosic feedstocks. National Renew. Energy Lab. Rep.

Menetrez, M.Y., 2010. The Potential Environmental Impact of Waste from Ethanol Production. J. Air Waste Manage. Assoc. 60(2), 245-250.

Menetrez, M.Y., 2012. An Overview of Algae Biofuel Production and Potential Environmental Impact. Environ. Sci. Technol.46 (13), 7073-7085.

Meng, X., Yang, J., Xu, X., Zhang, L., Nie, Q., Xian, M., 2009. Biodiesel production from oleaginous microorganisms. Renewable Energy. 34(1), 1-5.

Milledge, J., 2011. Commercial application of microalgae other than as biofuels: a brief review. Rev. Environ. Sci. Biotechnol. 10(1), 31-41.

Monceaux, D.A., 2009. Alternative Feedstocks for fuel ethanol production, in: Ingledew W.M., The Alcohol Textbook. Lallemand Ethanol Technology.

Mononoa, E.M., Nyrenb, P.E., Berti, M.T., Pryora, S.W., 2012. Variability in biomass yield, chemical composition, and ethanol potential of individual and mixed herbaceous biomass species grown in North Dakota. Ind. Crops Prod. 41, 331-339.

National Research Council, 2008. Water Implications of Biofuels Production in the United States. National Academy Press, Washington, DC.

NC State University A&T State University, Cooperative Extension, 2012. Oilseed Crops for Biodiesel Production, Available at http://www.extension.org/pages/28006/oilseed-crops-for-biodiesel-production. (accessed on 28 November 2014).

Olsson, L., and Hahn-Hägerdal, B., 1996. Fermentation of lignocellulosic hydrolysates for ethanol fermentation. Enzyme Microb. Technol. 18(5), 312-331.

Palmqvist, E., Hahn-Hägerdal, B., 2000. Fermentation of lignocellulosic hydrolysates. I : Inhibition and detoxification. Bioresour. Technol. 74(1), 17-24.

Parker, M., 2012a. Ethanol Rebounds from One-Week Low as Heat Threatens Corn Crop. Bloomberg News. Available at http://www.businessweek.com/news/2012-05-23/ethanol-rebounds-from-one-week-low-as-heat-threatens-corn-crop(accessed on 28 November 2014).

Parker, M., 2012b. Ethanol Output in U.S. Rose to Three-Month High, Report Shows. Bloomberg News. Available at http://www.businessweek.com/news/2012-05-23/ethanol-output-in-u-dot-s-dot-rose-to-three-month-high-report-shows . (accessed on 28 November 2014).

Parker, M., 2012c. Ethanol Caps Biggest Monthly Gain Since 2008 on Higher Corn. Bloomberg News. Available at http://www.businessweek.com/news/2012-07-31/ethanol-caps-biggest-monthly-gain-since-2008-on-higher-corn. (accessed on 28 November 2014).

Parker, M., 2012d. Ethanol Falls Against Gasoline on Concern Margins to Spur Output. Bloomberg News. Available at http://www.businessweek.com/news/2013-05-14/ethanol-falls-against-gasoline-on-concern-margins-to-spur-output. (accessed on 28 November 2014).

Parker, M., 2013a. Ethanol Stronger Against Gasoline as Shutdowns Curtail. Bloomberg News. Available at http://www.businessweek.com/news/2013-02-04/ethanol-stronger-against-gasoline-as-shutdowns-curtail. (accessed on 28 November 2014).

Parker, M., 2013b. Cellulosic biofuel RINs generated in June. Bloomberg News. Available at http://www.ethanolproducer.com/articles/10095/cellulosic-biofuel-rins-generated-in-june. (accessed on 28 November 2014).

Parker, M., 2013c. Ethanol Weakens Against Gasoline on Higher Inventory and Imports. Bloomberg News. Available at http://www.businessweek.com/news/2013-01-22/ethanol-weakens-against-gasoline-on-higher-inventory-and-imports. (accessed on 28 November 2014).

Parker, M., 2013d. Ethanol Weakens as Dry Midwestern Weather Boosts Corn Planting. Bloomberg News. Available at http://www.bloomberg.com/news/2013-05-06/ethanol-weakens-as-dry-midwestern-weather-boosts-corn-planting.html. (accessed on 28 November 2014).

Parker, M., 2013e. Ethanol's Discount to Gasoline Tightens on Lower Stockpiles. Available at http://www.bloomberg.com/news/2013-08-05/ethanol-s-discount-to-gasoline-tightens-on-lower-stockpiles.html. (accessed on 28 November 2014).

Pedroso, G.M., De Ben, C., Hutmacher, R.B., Orloff, S., Putnam, D., Six, J., van Kessel, C., Wright, S.D., Linquist, B.A., 2011. Switchgrass is a promising, high-yielding crop for California biofuel. California Agric. 65(3):168-173.

Perlack, R.D., Erbach, D.C.,Graham, R.L., Stokes, B.J., Turhollow, A.F., Wright, L.L., 2005. Biomass as Feedstock for a Bioenergy and Bioproducts Industry: the Technical Feasibility of a Billion-Ton Annual Supply. Environmental Science Division, Oak Ridge National Laboratory.

Piedmont Biofuels, 2013. Enzymatic Biodiesel. Available at http://www.biofuels.coop/wp-content/uploads/2011/01/2012-FAeSTER-Letter1.pdf. (accessed on 28 November 2014).

Platts McGraw Hill Financial, 2013. US RINs prices fall amid selloff, ending six-session ascent. Available at http://www.platts.com/latest-news/agriculture/houston/us-rins-prices-fall-amid-selloff-ending-six-session-21447191. (accessed on 28 November 2014).

Public Law 110-140-Dec. 19, 2007, 121 STAT. 1492, Energy Independence and Security Act of 2007. Available at http://www.gpo.gov/fdsys/pkg/PLAW-110publ140/content-detail.html. (accessed on 28 November 2014).

RFS Renewable Fuels Association, 2012. Renewable Fuels Standard. Available at http://www.ethanolrfa.org/pages/renewable-fuels-standard (accessed on 28 November 2014)

Rosenberg J.N., Oyler GA, Wilkinson L., Betenbaugh M.J., 2008. A green light for engineered algae: Redirecting metabolism to fuel a biotechnology revolution. Curr. Opin. Biotechnol. 19(5), 430-436.

Schroeder, J.W., 2003. Distillers Grains as a Protein and Energy Supplement for Dairy Cattle. NDSU Extension Service, AS-1241, North Dakota State University, Fargo, North Dakota.

Shaffer, D., 2012. Poet breaks ground on cellulosic ethanol plant in Iowa, Star Tribune Updated. Available at http://www.startribune.com/business/142515155.html. (accessed on 28 November 2014).

Shen, Y., Pei, Z., Yuan, W., Mao, E., 2009. Effect of nitrogen and extraction method on algae lipid yield. Int. J. Agric. Biol. Eng. 2(1), 51-57.

Snow, A.A., Smith, V.S., 2012. Genetically Engineered Algae for Biofuels: A Key Role for Ecologists. BioScience. 62(8), 765-768.

Sukenik, A., Levy, R.S., Levy, Y., Falkowski, P.G., Dubinsky, Z., 1991. Optimizing algal biomass production in an outdoor pond: a simulation model. J. Appl. Phycol. 3(3), 191-201.

Talukder, M.R., Das, P., Shu Fang, T., Wu, J.C., 2011. Enhanced enzymatic transesterification of palm oil to biodiesel. Biochem. Eng. J. 55(2), 119-122.

Tannura, M., Irwin, S., and Good, D., 2008. Are corn trend yields increasing at a faster rate? Marketing and Outlook Briefs, Department of Agriculture and Consumer Economics, University of Illinois at Urbana-Champaign. MOBR 08-02.

The Wall Street Journal, 2013. January 28, 2013, 7:06 P.M. ET Zero Dark Ethanol, Available at http://online.wsj.com/article/SB100014241278873243292045782700439673301294.html?KEYWORDS=biofuel&mod=dist_smartbrief#printMode. (accessed on 28 November 2014).

Thompson, W., Meyer, S., Westhoff, P., 2010. The New Markets for Renewable Identification Numbers. Appl. Econ. Perspect. Policy. *32*(4), 588-603.

Unkefer P.J., Knight T.J., Martinez R.A., 2004. Use of prolines for improving growth and other properties of plants and algae. U.S. Patent No. 6,831,040.

USA Biodiesel Prices.com , 2012. Biodiesel Stations and Prices for the entire US. Available on http://www.usabiodieselprices.com/. (accessed on 28 November 2014).

U.S. Biodiesels Digest, 2011. US biodiesel production sets all-time record. Available at http://www.biofuelsdigest.com/bdigest/2011/11/29/2011-us-biodiesel-production-sets-all-time-record/. (accessed on 28 November 2014).

U.S. Department of Agriculture, 2011. Principal Crops Area Planted and Harvested – States and United States: 2009-2011. Available at http://usda01.library.cornell.edu/usda/current/CropProdSu/CropProdSu-01-12-2012.pdf. (accessed on 28 November 2014).

U.S. Department of Agriculture, 2012. Press Release, National Agricultural Statistics Service, United States Department of Agriculture. Washington, DC, Available at http://www.nass.usda.gov/Statistics_by_State/Washington/Publications/Current_News_Release/wwhtaug.pdf. (accessed on 28 November 2014).

U.S. Department of Agriculture, 2006. Rural Development. Section 9006, Renewable Energy Systems and Energy Efficiency Improvements Program. (Available at http://www.farmenergy.org/)

U.S. Department of Energy. Biomass Program Multi-Year Technical Plan. (Available at http://www1.eere.energy.gov/biomass/pdfs/mytp.pdf). (accessed on 28 November 2014).

U.S. Department of Agriculture, 2013. Statement on the Detection of Genetically Engineered Wheat in Oregon. Available at http://www.usda.gov/wps/portal/usda/usdahome?contentid=2013/06/0127.xml. (accessed on 28 November 2014).

U.S. Department of Energy, 2004. Assumptions for the Annual Energy Outlook, with Projections to 2025, Energy Information Administration, DOE/EIA-0554.

U.S. Department of Energy, 2007a. National Renewable Energy Laboratory (NREL), Research Advances Cellulosic Ethanol, NREL/BR-510-40742.

U.S. Department of Energy, 2007b. DOE Selects Six Cellulosic Ethanol Plants for Up to $385 Million in Federal Funding, DOE Office of Public Affairs, Washington, D.C.

U.S. Department of Energy (DOE), 2008. Office of Science, Genomics: GTL Systems Biology for Energy and Environment, Cellulosic Ethanol: Fuel Ethanol Production. Available at http://genomicsgtl.energy.gov. (accessed on 28 November 2014).

U.S. Energy Information Administration, 2012a. Biodiesel Performance, Costs, and Use. Available at http://www.eia.gov/oiaf/analysispaper/biodiesel/. (accessed on 28 November 2014).

U.S. Energy Information Administration, 2012b. Biodiesel demand estimates now provided in petroleum supply and demand balances. Available at http://www.eia.gov/biofuels/biodiesel/production/table1.pdf. (accessed on 28 November 2014).

U.S. Energy Information Administration, 2012c. Petroleum & Other Liquids, Gasoline and Diesel Fuel Update, Sept. 17, 2012. Available at http://www.eia.gov/petroleum/gasdiesel/. (accessed on 28 November 2014).

U.S. Environmental Protection Agency, 2008. Fact Sheet: Final Changes for Certain Ethanol Production Facilities Under Three Clean Air Act Permitting Programs, EPA-HQ-OAR-2006-0089. Available at http://www.epa.gov/nsr/fs20070412.html. (accessed on 28 November 2014).

Venkata Mohan, S., Bhaskar, Y.B., Krishna, T.M., Chandrasekhara Rao, N., Lalit Babu, V., Sarma, P.N., 2007a. Biohydrogen production from chemical wastewater as substrate by selectively enriched anaerobic mixed consortia: Influence of fermentation pH and substrate composition. Int. J. Hydrogen Energy. 32(13), 2286-2295.

Venkata Mohan, S., Mohanakrishna, G., Raghuvulu, S.V., Sarma, P.N., 2007b. Enhancing biohydrogen production from chemical wastewater treatment in anaerobic sequencing batch biofilm reactor (AnSBBR) by bioaugmenting with selectively enriched kanamycin resistant anaerobic mixed consortia. Int. J. Hydrogen Energy. 32(15), 3284-3292.

Wageningen University, 2011. Research on microalgae within Wageningen UR. Available at http://www.algae.wur.nl/UK/technologies/production/heterotrophic_organisms/. (accessed on 28 November 2014).

Wallace, R., Ibsen, K., McAloon, A., Yee, W., 2005. Feasibility Study for Co-Locating and Integrating Ethanol Production Plants from Corn Starch and Lignocellulosic Feedstocks (Revised) (No. NREL/TP-510-37092). National Renew. Energy Lab., Golden, CO (US).

Weihrauch, J., 2007. Registration, Recordkeeping, and Reporting Requirements [for RINs], U.S. EPA Office of Transportation and Air Quality. Presentation at a Renewable Fuels Standard Workshop, May 10, 2007.

Weissman, J.C., Benemann, J.R., 2003. Comparison of Marine Microalgae Culture Systems, Second National Conference on Carbon Sequestration. Available at http://www.netl.doe.gov/publications/proceedings/03/carbon-seq/PDFs/217.pdf. (accessed on 28 November 2014).

Wisner, R., 2009. Renewable Identification Numbers (RINs) and Government Biofuels Blending Mandates. AgMRC Renewable Energy Newsletter.

Woertz, I., Feffer, A., Lundquist, T., Nelson, Y., 2009. Algae grown on dairy and municipal wastewater for simultaneous nutrient removal and lipid production for biofuel feedstock. J. Environ. Eng. 135(11), 1115-1122.

Zfacts, 2013. Available at http://zfacts.com/p/436.html. (accessed on 28 November 2014).