

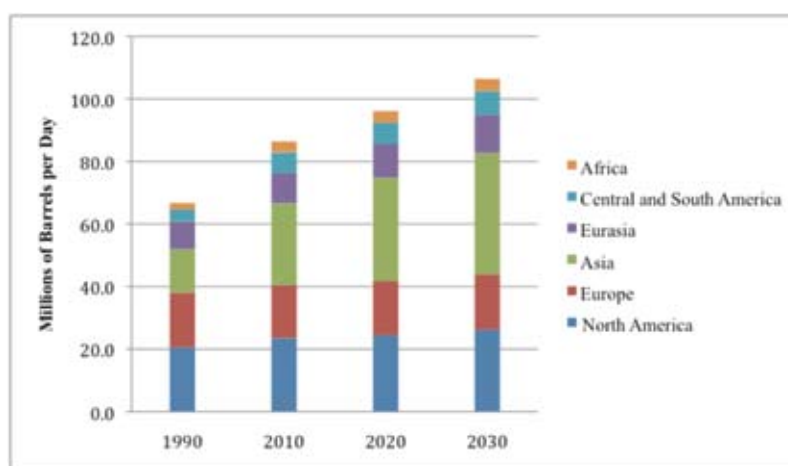
## Trends in Biofuel Industry

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Each day, the world consumes approximately 85 million barrels fossil based petroleum products<sup>1</sup>. United States leads that consumption by guzzling down nearly 20 million barrels per day. The world consumption is expected to reach 100 million barrels in 2030, driven primarily by the rapid economic growth in Asia and South America (Figure 1). Majority of the world petroleum consumption goes into the transportation fuel, which is responsible for about one-quarter and a rising share of energy-related greenhouse gas (GHG) emissions.



**Figure 1:** World petroleum consumption (source: EIA International Energy Outlook<sup>1</sup>)

As countries look for sustainable economic growth and energy security, while reducing greenhouse gas (GHG) emissions and dependence on fossil based petroleum products, alternative energy sources such as biofuels are gaining increasing attention. There has been a resurgence of research and development activities to develop next generation biofuels that use sustainable feedstock and provide GHG reduction. Escalating crude prices and favorable government policy also provide financial incentives, enticing companies in industries including petroleum, agribusiness, biotechnology, chemicals, engineering, and financial services, to join the effort.

Biofuels are fuels derived from biomass via chemical, biological, or thermochemical means. Currently, biofuels provide approximately 2% of the world's transportation fuel and is expected to reach 7% in 2020, according to the recent report published by EIA<sup>2</sup>. Compared to fossil based petroleum products, biofuels offer the following advantages.

- \* They are renewable, thus can be naturally replenished if managed properly
- \* They help to reduce GHG emissions because their feedstock (bio-

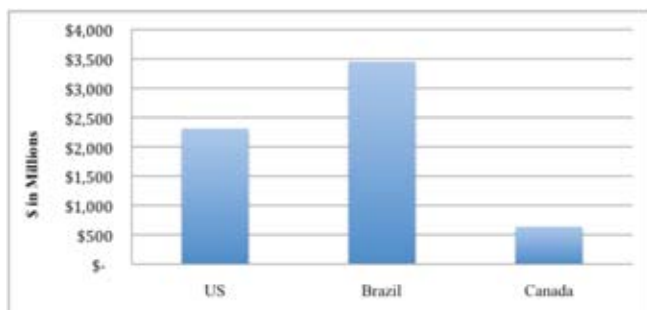
mass) can sequester carbon dioxide during growth. Estimated GHG reductions for various types of biofuel feedstock are listed in Table 1

- \* They can be globally and evenly distributed and can be exploited via environmentally friendly means. Thus, they provide an opportunity for regional energy sufficiency across the globe.
- \* They provide agriculture diversity and create new job opportunities.

Biofuel Feedstock	Relative GHG Reduction
Starch (corn, wheat)	15-40%
Vegetable Oil (soy bean, rapeseed)	45-75%
Sugar (sugar cane, sugar beet)	45-90%
Waste (agriculture residue, sewer)	65-100%
Lignocellulose (switch grass, poplar)	70-110%

**Table 1:** Green house gas reduction of various biofuel feedstocks as related to fossil derived petroleum<sup>3</sup>

The past few years witnessed tremendous growth in biofuels, with multi-billion dollar investment pouring into the industry. Despite the recent economic crisis, in 2009 alone, various institutions in the United States committed more than \$2 billion in the biofuel industry. This number is dwarfed by the nearly \$3.5 billion pump into biofuel industry in Brazil (Figure 2). Most of investments came from the private sector. But the United States government is one of the biggest investors. In September of 2009, The United States Department of Energy released \$564 million to support 19 integrated biorefinery projects at pilot, demonstration, and commercial scales.

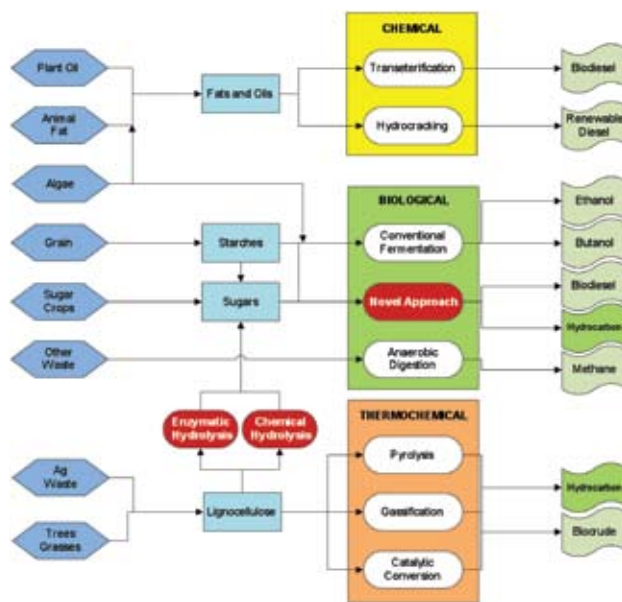


**Figure 2:** Global biofuel investment in 2009 (source: biofuelsdigest.com)

There are a number of approaches to make biofuels, as depicted in Figure 3, all of which begin with biologically

produced feedstock, such as crops, seeds, trees, fats, algae or waste derived from these materials. The feedstock harnesses the solar energy via direct or indirect photosynthesis, stored it as chemical energy in the bonds of the biological materials therein. Broadly speaking, there are three types of conversion processes: chemical, biological, or thermochemical. Using the chemical process, plant derived fats and oils undergo chemical reactions such as transesterification or hydrogenation. The resulting products can be used as biodiesel or renewable diesel. Using the thermochemical process, biomass is converted into bio oil or syngas under extremely high temperature and in the absence of air. The bio oil or syngas is then catalytically converted into variety of fuel products. Using the biological process, raw feedstock is broken down into fermentable sugars via chemical or enzymatic reactions. The sugars are then biologically converted to fuel products using different microorganisms. Examples of classic biological conversion methods include yeast fermentation and anaerobic digestion. During yeast fermentation, sugars are converted into ethanol. During anaerobic digestion, a variety of waste products can be broken down and converted into methane (biogas), with the help of a mixture of different microorganisms. The biogas can be used directly as a fuel or convert to other products.

The recent boom in the biofuels industry also saw a rally of research efforts in developing new technologies, including advanced synthetic biology, metabolic engineering, and bioprocess engineering. The focus has been turned towards two major conversion technologies: lignocelluloses to sugar,



**Figure 3:** Many routes to biofuels

and sugar to fuel. These two approaches are highlighted in red in Figure 3.

There are many routes to make biofuels, some of which are based on well established technologies while others are still in the early development stages. It is difficult to predict which process(es) will become the ultimate winner. However, the key criteria for success are clear, including cost, scale, sustainability, and compatibility.

### **Cost**

Even with its “green” and “renewable” appeal, biofuels will need to be cost competitive relative to fossil based petroleum fuels to be accepted by the general public and become commercially viable. Cost competitiveness depends on a few factors, including the cost of feedstock and conversion technology, and government policies. The cost of feedstock can vary significantly by region and fluctuate in the years ahead. For example, sugar price in the United States is currently two to three times higher than it is in Brazil. The cost of cultivating, harvesting and transporting feedstock must be well understood and driven down, and scaling up the technology to commercial scale must be economical. Depending on the future fuel price, only those low cost producers who possess most efficient conversion technologies will be the ultimate winners. Governmental policies, whether in the forms of mandates, credits, or subsidies, may also alter the industry’s ground rules to match changing priorities in climate change, energy security and economic development.

### **Scale**

The developing world’s appetite for energy is huge and will continue to grow. In order to make an impact, the biofuel technology must be readily scalable to meet the ever increasing energy demand. What impact the scalability the most are feedstock availability and conversion technology. Tens of millions of tons of biomass are required annually to produce enough fuels to make a difference, given the size of the global marketplace. The question then becomes: is there enough feedstock to meet the scale or demand; and can the conversion technology be scaled up without compromising the conversion efficiency or incurring too much capital expenditure?

### **Sustainability**

The types of feedstock for producing biofuels have been a point of contention for many years. First generation biofuels utilize crops such as corn and soybean, which are edible foods. Two years ago, the rising of corn and

soybean prices as a result of biofuel production led to a global debate on food vs. fuel, which triggered a great deal of attention on the dilemma and risk of diverting farmland or crops to biofuels production to the detriment of the food supply. While there are valid arguments on both sides, ultimately the feedstock must come from non-food sources, such as energy crops that can be grown on marginal land with little or no fertilizer usage, agriculture or forestry waste materials, and most recently algae. Much research and development effort is currently placed on the production of next generation biofuels from non-food crops, crop residues, or waste materials. Accordingly, environmental and socioeconomic issues of land, water, and biomass utilization must also be understood and considered, in the production and use of biofuels.

### **Compatibility**

Significant amount infrastructure already exists to transport, store and distribute the fuel to the end users. There are estimated over two million kilometers of fuel pipelines, hundreds of thousands of filling stations, and not to mention hundreds of millions of cars and other transportation vehicles that use conventional fossil fuels. To enable rapid market acceptance, it is important that biofuels be compatible with existing infrastructure and vehicles, and be a true drop-in replacement for conventional fuels. That is the challenge that the first generation biofuels have faced.

### **First Regeneration Biofuel**

Despite the many paths to produce biofuels that are known, currently there are only two types of biofuels that are commercially available. They are yeast fermentation derived ethanol and transesterification derived biodiesel, which are generally regarded as first generation biofuels.

Ethanol is the most widely used biofuel today. Global production of ethanol is estimated to be 23 billion gallons in 2010, led by Brazil, which is expected to contribute nearly 45% of the total ethanol production, followed by the United States<sup>4</sup>. Ethanol is a short chain alcohol and traditionally made through yeast fermentation that converts sugar into ethanol, which is then distilled into its final commercial form. This ethanol is then blended into gasoline for use in passenger vehicles. Gasoline with up to 10 percent ethanol (E10) can be used in most vehicles without further modification, while special flexible fuel vehicles can use a gasoline-ethanol blend that has up to 85 percent ethanol (E85). Sugar cane is the most important crop for producing ethanol biofuels today. Sugar cane derived ethanol accounts for more than 40 percent of all

fuel ethanol. Corn ranks a close second, serving as the primary source for ethanol biofuel production in the United States.

Biodiesel is a renewable diesel fuel substitute that is currently made by combining natural oils or fats with an alcohol (usually methanol) to form fatty acid methyl ester. It is currently the most widely used biofuel in Europe, which accounts for 80 percent of the biodiesel produced in the world. It is produced mainly from rapeseeds or sunflower seeds in Europe, and soy beans in the United States. Biodiesel can be blended with petroleum diesel in various proportions. The most common blend of biodiesel in the United States is 20 percent biodiesel (B20). In the 21st century, biodiesel has moved out of experimental and niche markets and into the mainstream. In 2001 the American Society for Testing and Materials (ASTM) created the new standard, #6751, for biodiesel, paving the way for car manufacturers to adopt biodiesels as fuels for their vehicles by providing a consistent set of performance parameters for biodiesels. The Environmental Protection Agency (EPA) has also contributed by providing various standards to evaluate biodiesels for on-road use. Tier I and Tier II tests established and promulgated by the EPA renders those in compliance clean alternatives to petroleum-based diesel fuels. Gradually, biodiesel fuels have become attractive to public transportation providers who are concerned about exposing adults and children to toxins and carcinogens that existed in petroleum based diesel products. Biodiesel is simple to use, biodegradable, nontoxic, and essentially free of sulfur and aromatics.

Most analyses continue to indicate a net benefit in terms of GHG emissions reduction and energy balance in the first generation biofuels. But these biofuels have several apparent drawbacks. Current concerns for many, but not all, of the first generation biofuels include:

- \* Ethanol and biodiesel having lower energy density (Table 2)
- \* Ethanol being incompatible with existing infrastructure
- \* Feedstock competing with food sources
- \* Lacking in cost competitiveness when taking account of total production costs excluding government grants and subsidies
- \* Providing only marginal GHG reduction benefits
- \* Competing for arable land usage and scarce water resources in some regions.

Type of Biofuel	Volumetric Energy (MJ/L)	Energy Density Relative to Petroleum Fuel
Ethanol	20	70%
Biodiesel	31-35	85%
Long Chain Hydrocarbon (Alkane)	~40	100%

**Table 2:** *Energy density of biofuels*

### Next Generation Biofuel

The goal of developing next generation biofuels is to extend the amount of biofuel that can be produced sustainably by utilizing non-food based feedstock, and to produce drop-in replacement of petroleum fuel, as well as addressing certain constraints of the first generation biofuel, such as the marginal GHG reduction and lack of infrastructure compatibility. Compared to first generation biofuel, next generation biofuels offer the following advantages.

- \* Utilization of a wider range of biomass feedstock, and avoidance of competition with food production.
- \* More favorable GHG reduction. Lignocellulosic biomass derived biofuel could provide about 90% greater CO<sub>2</sub> reduction than fossil fuels, whereas corn ethanol reduces CO<sub>2</sub> levels by 40%.
- \* Higher land yield and less water usage. With advancement in plant sciences, energy crops can easily triple the yield per hectare.
- \* Producing at cost competitive prices, using low-cost biomass feedstock.
- \* Producing fuels of higher quality than first generation biofuels, and enabling true drop-in replacement fuels.

Among the many recent developments in making next generation biofuels biologically, two types of conversion technologies that complement each other, lignocelluloses to sugar, and sugar to fuel, have been the main focus.

### Lignocullose to Sugar

Lignocullose is the most abundant biomass on Earth. It exists in various high energy crops, such as switch grass, poplar, and in agriculture residues or forest wastes, such as corn stovers, and wood chips. Technologies have been developed in the “lignoculloses to sugar” path is to break down the rigid lignocullosic material into cellulose, hemiculloses, and lignin, using chemicals or enzymes. Lignin can be separated and allowed

to regain its heat value through burning. Cellulose and hemicelluloses are further broken down to simple C6 and C5 sugars, such as glucose, xylose, manose. Currently, there is no clear commercial or technical advantage differentiating the enzymatic or chemical pathways, even after many years of research and the development of near-commercial demonstrations. Both technologies remain unproven at the fully commercial scale, are under continued development and evaluation, and have significant technical and environmental barriers that are yet to be overcome. Given the current investments poured into improving these technologies, it is expected that some certain aspects of these technologies will move into commercialization in the next decade. This will allow greater volumes of lignocellulosic materials to be produced while avoiding many drawbacks of first generation biofuels.

### Sugar to Fuel

Once the cellulosic biomass is broken down to simple C5 and C6 sugar, there are a number of cutting edge technologies that can convert them into infrastructure-compatible fuels, for example, via the microbial metabolic pathways. Researchers are investigating various alternatives for the most efficient conversion to produce the most desirable fuels. Table 3 provides a comparison of these different technologies from conversion and energy efficiency stand point.

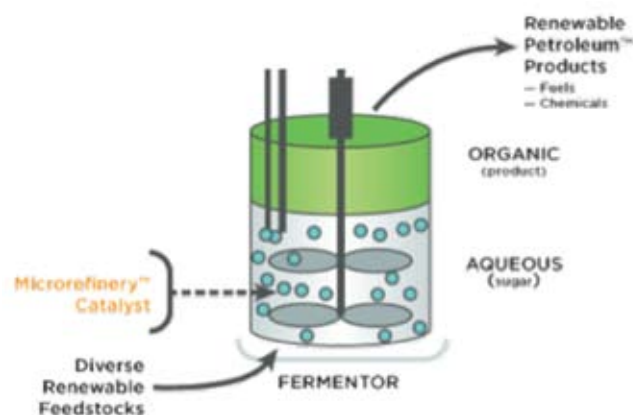
Most of the microbial metabolic pathway approach

except ethanol and butanol listed in Table 3 are in the development stages. Ethanol has the inherent challenge of not being fully compatible with existing infrastructure. Butanol is a promising gasoline replacement. The remaining technologies focus on long chain hydrocarbon molecules that can serve as diesel replacements. The clear differentiators include conversion efficiency and post fermentation modification requirements. Conversion technology determines the amount of the feedstock needed for production. Using the biological approach, the cost of the feedstock can account for more than 70% of the total production cost. Thus, the higher the conversion efficiency, the lower the cost of production. Post fermentation modification will be required if the selected microbial pathway cannot produce the finished product, thus needing additional chemical conversion steps after fermentation. These post fermentation steps contribute to additional operational and capital costs to the production economics. This is where technologies employed by companies like LS9, Inc. become important differentiating factors. Using fatty acid biosynthesis as a core technology platform, LS9 applies synthetic biology and metabolic engineering to produce high-performance proprietary drop-in fuels that are compatible with existing fuel distribution infrastructure. The same technology is also tweaked to produce valuable industrial chemicals. The Company has identified and pursued patent protection for the industry's most efficient microbial metabolic pathway, which enables it to produce renewable fuels in a one-step conversion process. Using proprietary synthetic biology tools, LS9 can genetically

Product	Type of Fuel	Pathway	Metabolic Mass Yield	Gal of Product /Ton Glucose	Energy Density (MJ/kg)	Infrastructure Compatible	Post Ferm Modification Required
Ethanol	Gasoline Replacement	Fermentation	51%	155	29.7	N	N
Butanol	Gasoline Replacement	Fermentation	41%	121	36.1	Y	N
Fanesene	Diesel Replacement	Mevalonate	25%	71	47.0	Y	Y
		Deoxyxylose	29%	83			
Ethyl hexadecanoate	Diesel Replacement	Fatty Acid	35%	98	39.4	Y	N
Pentadecane	Diesel Replacement	Fatty Acid	29%	90	47.0	Y	N

**Table 3:** Theoretical yields on glucose for selected microbial fuels from different metabolic pathways (modified from table 1 reference 4)

control and modify these core pathways to transform a spectrum of feedstock into a broad portfolio of products that meet the diverse demands of the petroleum economy. The finished products produced by genetically modified microorganisms are secreted outside the cells and are spontaneously separated from the fermentation broth, rendering great efficiency to a simple product recovery and separation process (Figure 4). The combined efficiency of the fermentation and downstream processing enables LS9's UltraClean™ Diesel to be competitive with crude oil at \$40-\$50 per barrel, without subsidies.



**Figure 4:** LS9 platform technology

Despite the great promise and excitement for next generation biofuels, there remain many challenges before commercialization. These challenges include the following:

- \* Most of the technologies are in various stages of development, pilot testing, and technology demonstration. Technical barriers remain for the next generation biofuel production.
- \* Production costs are uncertain and vary with the cost and availability of feedstock.
- \* Scale up risk remains when moving from lab or pilot scale to commercial scale.
- \* Capital requirements for commercial plants are unclear. Demonstration scale testing and monitoring are thus essential for providing accurate comparative data.
- \* Some of the currently existing technologies will probably not become fully commercially viable without significant additional government support in the near future.

- \* Lack of technology integration between the lignocelluloses to sugar and the sugar to fuel paths. More development effort is necessary to ensure that sugar to fuel conversion can be truly feedstock agnostic.

## Summary

The cause for transition from fossil fuels to renewable alternatives is compelling. In the near term, the biofuel industry will grow at a steady rate and rely on first generation technologies to meet the agreed-to environmental, sustainability, and economic policy goals. As we embracing the new and emerging technologies, the transition to an integrated next generation biofuel landscape is imminent, as the knowledge and experiences of scaling and perfecting the next generation biofuel to its commercial scale grow. Specifically for the path to biologically making biofuels, much remains to be done in terms of improving feedstock characteristics, improving overall conversion efficiency, lowering capital and production costs, and enhancing overall process integration. Concerted efforts across different technical areas, favorable funding environment, and strong and supportive government policies would be required to overcome these challenges in the coming years. Bring together the world's two most important value chains, energy and agriculture; biofuels will have profound impact on the future of humanity. Biofuel has become and will continue to be a growing industry in the coming years, as it provides regional energy security, stalls or reverses global climate change, and promotes sustainable economic growth.

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