Chapter 2
Biofuel Context

Abstract Liquid transportation fuels are used globally on a daily basis at a high consumption rate that is projected to rapidly increase over the next several decades. With dwindling, finite supplies of oil-based transportation fuels, there is an urgent need for alternative fuels. However, most proposed alternative fuels and fuel production schemes have potential impacts on food supplies, the environment, or finances. There are a diversity of potential starting materials for biofuel production, target fuel compounds, and organisms to use for production. Determining the best combinations of input, output, and process to satisfy the fuel demand while addressing additional societal concerns could lead to a sustainable fuel source.

Around the world and particularly in the United States, liquid transportation fuels for use in vehicles with internal combustion engines are a dominant, everyday convenience or necessity. Rough estimations of gasoline consumption in the US give that approximately 388.6 million gallons of gasoline are consumed each day (U.S. Energy Information Administration–www.eia.gov) and with a total population of about 314 million people (U.S. Census Bureau–http://www.census.gov/population/www/popclockus.html) which means that on average each person in the US uses almost one and a quarter gallons of gasoline each day. This level of consumption is a direct reflection of our reliance and the impact of liquid transportation fuels.

If the liquid transportation fuels that are currently used were plentiful and sustainable sources existed, the prominent role of these fuels in everyday life would not be a concern. However, since our current oil-derived fuels do not have sustainable sources, alternative fuels are desirable and necessary long term. A real issue arises when proposed avenues for sourcing alternative fuels have tradeoffs in other areas of everyday concern such as food supplies or money. Due to these potentially competing interests it is important to evaluate the different facets of potential biofuels and to consider their impact on fuel supply and other areas.

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2.1 Biofuels in the Energy Landscape

Most people readily acknowledge the need to develop additional or alternative sources of energy. What is typically not realized is the urgency to develop these alternative energies due to the extent of our (US and the world) current and projected future energy consumption. Above was given an example of the gasoline consumption in the US. The current human global energy demand of approximately 14.9 terawatt (TW) is predicted to rise to 23.4 TW by 2030 (an increase of more than 50 % in a just a fifth of a century) (Hambourger et al. 2009). Similar consumption projections hold true for liquid transportation fuels. Due to the finite amount of oil, there is an immediate need to develop alternative fuel sources that will work in our current engines and be easily accessible to the population. Biofuels (gasoline alternatives and biodiesel) are growing in popularity but there is no clear single fuel substitute for use with our current infrastructure.

2.1.1 Current Biofuel Situation

The term biofuel is a broad umbrella term referring to a fuel that is derived from a biological starting material. As a general term, there are a vast number of different biofuels that have been proposed that vary in combinations of starting material and target fuel. The challenging decision is to determine the best process for efficient production of large quantities of fuel when choosing from different starting materials (corn, corn stover, switchgrass, sugar cane/beets, soybeans, vegetable oil, etc.) to potentially produce different end products (ethanol, propanol, butanol, isoprenes, diesel). Since the goal of developing alterative fuels is to increase the availability of fuels, care must be given to how much energy goes into a production process to yield more usable energy. The processing, separation, and purification of biofuels use up energy that has to be accounted for when evaluating energy-efficient fuel sources. While ethanol production from corn is one classic example of first-generation biofuels, it also does not have a very high net gain of energy.

Production of biofuel from algae is one option for using a low amount of input energy for producing and processing its biofuel. Growth of algae is relatively simple with minimal growth requirements and sunlight is a primary energy input due to photosynthetic capabilities. Furthermore, algae are easily harvested and processed since culture takes place in a liquid mixture. Harvesting, transportation, and processing of land-based plant material are not as easily accomplished.

Processes for production of biofuels that utilize other photosynthetic microorganisms (such as cyanobacteria) will offer the same benefit as algae if the efficiency of fuel production with minimal energy usage is comparable. Larger eukaryotic organisms also have more metabolic processes occurring simultaneously and therefore use more of their energies on undesirable products. Microbial production of biofuels can be engineered for a maximum production of the biofuel, which will optimize the conversion of the energy supplied to the desired product.

2.1.2 Starting Materials: Feedstocks

Energy is neither created nor destroyed and therefore we need to find the most efficient way to take existing energy on our planet and convert it into fuel that we can use to power our current and future lifestyle. On the molecular level, breaking carbon–carbon bonds are the main source of energy but it takes just as much energy to make them as to break them. Currently, we are tapping into a source of long-chain carbon bonds in the form of crude oil stored under the surface of the earth but the formation and storage of this substance happens very slowly over time and we will run out of this source sooner than later. We need to find a solution that creates carbon–carbon bonds continuously and does so in a way that outputs more fuel than we use to produce the fuel.

Currently, the most commonly considered starting material for a biofuel production process is plant biomass (lignocellulosic biomass). Lignocellulosic biomass consists of cellulose, hemicellulose, and lignin that are three types of sugar polymers. Cellulose is a chain of glucose, hemicellulose contains xylose, glucose, mannose, galactose, and arabinose in varying quantities depending on the organism, and lignin is a chain of phenylpropanoid units. In order to use the sugars in lignocellulosic biomass, these polymers need to be broken down into their monomers, and that can be done enzymatically or chemically. Cellulases are used to break down cellulose. The enzymatic process is generally preferred due to its mild conditions but the biomass does have to be pretreated by steam or dilute acid to be digestible by the cellulases.

A number of organisms natively use feedstocks that are continuously produced naturally to grow and form carbon–carbon bonds as a part of their natural functions. Different types of organisms use different energy sources. Lignocellulosic biomass from trees and other wood sources as well as simple sugars from plants both provide carbon–carbon bonds that can be broken down for energy which is a much more sustainable process than using fossil fuels, but it is still a roundabout way of using the sun to photosynthesize plant mass and then break down the plant mass to produce fuel. This is why using photosynthetic organisms that produce biofuel directly from the sun's energy has gained considerable attention as a more permanent long-term solution.

Different classes of organisms undergo different processes that have the capability of producing a wide range of carbon chains. A diversity of metabolic capabilities exists in photosynthetic and non-photosynthetic prokaryotes and eukaryotes that can result in production of carbon-containing compounds with chain lengths ranging from 2 to 24 carbons. Carbon bonds take a relatively high amount of energy to create and therefore organisms tend to only make the bonds that benefit their growth and survival. Because of this, ethanol, a two-carbon molecule, is one of the easiest targets for sustainable fuel production. With each additional carbon added onto the chain as in propanol or butanol, it gets much harder to produce the molecules in organisms because the organisms will choose thermodynamically easier paths to process its metabolites and will not favor the upstream, high energy-requiring reaction.

2.1.3 Target Fuel Compounds

Finally, recent advances in synthetic biology have enabled scientists to use more commonly used industrial or laboratory organisms such as *Escherichia coli*, *Corynebacterium glutamicum,* and *Saccharomyces cerevisiae* as platforms for metabolic engineering, expanding native functionality by both broadening substrate range and extending chemical production capabilities (Jarboe et al. 2010; Krivoruchko et al. 2011). What this translates to is that when selecting a desirable target chemical for production, we are not limited to only those compounds that are natively produced in an organism. If a desirable chemical is identified, it is often possible to design a method for biological production of that chemical. In representative cases, Liao et al. have engineered butanol production in *E. coli* by introducing genes from *Clostridium acetobutylicum* (Astumi et al. 2008) and using systems approaches to explore ways to improve butanol tolerance in *E. coli* (Senger and Papoutsakis 2008; Lee et al. 2008).

Thus, it is possible to consider a variety of different chemical compounds as possible biofuels based solely upon their chemical characteristics without initial regard to the feasibility of production. A number of these compounds have been identified and targeted as compounds that are either natively produced or can be engineered into organisms (Table 2.1).

Due to the diversity of organisms, starting materials, and potential target compounds, biofuel production processes take many different forms. While incremental research progress is being made in a large number of different biofuel processes, it is still not clearly defined as to what process combinations will prove most fruitful. It is now becoming possible to answer this question from a more global, top-down perspective by independently evaluating chemical characteristics, starting material characteristics, and organism traits. By identifying the most promising starting materials and target compounds, the most suitable organisms can be targeted as host bioprocessing platforms.

2.2 Broader Impacts of Biofuels

2.2.1 Effect on Food Supply

There is a lot of concern as to how biofuel production will impact the global fuel supply since many biofuel sources are grown as crops. Corn, sugar cane, switch grass, and soybeans are the main crop-based energy sources and out of all of them, corn is by far the largest competitor with possible food production. Because the

Table 2.1 Examples of potential target biofuel chemicals and organisms that have been shown to produce that chemical

Biofuel	Pathway	Examples of organisms
Ethanol	Native	Zymomonas mobilis
		Pichia stipitis
		Clostridium thermocellum
		Clostridium phytofermentans
		Saccharomyces cerevisiae
		Escherichia coli
	Imported	Corynebacterium glutamicum
Biobutanol	Native	Clostridium acetobutylicum
	Imported	Escherichia coli
		Saccharomyces cerevisiae
Lipid fuels	Native	Cyanobacteria and algae
		Yarrowia lipolytica
	Imported	Escherichia coli
Hydrogen	Native	Cyanobacteria and microalgae
	Imported	Escherichia coli
Higher alcohols and alkanes	Native	Vibrio furnissii
	Imported	Escherichia coli

corn produced for biofuel is engineered to optimize its starch content, it is inedible and is taking up farm land that could be used to produce corn for consumption. This same argument and concern holds true for almost all land-based plant matter that is considered as a starting material for biofuel production. The common argument is that if arable land is used, it would be better used for food purposes rather than fuel purposes.

Potential alternatives to land-based plants are the use of aquatic, photosynthetic organisms such as algae or cyanobacteria. Biofuel production from algae will not affect the food industry because it can be grown in diverse water systems and only has to have access to sunlight. Most energy options depend ultimately on the energy gained from the sun via photosynthesis to some extent. Therefore, in a continuous process, photosynthesis is usually the limiting process in energy acquisition. Solar energy, reaching the surface of the earth at a rate of approximately 120,000 TW, is a sustainable resource exceeding predicted human energy demands by >3 orders of magnitude. If solar energy can be concentrated and stored efficiently then it has the capacity to provide for future human energy needs (Bungay 2004). There is a large amount of research going into optimizing photosynthesis in many different organisms for the advancement in energy production avenues. As with many other organisms, the potential yield of algal biofuel is limited by the fundamental inefficiencies in the photosynthetic conversion of solar energy to biofuel. As of now our direct use of sunlight through solar panels is far exceeding that of indirect use through photosynthesis. Compared with synthetic solar panels with reported 30 % efficiencies, photosynthesis has a maximum efficiency of 8–10 % (Hambourger et al. 2009).

2.2.2 Environmental Impact: Greenhouse Gas Emissions

Global warming awareness has forced the fuel industry to reduce their carbon footprint and there is a heavy focus to minimize the carbon emission levels of perspective biofuels. Both the carbon produced for the production of the biofuel and the burning of the biofuel are considered in the overall emissions amount. This also takes into consideration the carbon that photosynthetic organisms absorb which can make their carbon emissions negative. General consensus to date suggests that from a greenhouse gas emissions standpoint, biofuels would be a better option than oil-based fuels.

2.2.3 Economic Feasibility

In addition to all the other considerations that can influence the choice of biofuel and biofuel production process, economics may be the most influential. Due to the size of the automotive and fuel industries, there are considerable investments made in infrastructure, production facilities, and even government policy. The more practical aspect of the economics is the cost of the final fuel product when considering production costs. Blending larger amounts of biofuels with gasoline while keeping the price of gas the same will ensure a path to sustainability and fuel independence.

2.2.4 Sustainability

All of the different considerations related to biofuels can be restated as an issue of sustainability. While there are numerous contexts and definitions for sustainability, the consistent general concept involves maintaining and preserving the current quality of life for current and future generations of humans within the context of our natural environment. The idea of sustainability is not limited to environmental impacts alone but is comprehensive in touching upon almost every salient aspect and concern that motivates the continued development of biofuels.

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