

Biotechnology for Production of Fuels, Chemicals, and Materials from Biomass

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ABSTRACT

Biological systems can convert renewable resources, including lignocellulosic biomass, starch crops, and carbon dioxide, into fuels, chemicals, and materials. Ethanol and other products are now derived from starch crops, such as corn. Enzyme-based technology is under development for conversion of lignocellulosic biomass (e.g., wood, grasses, and agricultural and municipal wastes) into fuel ethanol. The simultaneous saccharification and fermentation (SSF) process is employed to convert the cellulose fraction into ethanol at improved rates, higher yields, and higher ethanol concentrations than using sequential processing through careful selection of improved cellulase enzymes and fermentative microorganisms. Medium-BTU gas can be derived from lignocellulosic biomass by anaerobic digestion and cleaned up to a pipeline-quality gas. A high-solids fermenter achieves higher gas generation rates than conventional devices and promises to help make such gas economical. An extensive collection of more than 500 productive strains of microalgae has been established to produce lipid oils for diesel fuel and other compounds from carbon dioxide. Acetyl CoA carboxylase (ACC) has been shown to be a key enzyme in lipid oil synthesis, and genetic engineering approaches are being applied to enhance the rates and yields of product formation. In addition to fuels, a biorefinery could produce a wide range of chemicals and materials through microbial conversion of renewable resources, and technology is being developed for production of chemicals and materials from biomass.

Index Entries: Ethanol; methane; biodiesel; chemicals; biomass.

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INTRODUCTION

About 40% of the total of 8.6×10^{10} gigajoules (GJ) (81.5 quads) of energy consumed in the United States in 1991 was derived from petroleum, representing approx 3.5×10^{10} GJ (34 quads) of energy. Nearly half of the petroleum used in this country is imported. If we look further at petroleum consumption, we see that the largest use, just over 64%, is for transportation. Furthermore, the second largest petroleum-consuming sector is industry, representing about 25% of the total petroleum used (1).

Because of this heavy dependence on petroleum, particularly on imported supplies, the United States is vulnerable to oil supply disruptions. This has strategic implications in that our vital supplies of oil can be shut off in times of hostility. In addition, a large fraction of our balance-of-payments deficit, on the order of 44%, is attributed to imported petroleum (2). Therefore, it is desirable to look for alternatives to petroleum.

One way to replace petroleum is through biological conversion of indigenous biomass resources into products now derived from petroleum. Many biological reactions offer high selectivity, maximizing product yields. Furthermore, biological reactions occur at mild conditions that reduce the cost of containment. The modern application of biological transformations, known as biotechnology, is also an evolving field that has great promise for substantial improvements and significant cost reductions. One disadvantage of biological conversions is that biological reactions occur at slow rates; it is important to increase the reaction rates. Another concern is the often low product concentrations, which result in high product recovery costs with existing technology. Also, production of multiple products in some systems results in lower yields of targeted products.

In this article, the application of biotechnology will be described to reduce our dependence on petroleum by producing fuels, chemicals, and materials from biomass. In particular, current technology for fermentation of starch crops into ethanol will be discussed as well as emerging technology for lignocellulosic biomass conversion into ethanol as a replacement for gasoline. Another fuel application included is methane production by anaerobic digestion of biomass to replace natural gas either for industrial applications or for transportation. The use of microalgae to produce lipid oils for conversion to a diesel fuel substitute will be summarized. Finally, the application of biotechnology to produce commodity chemicals and materials will be looked at briefly.

ETHANOL FROM CORN

The largest-scale application of biotechnology in the United States today is the conversion of starch from corn and other crops into ethanol.

Corn is composed of about 70–75% starch, with the remaining fractions consisting of about 10% protein, 4.5% oil, and 10–15% other materials, such as fiber, ash, and sugars (3). Corn can be converted into ethanol in the dry milling process, in which the corn is first milled to a fine particle size. Enzymes are added to the corn to break down the starch into the sugar glucose, which yeasts ferment into ethanol. About 9.5–9.8 L (2.5–2.6 gal) of ethanol are produced/bushel of corn processed in the dry milling operation. 7.3–7.7 kg (16–17 lb) of carbon dioxide evolves during the dry milling process. In addition, an approximately equal weight (7.7–8.2 kg (17–18 lb)) of an animal feed coproduct called distillers dry grain with solubles (DDGS) is produced. DDGS contains about 27% protein (4) and is typically sold as animal feed.

Another process used for the production of ethanol from corn is the wet milling process. In wet milling, corn is first subjected to a series of processing steps to remove the oil while also separating off an animal feed coproduct called corn gluten feed, which contains about 21% protein, and a second animal feed coproduct called corn meal, which contains about 60% protein. The starch remaining after this separation is converted into sugars by enzymes, and the sugars are fermented to produce about 9.5 L (2.5 gal) of ethanol/bushel of corn (4). Wet milling facilities produce about two-thirds of the approx 3.8 giga liters (GL) (1.0 billion gal) of ethanol now sold annually in the United States.

Currently, ethanol produced from corn is blended with gasoline. Usually the blend contains 10% ethanol with 90% gasoline. Such blends reduce carbon monoxide and unburned hydrocarbon emissions by ensuring more complete combustion of gasoline because of the presence of oxygen in the ethanol molecule (5–7). Ethanol can also be converted into ethyl tertiary butyl ether (ETBE) for blending with gasoline, giving similar improvements in terms of carbon monoxide and hydrocarbon emissions. Although direct blends of ethanol increase the vapor pressure of the overall gasoline mixture by about 3.4 kilopascals (kPa) (0.5 psi), ETBE lowers the vapor pressure of gasoline, thereby reducing smog formation caused by evaporative and running losses of fuel (8,9). Alternatively, ethanol can be used as a “neat” fuel (virtually 100% ethanol) as in Brazil. Neat ethanol has the potential to reduce smog formation in cities, such as Los Angeles, substantially, because ethanol has a lower volatility than gasoline and is less photochemically reactive (2,10).

ETHANOL FROM LIGNOCELLULOSIC BIOMASS

Ethanol can be produced from sugar crops and lignocellulosic materials as well as from starch crops. Over 11 billion liters (3 billion gallons) per year of ethanol are now produced in Brazil from sugar crops (11). However, sugar prices are controlled at about \$0.42–\$0.44/kg (\$0.19–\$0.20/lb)

in the United States (12), making sugar too expensive for ethanol production in this country compared to starch. Lignocellulosic biomass, such as agricultural, forestry, and municipal wastes, as well as short rotation woody crops and herbaceous energy crops offers a feedstock even lower in price than starch for ethanol production (2,10). However, lignocellulosic biomass is more resistant to breakdown, and substantial advances have been required in the technology for producing ethanol from lignocellulosic biomass at prices competitive with those of starch, even though the substrate itself is less expensive.

Although carbon dioxide is released during the manufacture and use of ethanol from lignocellulosic biomass, carbon dioxide is recycled to form new biomass. This means that no net carbon dioxide buildup results from the use of lignocellulosic biomass for ethanol production, avoiding possible contributions to global climate change caused by the greenhouse effect (2,10). In addition, sufficient lignocellulosic biomass resources are available in the United States to produce enough ethanol to replace all gasoline used in this country (2,10). Thus, large-scale substitution of ethanol for gasoline reduces our vulnerability to disruptions in the supply of imported oil and substantially improves our balance of trade for imported oil. Finally, ethanol production from lignocellulosic biomass offers a new market for agricultural products and could help to revitalize the farm economy.

Several forms of lignocellulosic biomass can be used to produce ethanol. These include underutilized wood in existing forests, agricultural residues, such as corn cobs or corn stover, and municipal solid waste, which together could be used to produce approx 189 GL (50 billion gallons) of ethanol annually as shown in Table 1. In addition to waste streams, dedicated wood or herbaceous crops could be grown on a large scale (energy crops) for ethanol production. As shown in Table 1, land is potentially available to grow between 763 and 2448 megatonnes (842–2698 million tons)/yr of lignocellulosic energy crops, enough to yield between 318.7 and 1021.2 GL (84.2–269.8 billion gallons)/yr of ethanol. Another 262 megatonnes (289 million tons) of biomass could be grown on forestland, potentially yielding 109.4 GL (28.9 billion gallons) of ethanol. Because approx 530 GL (140 billion gallons) of ethanol would be required to replace all gasoline used in the United States on an equivalent mileage traveled basis (2), the potential amount of ethanol derived from lignocellulosic biomass in Table 1 is more than sufficient to provide enough ethanol to replace all gasoline now used in the United States.

As shown in Fig. 1, even though the external appearance of various forms of biomass is substantially different, they are all quite similar in composition. The largest fraction of lignocellulosic biomass, on the order of 40–50%, consists of cellulose, a polymer of glucose sugar. The next largest fraction, typically 25–35%, is hemicellulose, a polymer of a variety of sugars often predominated by xylose. The remaining portion of the

Table 1
Potential Lignocellulosic Biomass Availability and Ethanol Production (2)

	Biomass availability		Ethanol potential		Energy potential	
	Megatons	10 ⁵ Tons	GL	Billions of gallons	GJ	Quads (2)
Waste material						
Agricultural	203	224	84.8	22.4	1.8×10 ⁹	1.7
Forestry	167	184	69.6	18.4	1.5×10 ⁹	1.4
Municipal solid waste	111	122	34.8	9.2	7.4×10 ⁸	0.7
Cellulosic biomass						
Cropland						
Idled (1988) ^a	358-	395-	149.5-	39.5-77.6	3.2×10 ⁹	3.0-5.9
	704	776	293.7		6.2×10 ⁹	
Excess (2012) ^a	680-	750-	283.9-	75.0-150.0	6.0×10 ⁹ -	5.7-11.4
	1361	1500	567.8		1.2×10 ¹⁰	
Potential	405-	447-	169.2-	44.7-119.8	3.6×10 ⁹	3.4-9.1
	1087	1198	453.4		9.6×10 ⁹	
Forest land	262	289	109.4	28.9	2.3×10 ⁹	2.2
Total	1507-	1661-	617.3-	163.1-348.7	1.3×10 ¹⁰ -	12.4-26.5
	3190	3517	1319.8		2.8×10 ¹⁰	

^aValues are for biomass production potential in 1988 and 2012 based on expected availability of land, and current and future biomass production (2).

^bEthanol rated at the lower heating value.

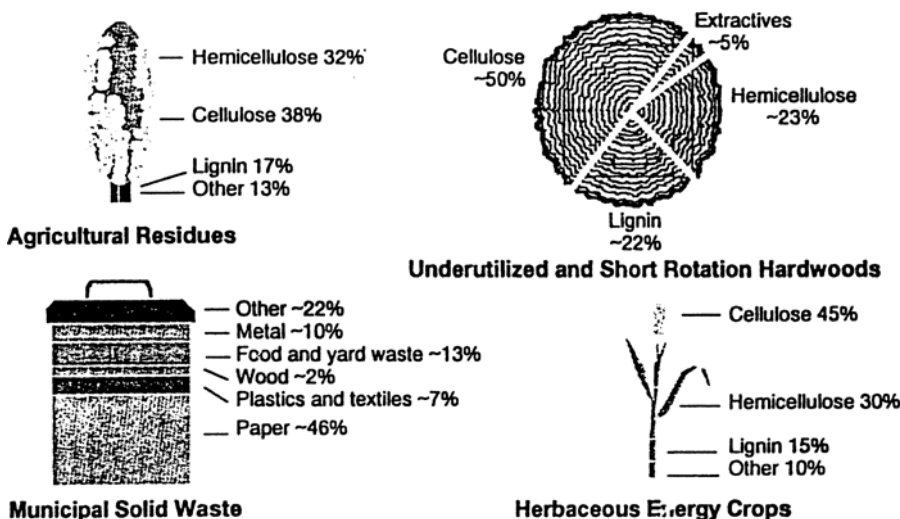


Fig. 1. Composition of lignocellulosic biomass.

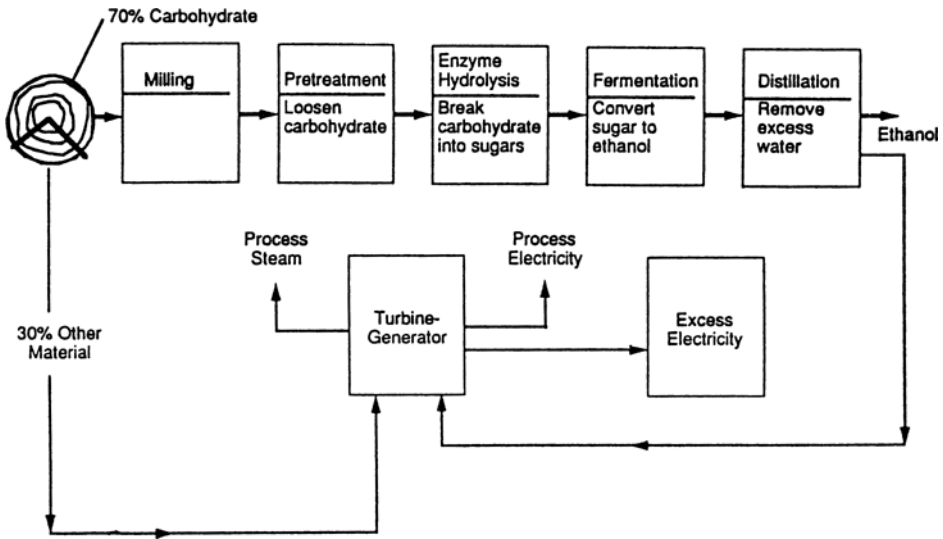


Fig. 2. Typical processing sequence to convert the cellulose and hemicellulose fractions of lignocellulosic biomass into ethanol while burning the lignin fraction to provide heat and electricity.

biomass, on the order of 15–25%, consists of lignin and other materials called extractives and ash.

As shown in Fig. 2, the processing of lignocellulosic biomass to ethanol proceeds through a series of operations, starting with milling and pretreatment to overcome the natural resistance of lignocellulosic biomass to form sugars. Cellulase enzymes are produced and added to the pretreated biomass to breakdown the cellulose to form glucose, which yeast or other organisms ferment to ethanol. Hemicellulose can be broken down during the pretreatment and/or hydrolysis step to produce sugars, such as xylose for fermentation to ethanol. The mixture of ethanol, residual biomass, and water from the fermentation system is then sent to distillation or another purification process for recovery of the ethanol. The residual solid material recovered from the product stream is burned as a boiler fuel to produce process steam and electricity. The excess electricity or heat beyond that required for the process is sold to generate additional revenue (13).

Over the years, front-running process options have been identified for each one of the individual steps in conversion of lignocellulosic biomass to ethanol. This article will focus on some of the advances achieved in cellulose conversion. The key requirement in this area is to achieve high yields of ethanol from the cellulose fraction. A number of evaluations have been conducted on various options for ethanol production, and the simultaneous saccharification and fermentation (SSF) process has been identified as a promising route (14). In SSF, pretreated biomass, cellulase enzyme from the enzyme production step, and the fermentation microorganism (typically yeast) are all added to the same fermentation

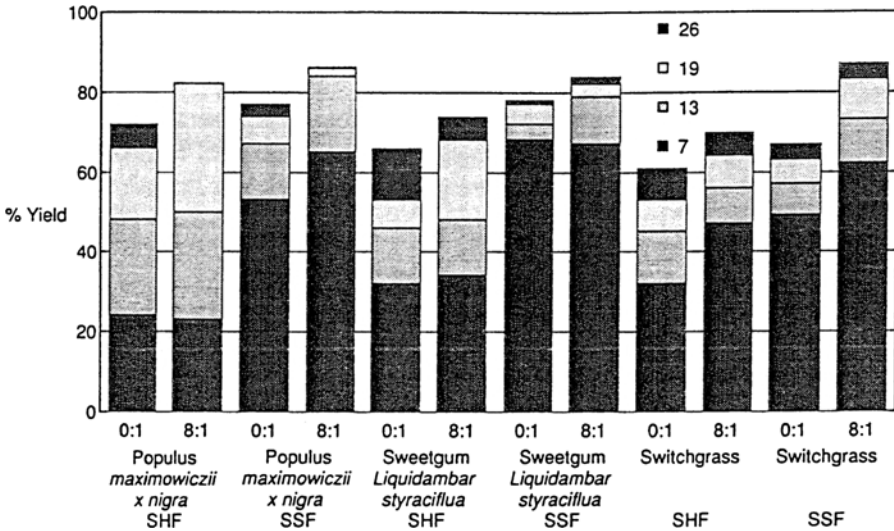


Fig. 3. Comparison of final ethanol yields for SHF and SSF processes with varying cellulase enzyme loadings (with [8:1] and without [0:1] β -glucosidase supplementation).

vessel. The enzyme catalyzes the breakdown of the cellulose to glucose, which the yeast rapidly converts into ethanol. By having the yeast present to rapidly convert glucose to ethanol, accumulation of the sugars that severely inhibit enzyme action is minimized. As a result, higher rates, yields, and concentrations of ethanol are achieved in the SSF process than if the saccharification and fermentation were done sequentially (SHF). Furthermore, by combining these two processes in one set of fermenters, about half as much equipment is required for the fermentation operation, substantially decreasing capital costs. Because not many organisms other than yeast are tolerant to high ethanol concentrations in the fermentation broth, SSF also reduces the probability of contamination by unwanted microorganisms that would rapidly consume glucose.

Over the last several years, a number of studies identified combinations of cellulase enzymes and yeast to speed the rate of conversion of lignocellulosic biomass to ethanol (15-19). Figure 3 illustrates the results of conversion of lignocellulosic biomass into ethanol for selected feedstocks with the SSF process and the SHF process. As shown for each of the feedstocks, the SSF process is able to achieve higher yields of ethanol at the same enzyme loading as sequential processing by the SHF approach. Furthermore, enhanced performance results from the addition of the enzyme β -glucosidase to the fermentation broth. β -glucosidase converts the glucose sugar dimers called cellobiose into glucose molecules for fermentation to ethanol. Cellobiose is known to be a powerful inhibitor of cellulase enzyme, and it appears that by adding β -glucosidase, the accumulation of cellobiose in the fermentation broth is minimized. Therefore,

higher yields are achieved when the β -glucosidase is added to the fermentation broth than when it is not. Similar benefits have been found by employing cellobiose-fermenting yeast or cellulase enzyme with greater β -glucosidase activity in the fermentation system to speed rates, increase yields, and increase concentrations of ethanol (17-19).

Seven or 8 yr ago, it required on the order of 2 wk to convert the cellulose into ethanol in an SSF process at yields of approx 70% (18,20). By selection of cellulase enzyme with enhanced characteristics, such as higher levels of β -glucosidase, as well as selection of yeasts that perform well under SSF conditions, on the order of 90-95% cellulose conversion into ethanol can be achieved in about 3-7 d for a variety of feedstocks (15-17).

Xylose cannot be directly fermented by yeast to ethanol at high yields; however, recent research has been successful in converting xylose into xylulose, which can be fermented to ethanol (21,22). In a highly promising approach, genetic engineering of organisms has been successfully applied to utilize the C₅ fraction of biomass productively (23,24). Micro-aerophilic organisms have also been identified that are capable of converting xylose to ethanol (25).

If we look at the combination of improvements in ethanol production by selecting better enzymes and yeast combinations, the successful development of technology for converting the xylose fraction of biomass into ethanol, and advances in various other steps in the process, we see that the price of ethanol has been reduced substantially in the last 10 yr. In 1980, the projected cost of ethanol production from lignocellulosic biomass by enzymatic conversion was on the order of \$0.95/L (\$3.60/gal) (26). A recent study conducted by Chem Systems has estimated the cost at about \$0.34/L (\$1.27/gal) (27). Furthermore, a number of opportunities have been identified to reduce the cost from \$0.34/L (\$1.27/gal) to \$0.18/L (\$0.67/gal) (13). Key target areas include improved glucose and xylose yields from pretreatment, increased ethanol yields of 90% or greater from cellulose and xylose fermentations, decreased stirring and pretreatment power requirements, better productivities through continuous processing, low-cost production of octane enhancers or chemicals from lignin, increased ethanol concentrations, and reduction of fermentation times. Feedstock costs are a significant fraction of the final product selling price, so improvements in feedstock production, collection, and genetics could provide additional cost reductions through economies of scale for larger ethanol plants, decreased feedstock costs, and less nonfermentable feedstock. Many of these goals have been met individually, and the evidence that the rest can be achieved is great; the primary need is to meet them simultaneously. Fortunately, there are enough options for lowering the selling price of ethanol so that not all the technical goals must be achieved to reach the target. At the target price, ethanol would be competitive with gasoline derived from oil at \$25.00/barrel, and with an aggressive R&D effort, the goal could be met in about the year 2000 (2).

METHANE FROM BIOMASS

Anaerobic digestion is currently used to convert waste streams, such as municipal sewage sludge and industrial waste, into methane gas. In these operations, bacteria break down organic matter to form soluble sugars that are converted into organic acids by another set of bacteria, and finally converted into methane and carbon dioxide by a third set of organisms. This technology could also be used to convert lignocellulosic biomass into methane and carbon dioxide. The gas can be used directly as a medium-BTU gas, or the carbon dioxide can be removed to produce pipeline-quality methane. In 1991, 2.0×10^{10} GJ (18.8 quads) of natural gas were used in the United States, representing 25% of total energy consumption (1). Assuming similar yields of methane as for ethanol in Table 1, sufficient quantities of waste and lignocellulosic biomass could be available for large-scale application of anaerobic digestion technology.

The breakdown of cellulose to sugars is very slow, and reaction times of close to a month can be required to achieve high yields of methane. If economic application of anaerobic digestion technology for conversion of biomass to methane gas is to be achieved, the rates of fermentation to methane must be accelerated. Gas production rates increase as the solids concentration in the digester increases, and one approach to realizing this goal is high solids anaerobic digestion. However, in conventional fermentation vessels, solid contents are limited to about 5–10% biomass, because at higher concentrations, the viscosity of the fermentation broth becomes so thick that conventional agitators can no longer mix the material (28). Recently, a high solids device was developed that allows mixing of any biomass concentration. Various agitators were tested in this device, and one was selected that achieves good mixing with low-power requirements. The device consists of a cylindrical vessel with a horizontal axis. The agitator shaft runs along this axis with prongs protruding from the shaft in the radial direction at regular intervals. As the agitator shaft turns, the action of the rods gently pushes the biomass from side to side as the rods move through the mixture. This provides effective mixing of the biomass mixture at high-solids concentrations with low-power input (28).

Evaluations have been made of the impact of solids concentration on gas production rates (Fig. 4). Initially, solids levels were started off at around 5%, which is typical for conventional devices. Then the solids concentration was gradually increased, and the gas production rate also increased up to about 35–40% solids. Further increases in solids concentrations above that level resulted in lower gas production rates. Apparently, the moisture in the biomass was insufficient for the anaerobic digester microorganisms, and they started to lose viability as the solids concentrations increased further. Therefore, the high-solid fermenters are typically operated around 32–35% solids. At these concentrations, gas production

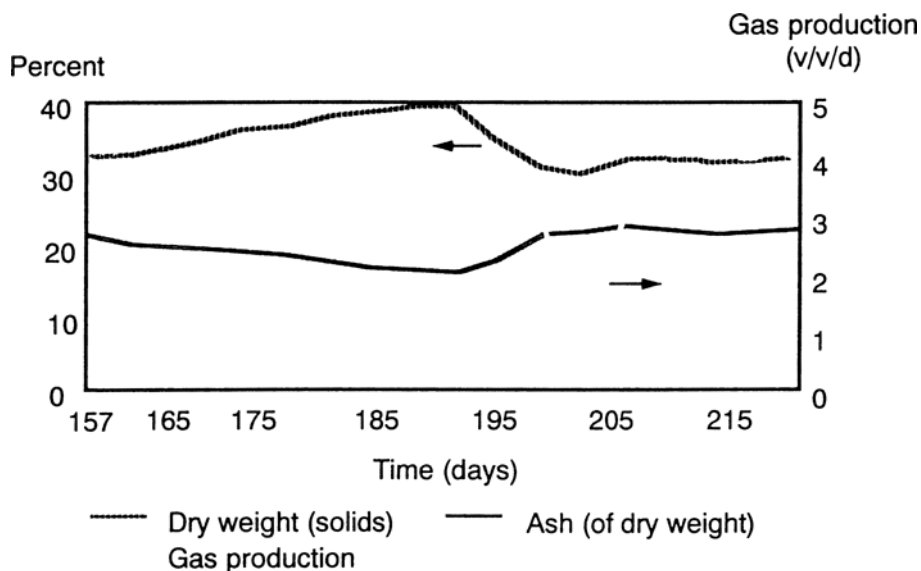


Fig. 4. Gas production rate of a high-solids reactor with changing solids concentration.

rates of seven to nine times those possible on conventional devices are achieved (28).

Through the application of high-solids anaerobic digestion, the price of methane from municipal solid waste has dropped to around \$4.27/GJ (\$4.50/million BTU) with a tipping fee of around \$44.00/tonne (\$40.00/t). Opportunities have been identified to drop the price further to under \$1.90/GJ (\$2.00/million BTU) for large-scale applications (29). Research needs include enhancing enzyme levels within the fermenters, improving organisms to tolerate wider pH differences within the system, and improving methods to maintain the stability of the fermenter (30).

DIESEL FUEL FROM MICROALGAE

Microalgae are single-celled plants that utilize the sun's energy to combine carbon dioxide and water to form a variety of products. They are unique in that they can form 60% or more of their body weight as lipid oils or triglycerides that can be converted into a low-sulfur diesel fuel substitute (31). Many algae are unusual in that they thrive in high-salinity water that is unsuitable for other purposes. Therefore, it may be possible to grow algae for diesel fuel production in the Desert Southwest where there is abundant, inexpensive, flat land available with plentiful high-salinity waters in aquifers. This would complement ethanol production from lignocellulosic biomass nicely in that as gasoline is replaced by ethanol, the fraction of petroleum that is used for diesel fuel production

must also be replaced. Currently, 17% of transportation energy is used in the form of diesel fuel (32).

To produce oil from algae, it is necessary to identify algae that are tolerant of very high salinity, high light intensities, and swings between high and low temperatures. It is also important that the algae produce high lipid oil concentrations and grow fast. More than 3000 strains of algae have been screened for these desirable characteristics for diesel fuel production. From these, a collection of about 500 strains shows great promise. Some of these strains have tolerance to high salinity and can grow quite rapidly at salinities more than twice that of sea water.

The key requirement for successful production of diesel fuel from microalgae is to achieve high oil content in the algae. Studies of algal characteristics have shown that algae will accumulate large amounts of lipid oils when they are deprived of key nutrients (33). For instance, when diatoms are deprived of silicon, they rapidly (within 24 h) increase lipid oil content within the cell to about 60% of their body wt (Fig. 5A). Similarly, green algae will produce about 60% of their body wt as lipid oils (within 48 h) when they are deprived of nitrogen. Studies of the biochemistry of diatoms have shown that the increase in lipid content of the algae correlates with increases in levels of acetyl Co-A carboxylase (ACC) activity (Fig. 5B). Therefore, it is thought that ACC is a key enzyme in lipid oil synthesis (33,34). Current efforts are focused on developing techniques to modify algae genetically to achieve high level of ACC activity with the goal of enhancing oil levels at high productivities.

Progress on oil production from algae has been substantial over the last 10 yr. Although algal oil production is a longer-term technology than either ethanol production or anaerobic digestion, the price of algal oil has dropped from approx \$4.49–\$4.76/L (\$17.00–\$18.00/gal) of diesel fuel in the early 1980s to around \$0.92/L (\$3.50/gal) now. Furthermore, opportunities have been identified to reduce the price of diesel fuel to about \$0.26/L (\$1.00/gal). The primary requirement is to enhance the growth rate of algae while achieving high lipid oil concentration. However, a key cost component of oil production is the cost of carbon dioxide (31), and if penalties were levied on carbon dioxide release because of concerns about the impact on global climate change, the economics of algae oil production would change dramatically. In fact, algae may offer one of the best ways to reduce emissions of carbon dioxide from fossil fuel power plants (35).

CHEMICALS AND MATERIALS FROM BIOMASS

The previous discussions have centered on fuel production by application of biotechnology. However, these concepts could be extended to produce a wide range of chemicals and materials from biomass in a bio-refinery. In such a case, sugars could be derived from a variety of feedstocks, such as sugar crops, starch crops, and lignocellulosic biomass.

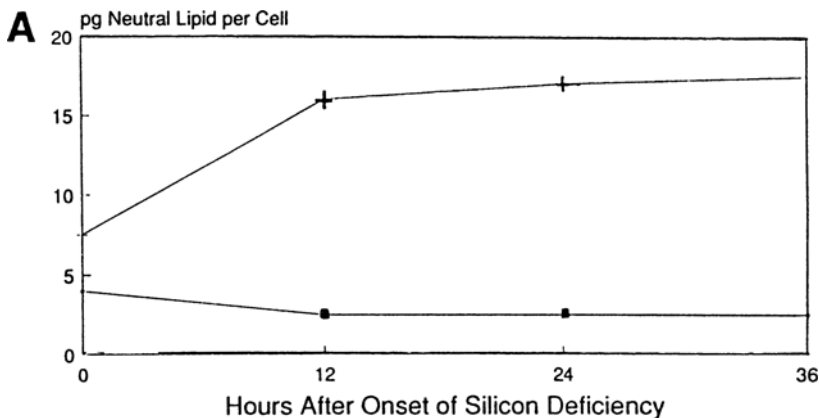


Fig. 5A. Change in neutral lipid content within diatoms after termination of silicon supply. + Si-deficient cells; —■— Si-replete cells.

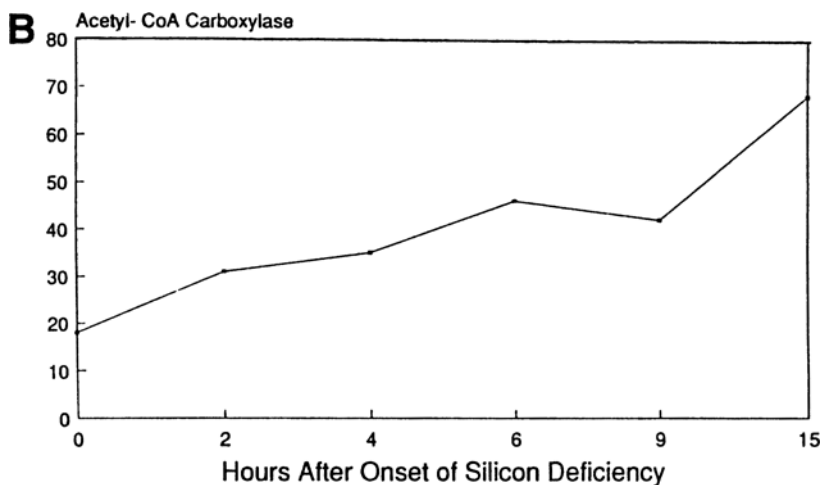


Fig. 5B. Accumulation of acetyl Co-A carboxylase enzyme in diatoms after termination of silicon supply.

These sugars could be biologically converted into a variety of chemicals, such as glycerol, lipid oils, acetone, *n*-butanol, butanediol, lactic acid, isopropanol, and acetic acid, as well as ethanol and methane gas. They could also be used to form biological materials, such as xantham gum, protein polymers, and polyhydroxybutyrate. Furthermore, the sugars or some of the fermentation products can be chemically converted into a variety of products, such as furfural, glycol, methyl ethyl ketone, and adipic acid. The remaining fraction of the biomass, such as the lignin in the case of lignocellulosic feedstocks, could be converted through hydrogenation processes into materials, such as phenols, aromatics, and olefins, or simply burned as a boiler fuel to power the overall process.

Table 2
Volumes and Prices of Major Organic Chemicals in the United States

Chemical	Production		Price	
	Millions of tonnes	Billions of pounds (37)	\$/kg	\$/lb (38)
Ethylene	17.79	39.23	.49	.22
Propylene	9.99	22.02	.37	.17
Ethylene dichloride	6.31	13.92	.12	.05
Benzene	5.34	11.78	.45	.20
Vinyl chloride	5.31	11.70	.40	.18
Methyl <i>tert</i> -butyl ether	4.35	9.60	.46	.21
Ethylbenzene	4.18	9.22	.61	.28
Styrene	4.09	9.01	.76	.34
Methanol	3.92	8.65	.12	.05
Terephthalic acid	3.47	7.66	.58	.26
Toluene	3.08	6.80	.35	.16
Formaldehyde	2.92	6.43	.13	.06
<i>p</i> -Xylene	2.46	5.43	.52	.24
Ethylene oxide	2.38	5.24	1.05	.48
Ethylene glycol	2.24	4.93	.47	.21
Cumene	1.94	4.28	.51	.23
Acetic acid	1.64	3.61	.37	.17
Phenol	1.58	3.49	.76	.34
1,3-Butadiene	1.32	2.91	.57	.26
Propylene oxide	.125	2.75	1.27	.58
Acrylonitrile	1.20	2.65	.59	.27
Vinyl acetate	1.20	2.65	.69	.31
Cyclohexane	1.05	2.31	.50	.23
Acetone	0.97	2.13	.47	.21

Currently, on the order of 5.28×10^9 GJ (5 quads) of fossil fuels are used annually for production of chemicals. Of this, approx 1.06 GJ (1 quad) is from petroleum (36). Assuming similar yields for chemicals as for ethanol in Table 1, potential quantities of lignocellulosic biomass are more than sufficient to produce enough chemicals to displace those made from petroleum. A wide range of organic chemicals are derived from petroleum feedstocks from the highest volume organic product, ethylene, through a wide range of other products, such as acetone (Table 2). When these products as well as other lower-volume chemicals are examined, it is found that the larger the volume of the product produced, the lower the unit price of that material. Therefore, in order to make a substantial impact on the production of chemicals in this country, it is necessary to achieve low prices. This is a major challenge for the application of biotechnology to the production of chemicals.

Two kinds of products can be made from renewable feedstocks. In one class, conventional chemicals can simply be derived from biomass. Examples include acetaldehyde, acetic acid, acetone, *n*-butanol, ethylene, and isopropanol. The advantage to such products is that the market is already established, and minimal effort is required to integrate these products into existing markets. On the other hand, competition will be strictly on the basis of price, because no performance advantages are possible for products derived through biotechnology over those produced from petroleum. Because established products are manufactured in plants with capital that is all or partially paid off, the advantage often is with existing products and processes, unless the cash cost of production of such products is greater than the total cost of biologically produced products, including return on investment.

New products could also be derived through biotechnology that are unique to bioprocessing. Materials with desirable properties that are not easily matched by petrochemical processing are particularly promising targets. Examples include new adhesives, biodegradable plastics, degradable surfactants, enzymes for catalysis, and various plastics and polymers. Substantial product development is, however, required to meet the specific needs of the customer. For such products, competition would probably be based on performance of the materials, and biomaterials might provide unique niches for initial introduction into the marketplace. Therefore, less price pressure would exist initially for such new products. However, to have a substantial impact on petroleum consumption, it is necessary to ensure that large markets would eventually result.

The same substrates, corn or lignocellulosic biomass, could be used to produce chemicals and materials as to produce ethanol. It would be possible to modify dry milling operations now employed for ethanol production to make such products. Corn would be ground, and enzymes added to it to convert the starch into glucose sugar. Appropriate organisms could then convert the glucose sugar into the desirable products and coproducts for this process. The solid fraction left after conversion of glucose into useful products could once again be sold as an animal feed coproduct, such as DDGS. Coproduct markets might be limited, however, and caution must be taken in considering their impact on overall economics, especially for large-scale implementation.

Wet milling is now applied to convert corn starch into lactic acid and a variety of other products. As before, corn is first processed to remove the corn oil and protein animal feed coproducts prior to conversion of starch to glucose for fermentation to desirable products. The primary requirement is to develop a wider range of products that can be made from glucose.

By analogy to corn dry and wet milling operations, lignocellulosic biomass could be converted to a variety of chemicals and materials. A sequence of operations comparable to those employed for ethanol production would be used to pretreat the biomass to open its structure for

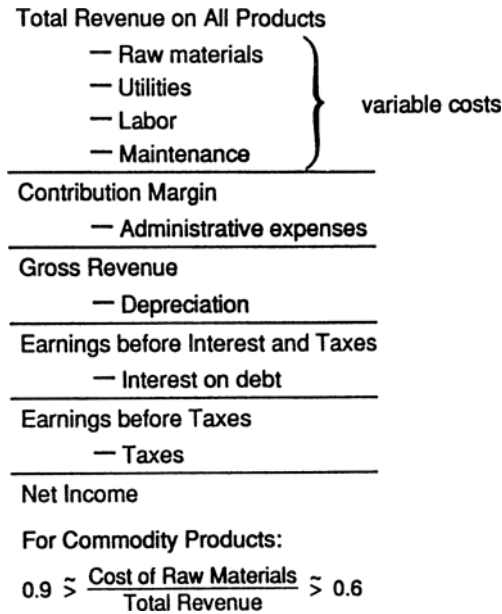


Fig. 6. Typical income statement for production of chemicals and materials from biomass.

conversion of the glucose, xylose, and other sugars derived from the cellulose and hemicellulose fractions into target products. The primary requirement is to identify satisfactory organisms that can convert these sugars into the chemicals and materials of interest at yields and concentrations required to be economical. The lignin fraction left after conversion of the sugars could be converted into chemicals and materials, or simply burned as a boiler fuel to provide the energy to run the overall process.

A cost breakdown for production of chemicals and materials from biomass should include the factors shown in Fig. 6. From the total revenue generated from all products, the variable costs for raw materials, utilities, labor, and maintenance should be subtracted to generate the contribution margin. The administrative expenses could then be subtracted to arrive at a gross revenue. Depreciation is subtracted from the gross revenue to determine earnings before interest and taxes. Interest on the debt is then subtracted to determine earnings before taxes, and net income is calculated by subtracting taxes from earnings.

If the ratio of the cost of feedstock is simply compared to the revenue that could be generated from the sale of this product, a quick test of the economic feasibility of this process could be obtained. For a single product, the cost-to-price ratio, which is the unit cost of the feedstock divided by the unit value of the product, is calculated by simply dividing the cost-to-price ratio by the fermentation yield (weight of the product divided by

the weight of the feedstock). This ratio must be < 1 for a product to be considered viable (39). On the other hand, in the production of commodity chemicals, the ratio of the cost of the raw materials compared to the total revenue generated often falls in the range of 60–90%. Therefore, it might be expected that the cost of raw materials could represent a large fraction of the final product price for commodity chemicals. For commodity materials, it might be expected that the ratio would lie at the lower end of the range.

If more than one product is produced in a process, it is necessary to consider the revenues possible from each one of these products in this test. The relationship that results will be the cost of the feedstock divided by the summation of the revenues from each one of the products:

$$\begin{aligned} \text{Fraction of revenue for feedstock} &= (\text{cost of feedstock} / \text{value of products}) \\ &= (C_f / \sum \alpha_i x_i y_i v_i) \end{aligned}$$

where i = component i of the feedstock, α_i = fraction of the maximum theoretical yield of product derived from component i , x_i = fraction of component i in the feedstock, y_i = theoretical yield of product derived from component i , C_f = unit cost of the feedstock, and v_i = value of the product derived from component i .

These criteria can be used as a simple test to evaluate the economic potential of a range of products. Actual yield data can be gathered for organisms that produce products of interest, and the yields employed in these relationships to determine the ratio of the cost of the feedstock compared to the value of the product. Furthermore, the maximum possible yield can be used as a test of the ultimate potential of a product (40).

These relationships provide a simple way to guide the direction of research and identification of opportunities for commercialization. First, the lower the cost of the feedstock compared to the value of the product, the better the chance of economic success. On the other hand, for commodity chemicals and materials, a typical cost-of-feedstock to value-of-products ratio of about 0.6 to 0.9 would be expected. To minimize the cost-of-feedstock to value-of-products ratio, the product of yield times the value should be maximized. Therefore, we should look for products that will achieve high yields, have high values, or both. However, it must be remembered that commodities will typically sell for low prices. Another important outcome is that we should seek to use all of the substrate, so we can generate the maximum revenue possible from the feedstock. Finally, it is very desirable to have a low-cost feedstock, such as lignocellulosic biomass.

CONCLUSIONS

Currently, about one billion gallons per year of ethanol are derived from corn in the United States. However, substantial opportunity exists to produce ethanol from lignocellulosic biomass. Ethanol production

from lignocellulosic biomass has the potential to reduce urban air pollution, decrease the buildup of carbon dioxide in the atmosphere and the potential for global climate change, reduce the vulnerability of our country to disruptions in oil supply, decrease the balance of payments for petroleum imports, and provide new markets for agricultural products. In the last 10 yr, substantial progress has been made on the enzymatic conversion of lignocellulosic biomass into ethanol, and the price of ethanol has dropped by almost a factor of three during that time. Furthermore, opportunities have been identified to reduce the cost of ethanol from lignocellulosic biomass so that it can compete with gasoline.

Anaerobic digestion offers an opportunity to convert lignocellulosic biomass into methane gas. High-solids digestion of biomass to methane gas has resulted in substantial increases in gas generation rates per volume of fermenter, decreasing the price of methane production. However, further advances in the technology, such as increasing the level of enzymes for breakdown of cellulose into fermentable sugars are required to achieve maximum economic potential.

Microalgae are single-celled plants that can produce high levels of lipid oil hydrocarbons in environments unsuitable for most other purposes. Progress has been made in identifying a wide range of microalgae that can produce lipid oil at high yields while tolerating high-salinity water, large swings in temperature, and other harsh conditions. The enzyme acetyl Co-A carboxylase has been found to play a key role in lipid oil synthesis. Research is in progress to engineer algae genetically to achieve higher levels of lipid oil production and faster growth rates. Algae offer the opportunity to use carbon dioxide from power plants for growth, thereby reducing the emissions of carbon dioxide into the atmosphere that could contribute to global climate change. Substantial reductions have been achieved in the cost of algal oil production, and opportunity exists to achieve commercial success if technology improvements are realized or penalties on carbon dioxide release are imposed.

In addition to producing fuels, such as ethanol, methane, and diesel, biotechnology can be applied to produce chemicals and materials from biomass. The current corn wet milling and dry milling industries could produce a wider range of chemicals and materials. Furthermore, the cellulose and hemicellulose fractions of lignocellulosic biomass could also be converted into chemicals and materials. Simple criteria have been devised to allow rapid screening of potential products from biomass for their economic merit. We now need to identify products that have economic potential and improve the technology to a point where these technologies can be applied in a cost-effective way.

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