



BIOFUELS

Bringing Biological Solutions to Energy Challenges

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Alternative fuels from renewable biomass (plant material) have the potential to significantly reduce U.S. dependence on imported oil. Several scientific breakthroughs, however, are needed to make biofuels a cost-effective alternative to fossil fuels. Building on advances in DNA technologies resulting from the Human Genome Project, the U.S. Department of Energy's (DOE) Genomics:GTL systems biology program is creating a new generation of biological research. This research will accelerate the understanding and application of plant and microbial systems central to producing biofuels, cleaning up the environment, and managing the global carbon cycle.

REDUCING DEPENDENCE ON FOREIGN OIL

The triple energy-related challenges of the 21st Century are economic and energy growth, energy security, and climate protection. Energy helps drive the U.S. and global economies and has a significant impact on our quality of life and the health of our people. The United States is dependent on foreign oil, in particular, for the transportation sector. In 2005, net imports of liquid fuels, primarily petroleum, accounted for 60% of domestic consumption. Furthermore, recent measurable changes to the Earth's climate and improved science-based predictions of possible changes in the future

have signaled a call to action to reduce the environmental impacts associated with energy use via reductions to the nation's carbon emission footprint. Science-driven innovations in technology will be critical to addressing these challenges. Biofuels—particularly cellulosic ethanol—represent one such innovation.

In 2007, the President set a goal of reducing gasoline usage in the United States by 20% in the next 10 years. To achieve this goal, 15% of the reduction will come from expanding the supply of alternative fuels and the remaining 5% from increasing motor vehicle effi-

Genomics:GTL (formerly Genomes to Life) is a research program that aims to develop a predictive understanding of the biological systems relevant to biofuel production and other DOE mission areas. Genomics:GTL is supported by the Office of Biological and Environmental Research and the Office of Advanced Scientific Computing Research within the DOE Office of Science (genomicsgtl.energy.gov).

ciency. Displacing 15% of the projected gasoline usage by 2017 will require a rapid expansion of renewable fuel production. Annual alternative fuel supplies would need to increase from about 5 billion gallons of corn grain ethanol produced in 2006 to about 35 billion gallons of alternative fuels from a variety of cellulosic biomass materials, including grasses, wood chips, and agricultural wastes. The President's "20 in 10" goal is an ambitious milestone towards achieving DOE's target of replacing 30% of transportation fuels with biofuels by 2030. ♦

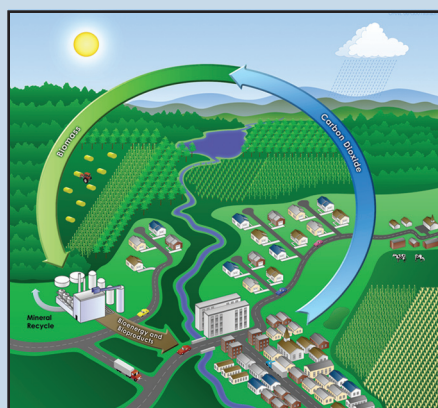


Fig. 1. New Generations of Bioenergy Crops Absorb CO₂ Emitted by Biofuel Use.

Benefits of Biofuels

- ▶ **Lower carbon dioxide and pollutant emissions.** The photosynthetic generation of new biomass takes up most of the CO₂ released from biofuel production and use. Biofuels also have lower sulfur, particulate, and toxic emissions than fossil fuels.
- ▶ **Abundant, renewable, and homegrown.** A joint DOE–U.S. Department of Agriculture study found that the United States could produce over 1 billion tons of biomass—enough to replace 30% of U.S. gasoline consumption—while still meeting demand for lumber, food, and fiber.
- ▶ **Revitalization of rural economies.** Conservative projections suggest that 10,000 to 20,000 jobs could be created for every billion gallons of biofuel produced.

SCIENTIFIC CHALLENGES TO BIOFUEL PRODUCTION

One of the most promising biofuels for near-term, commercial-scale deployment is ethanol from cellulosic biomass, the most abundant biological material on the planet. Examples of cellulosic biomass include wood chips, grasses, cornstalks, and other fibrous and inedible portions of plants.

Despite its abundance, cellulosic biomass is a complex feedstock that requires more extensive processing than corn grain—the primary feedstock for conventional fuel ethanol production. Figure 2 provides an overview of key cellulosic ethanol processing steps. Although cellulosic ethanol production has been demonstrated at a pilot scale, several scientific

breakthroughs are needed to make this process cost-competitive with gasoline and productive enough to displace up to 30% of U.S. gasoline consumption.

Three areas where focused biological research can bring down costs and increase productivity (as shown in Fig. 2, below) are

- developing energy crops dedicated to biofuel production (see step 1),
- improving enzymes that deconstruct cellulosic biomass (see steps 2 and 3), and
- optimizing microbes for industrial-scale conversion of biomass sugars into ethanol and other biofuels or bioproducts (see step 4).

Developing Energy Crops

Energy crops are biomass feedstocks grown specifically for energy production. The energy content of cellulosic biomass comes from the complex compounds that strengthen plant cell walls and support plant structure. A large portion of the plant cell wall contains long chains of sugars (polysaccharides) that can be converted to transportation fuels such as ethanol. Cellulose—the most abundant polysaccharide in plant cell walls—consists of tightly bound sugar chains embedded within a matrix of other polymers that can act as a protective coating to shield cellulose from enzymatic attack.

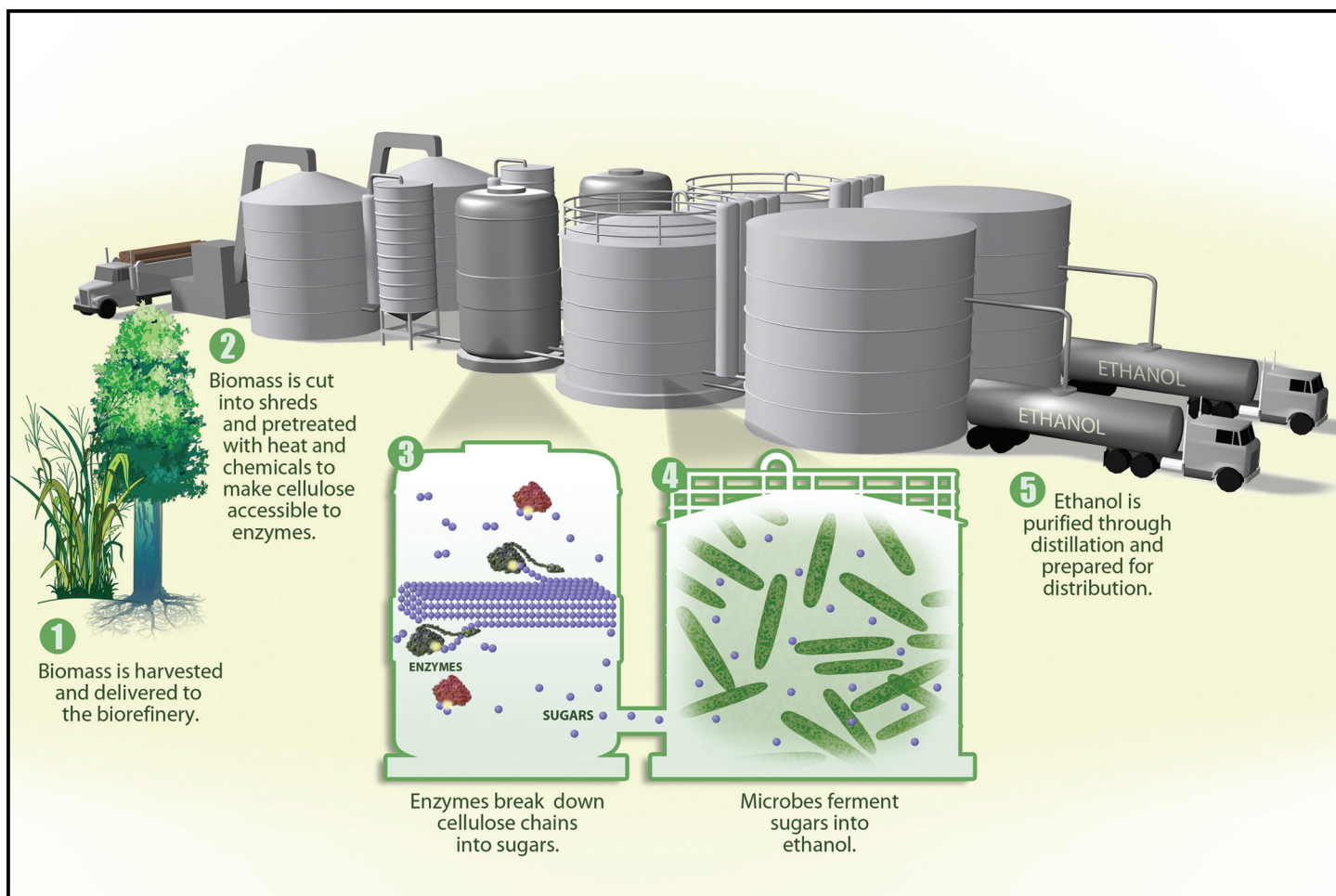


Fig. 2. *From Biomass to Cellulosic Ethanol.* This figure highlights some key processing steps in an artist's conception of a future large-scale cellulosic ethanol production facility.

The plant cell wall is so complex that several thousand genes are thought to be involved in cell-wall synthesis and maintenance. Only a few of these genes have been identified, and little is known about their functions. Understanding how genes control cell-wall composition in plants could lead to new energy crops with cell walls optimized for enzymatic deconstruction.

In addition to altering cell-wall composition, other energy crop improvements include maximizing biomass productivity per acre, increasing resistance to pests and drought, and minimizing the application of fertilizers and other inputs. Many potential energy crops are grasses or fast-growing trees that have not benefited from the years of agricultural research devoted to breeding better traditional crops such as corn or wheat. With new biological tools, the generation time of new energy crops could be reduced significantly.

Improving Enzymes that Deconstruct Biomass

The enzymes used to break down cellulose into sugars for biofuel production come from fungi and bacteria that are specialists at degrading biological materials in natural environments. In fact discovering, harnessing, and enhancing the best biomass-degrading enzymes in nature will have a significant impact on reducing the cost of cellulosic ethanol production. Scientists are just beginning to realize the vast diversity of enzymes in environments such as the termite gut and the cow rumen. Explorations are also under way to study other natural habitats likely to be sources of important enzymes.

Another challenge is to identify and produce the right mix of enzymes for industrial processing. Cellulose is so resistant to deconstruction that several enzymes with specialized functions must work in teams to break it down.

The most efficient team may consist of enzymes made by different types of microbes.

Optimizing Microbes that Convert Biomass Sugars into Ethanol

In addition to cellulose, other polysaccharides (collectively called hemicelluloses) in plant cell walls also are broken down into fermentable sugars when biomass is pretreated with heat and chemicals. Although cellulose is made of one type of six-carbon sugar called glucose that is readily converted into ethanol, microbial fermentation of the five- and six-carbon sugar mix from hemicelluloses is less efficient and represents a key area for improvement.

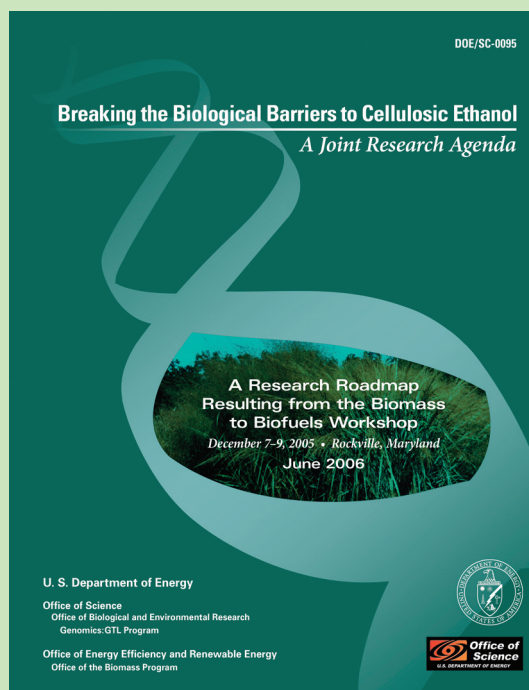
En route to the fermentation tank, biomass is subjected to physical, chemical,

and enzymatic processing steps that can create by-products and conditions that might inhibit microbial conversion of sugars into ethanol. Ethanol itself also inhibits microbial fermentation at high concentrations. Developing microbes robust enough to withstand the stresses of industrial processing and tolerate higher ethanol concentrations is an important near-term research target.

A more distant research target that could drastically reduce costs and simplify the entire production process is a method to combine cellulose deconstruction and sugar fermentation into a single step mediated by a "multitasked" single microbe or stable mixed culture of microbes. ♦

A New Research Roadmap for Biofuels

In June 2006, DOE released *Breaking the Biological Barriers to Cellulosic Ethanol: A Joint Research Agenda*. This research roadmap was jointly sponsored



by the Office of Biological and Environmental Research in the DOE Office of Science and the Office of the Biomass Program in DOE's Office of Energy Efficiency and Renewable Energy. The roadmap provides recommendations for using modern biology tools to overcome barriers to the rapid expansion of the cellulosic ethanol industry. It is the product of a workshop attended by more than 50 leading biomass and biofuel researchers and systems

biologists from academia, industry, and the government. Although ethanol was the focus, the scientific challenges addressed at the workshop also apply to other biofuels and bioproducts (genomicsgtl.energy.gov/biofuels/b2bworkshop.shtml).

THE TERMITE GUT

Nature's Microbial Bioreactor for Digesting Wood and Making Biofuels

The microbial community within a termite's gut is one of nature's most efficient bioreactors—typically converting 95% of cellulose into simple sugars within 24 hours. More than 200 species of microbes make up this community, and together they produce a bounty of wood-busting enzymes that industry could put to work in biorefineries making ethanol from several forms of cellulosic biomass.

This diverse array of microbial capabilities that could jumpstart a new biofuel industry is the result of a codependent strategy for survival. Without wood-eating microbes, a termite would not be able to extract nutrients and energy from wood, and, without the termite to grind wood into tiny pieces and

provide an oxygen-free habitat within its gut, the microbes would not be able to survive.

In addition to efficiently degrading cellulose into sugars, some termite-gut microbes are biochemically capable of generating other potential fuels such as hydrogen or methane. Hydrogen produced by one group of microbes is consumed by other gut microbes that create energy-producing by-products the termite can use. Investigations of the termite-gut community reveal a vast collection of biological pathways that may one day be put to use for multiple energy applications.

A collaboration of researchers from the Department of Energy's Joint Genome Institute (DOE JGI), the California

Institute of Technology, Diversa, and the National Biodiversity Institute of Costa Rica has sequenced and analyzed microbial DNA extracted from the guts of hundreds of termites harvested from a nest in a Costa Rican rainforest. Preliminary results already have identified several novel enzymes capable of degrading cellulose into sugars, and the San Diego-based biotechnology company Diversa has used insights from this discovery to create a high-performance enzyme cocktail for processing plant biomass into biofuels.

DOE JGI researchers continue to investigate other microbial communities in the guts of insects that consume different plant materials. The goal is to understand and reconstruct a diverse range of metabolic processes that could be scaled up for industrial biofuel production. ♦

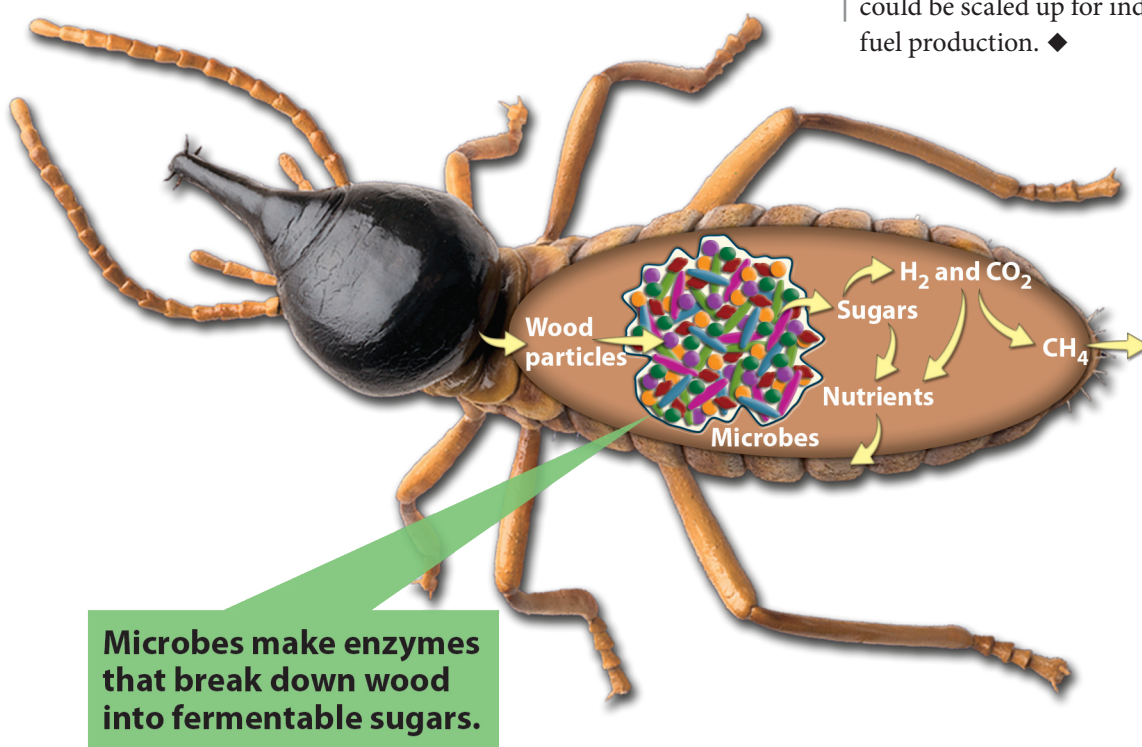


Fig. 3. Termite Microbes: A Potential Source of Enzymes to Digest Biomass for Bioenergy Production.