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Industrial Biotechnology Is Revolutionizing the Production of Ethanol Transportation Fuel

Biotechnology Is Playing a Key Role in the Rapid Expansion of the Ethanol Industry and in Development of Cellulosic Ethanol

In his Jan. 23, 2007 State of the Union address, President Bush proposed initiatives to catapult the U.S. biofuels industry into the next level of commercial development, to address climate change from transportation sources and to increase energy security:

> "It's in our vital interest to diversify America's energy supply – the way forward is through technology…. We must continue investing in new methods of producing ethanol using everything from wood chips to grasses, to agricultural wastes…. Tonight, I ask Congress to join me in pursuing a great goal. Let us build on the work we've done and reduce gasoline usage in the United States by 20 percent in the next 10 years.… To reach this goal, we must increase the supply of alternative fuels, by setting a mandatory fuels standard to require 35 billion gallons of renewable and alternative fuels in 2017 – and that is nearly five times the current target."

These proposed initiatives built on and accelerated the timeline for achieving the Advanced Energy Initiative announced by President Bush in his Jan. 31, 2006, State of the Union address. Stating "America is addicted to oil," the President that year proposed a 22 percent increase in clean-energy research at the Department of Energy, including funding for research to produce cellulosic ethanol at a cost that is competitive with gasoline by 2012. According to the President:

> “We’ll also fund additional research in cutting-edge methods of producing ethanol, not just from corn, but from wood chips and stalks, or switchgrass. Our goal is to make this new kind of ethanol practical and competitive within six years.
>
> “Breakthroughs on this and other new technologies will help us reach another great goal: to replace more than 75 percent of our oil imports from the Middle East by 2025. By applying the talent and technology of America, this country can dramatically improve our environment, move beyond a petroleum-based economy, and make our dependence on Middle Eastern oil a thing of the past.”

Jim Greenwood, president and CEO of the Biotechnology Industry Organization (BIO), applauded the President for the initiatives outlined in his 2007 State of the Union address:

> “The President is sending a dramatically positive signal to the investment community, to farmers, to biotech companies and to gasoline refiners that our government will work with the private sector to make the biofuels sector a major contributor to our energy independence. Biotechnology is the key enabling technology that can help the United States significantly reduce its use of foreign petroleum.”

In 2006, Greenwood also praised the President’s Advanced Energy Initiative:

> “The Advanced Energy Initiative will increase America’s competitiveness by providing clean, affordable energy sources that will enable us to lessen our dependence on foreign oil through biotechnology. Using crop wastes, we can produce tens of billions of gallons of ethanol. We could produce 25 percent of our transportation fuel need by 2015 if we dramatically ramp up biorefinery development.”

In June 2006, the U.S. Department of Energy (DOE) published a research roadmap for the advanced technologies needed to produce ethanol from cellulose. The DOE set a goal of producing 60 billion gallons of ethanol for transportation fuel – meeting 30 percent of current demand – by 2030:

> "Recent advances in science and technological capabilities, especially those from the nascent discipline of systems biology, promise to accelerate and enhance this development. Resulting technologies will create a fundamentally new process and biorefinery paradigm that will enable an efficient and economic industry for converting plant biomass to liquid fuels."

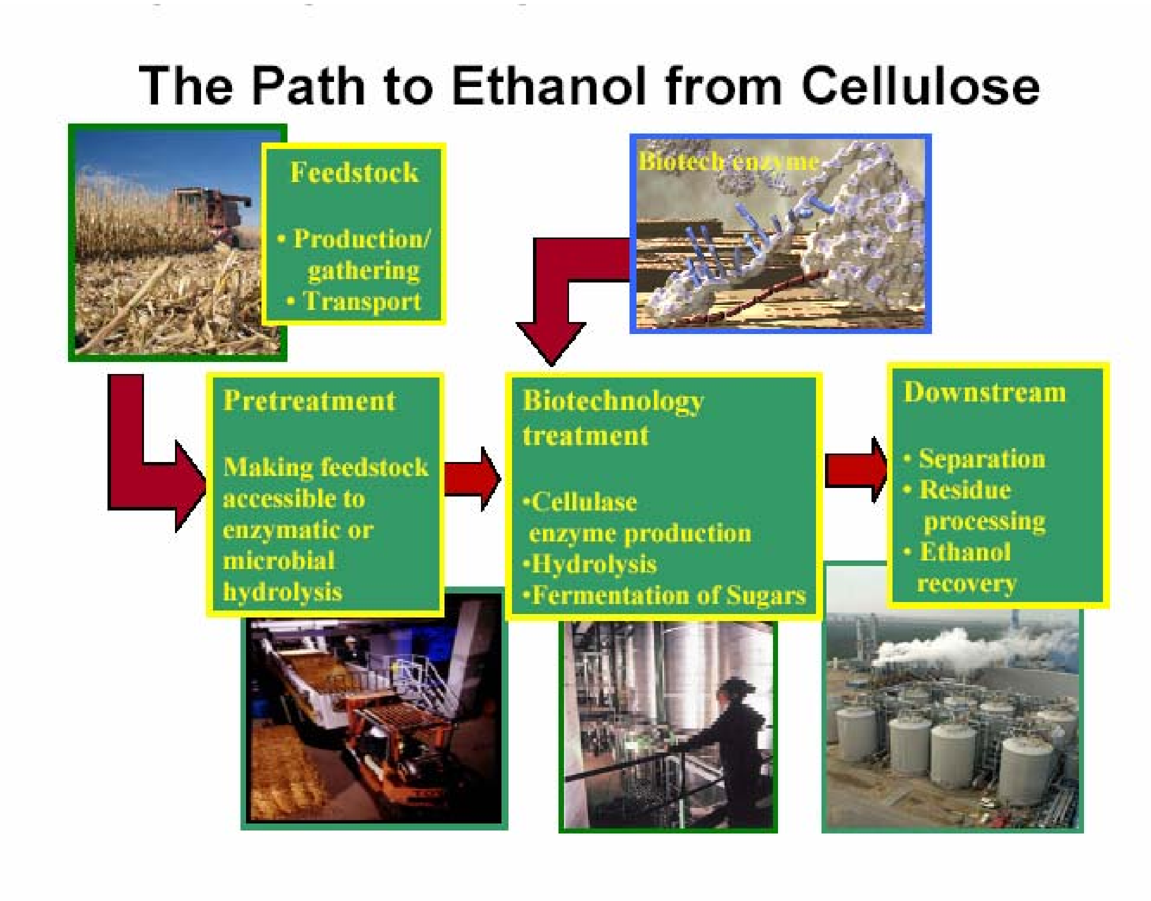
What Is Cellulosic Ethanol?

Currently, ethanol is primarily fermented from the sugar that makes up the starch in grain. Corn is the source of starch for more than 92 percent of ethanol production in the United States. The starch is converted to dextrose by amylase enzymes, and this sugar is then fermented with yeast, just as in making beer or distilled spirits. Overseas, countries such as Brazil, India and Malaysia use other dedicated crops as feedstocks, including sugarcane and palm oil.

But ethanol also can be made from cellulosic biomass – plant matter composed primarily of inedible cellulose fibers that form the stems and branches of most plants. Crop residues (such as corn stalks, wheat straw and rice straw), wood waste, and even municipal solid waste are potential sources of cellulosic biomass. Dedicated energy crops, such as switchgrass or fast-growing trees, are also promising cellulose sources because they can be sustainably produced in many regions of the United States.

Cellulosic biomass is a highly undervalued and underutilized energy asset in the United States and around the world. Cellulose-containing natural products are widely abundant: indeed, cellulose has been estimated to make up half of all the organic carbon on the planet. A 2005 analysis by the Natural Resources Defense Council found that ethanol from cellulose could supply half of U.S. transportation fuel needs by 2050, without decreasing production of food and animal feed.

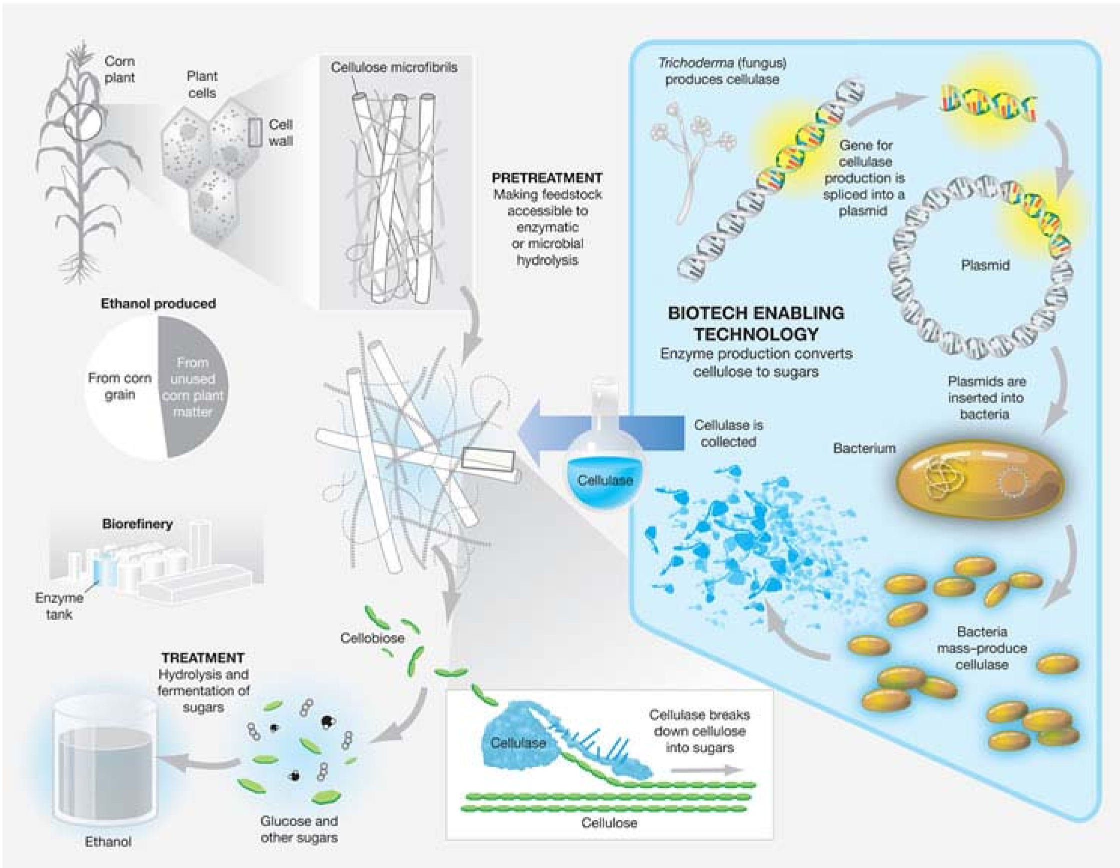
Cellulosic biomass has been a challenge for scientists to convert to ethanol, because the cellulose is tightly bound in a matrix with hemicellulose and lignin, which cannot be fermented. In the past, scientists have used harsh acids and high temperatures to try and break the cellulose matrix into its individual sugar components. However, an economical process has never been developed using traditional chemistry.



Recent advances in the relatively new field of industrial biotechnology are making it possible to convert cellulosic biomass to ethanol. Enzymes that break down cellulose – called "cellulases" – can economically convert plant matter to fermentable sugars. Cellulases are naturally produced by fungi as well as microbes, such as those from the guts of termites. Biotech tools are necessary to identify or produce the most efficient cellulases on a scale useful for ethanol production.

A pretreatment step using steam or dilute sulfuric acid is generally used to separate the biomass into its constituent parts – cellulose, hemicellulose, and lignin. The cellulose and hemicellulose are then hydrolyzed to sugars – both five-carbon sugars – xylose and arabinose – and six-carbon sugars – glucose, mannose and galactose. These sugars require specialized microbes or modified yeasts for fermentation. Biotechnology is used to identify or genetically enhance microorganisms to ferment these sugars.

How Biotechnology Is Used in Producing Ethanol from Cellulose



The Energy Information Administration (EIA) predicts in its Annual Energy Outlook 2007 that ethanol consumption will reach 11.2 billion gallons by 2012, outstripping the 7.5 billion gallons required in the Renewable Fuel Standard that was enacted as part of the Energy Policy Act of 2005. The Renewable Fuels Association counts 113 U.S. ethanol distilleries in operation and another 78 under construction, with capacity to produce 11.8 billion gallons within the next few years. Until 2030, the EIA predicts that demand for ethanol will grow 5.2 percent annually, reaching 14.6 billion gallons in 2030 (about 8 percent of total gasoline consumption by volume).

BIO supports the production of ethanol from all feedstocks. Agricultural biotechnology is helping to increase supplies of corn and other agricultural feedstocks for ethanol, while industrial biotechnology is helping to convert corn starch and crop residues into ethanol more efficiently. With ongoing advances in biotechnology, biofuels will help America meet nearly half its transportation-fuel needs by the middle of this century.

Modern biorefineries to produce cellulosic ethanol from a wide variety of biomass resources are currently being constructed throughout the United States, Canada and Europe. Included in the list below are existing, planned, and under construction facilities to produce cellulosic biofuels.

Company	**Plant Location**	**Feedstock**	**Technology**	**Capacity** (gallons/year)
Abengoa	Colwich, Kan.	corn stover, wheat straw, milo (sorghum) stubble, switchgrass, and others	thermochemical and biochemical processing	11.4 million
Abengoa	York, Neb.	corn stover, residual starch	enzymatic hydrolysis (*Chrysosporium lucknowense*), integrated with dry mill corn production	0.47 million (0.02 million from corn stover)
Abengoa	Salamanca, Spain	wheat straw, cereal	steam pretreatment, enzymatic hydrolysis	1.3 million
ALICO, Inc.	LaBelle, Fla.	yard and citrus wastes	gasification, fermentation of syngas	13.9 million
BioEthanol Japan	Osaka, Japan	wood construction waste	enzymatic hydrolysis, fermentation (*Klebsiella oxytoca* and *E. coli*)	0.37 million

BioFuels Energy Corp.	Raymondville, Texas	grass and tree trimmings		4 million
BlueFire Ethanol	Lancaster, Calif.	green waste	acid hydrolysis, fermentation	3.1 million
Colusa Biomass	Colusa, Calif.	rice straw and hulls	acid hydrolysis, fermentation	20 million
China Resources Alcohol Corporation	ZhaoDong City, Heilongjiang Province	corn stover	steam pretreatment, enzymatic hydrolysis	1.7 million
DuPont-BP Biofuels	Wissington, England	sugar beets	enzymatic hydrolysis, fermentation to biobutanol	9 million
Iogen	Shelley, Idaho	wheat straw, barley straw, corn stover, switchgrass and rice straw	enzymatic hydrolysis (*Trichoderma reesei, Saccharomyces*)	18 million
Iogen	Ottawa, Canada	wheat, oat and barley straw	enzymatic hydrolysis (*Trichoderma reesei, Saccharomyces*)	0.79 million
Lignol	Vancouver, Canada	softwood and hardwood	pulping liquor pretreatment, enzymatic hydrolysis	1.3 million
Mascoma	Rochester, N.Y.	paper sludge, wood chips, switch grass and corn stover.	enzymatic hydrolysis and fermentation (*Thermoanaerobacterium saccharolyticum*)	0.5 million

Poet	Emmetsburg, Iowa	corn fiber, corn stover	enzymatic hydrolysis, integrated with dry mill	30 million
Range Fuels	Soperton, Ga.	timber and forest residue	pyrolysis and catalytic conversion	40 million
Verenium	Jennings, La.	sugarcane bagasse and specially bred energy cane	enzymatic hydrolysis, fermentation (*Klebsiella oxytoca* and *E. coli*)	1.4 million
Western Biomass	Upton, Wyo.	Ponderosa pine wood chips, waste	CO2 pretreatment, enzymatic hydrolysis	1 million

What Is Industrial Biotechnology and How Is It Enabling Ethanol Production?

Industrial biotechnology companies develop biocatalysts, such as enzymes, to be used in industrial manufacturing involving chemical synthesis. Enzymes are proteins that are present in all living organisms. Enzymes trigger or speed up chemical processes that would otherwise run very slowly, but they are not consumed in the process of changing other molecules. They are unique due to their catalytic activity and their ability to break down compounds such as cellulose. As catalysts, they can also put broken protein fragments back together.

Scientists have learned how to convert these biological systems and enzymes into very useful industrial tools that replace less efficient and sometimes toxic chemical processes. Natural enzyme-based processes operate at lower temperatures and produce less toxic waste and fewer emissions than conventional chemical processes. They may also use less purified raw materials because they have precise chemical selectivity.

Industrial biotech companies conduct genomic studies to capitalize on the wealth of genetic diversity in microbial populations. Researchers then use DNA probes to look for genes that express enzymes with specific biocatalytic capabilities. Once snared, enzymes can be identified and characterized for their ability to function in specific industrial processes and, if necessary, they can be improved with advanced biotechnology techniques. This is exactly what scientists have been doing to improve enzymes for cellulosic ethanol production.

Once the needed enzyme is discovered and improved, it may be produced in commercial quantities using systems similar to those that produce human therapeutic proteins or bulk yeast for the brewing industry—bioreactors, cell cultures or fermentation.

Scientists are using genomics, proteomics and bioinformatics to improve the productivity of enzymes an to increase enzyme output rates. Biotechnology enables scientists to custom tailor the specificity of enzymes, improve catalytic properties or broaden the conditions under which enzymes can function so that they are more compatible with existing industrial processes.

Using enzymes in industrial processes has several benefits. Use of enzymes can reduce the amount of harsh chemicals in industrial processes that contaminate the environment. In addition to creating cleaner process outputs, enzymes help conserve energy and raw materials by reducing the amount of inputs in industrial processes. These enzymes are the key to ensuring that older industrial manufacturing processes evolve into cleaner, more sustainable industrial ones.

Biotech companies are producing improved enzymes for converting starch to ethanol and have been perfecting enzymes for converting cellulose to ethanol. Ethanol from cellulose can substitute for petroleum in many manufacturing processes – such as plastics – and could contribute in a major way to reducing America's "addiction to oil," while at the same time helping to address climate change, by reducing our need to burn fossil fuels.

Biotech Breakthroughs and Promising Developments in the Production of Cellulosic Ethanol

Dramatic Cost Reductions

In 2001, the cost of cellulase enzymes to convert cellulose to sugars was the greatest technical barrier to cost-effective production of ethanol from cellulose. Enzyme producers **Genencor International** and **Novozymes A/S** partnered with the U.S. Department of Energy to increase enzyme activity and reduce the cost of production. Both of these industrial biotechnology companies have achieved dramatic results.

Novozymes identified a range of new enzymes and enhanced their activity to dramatically boost sugar yields, reducing the cost of enzymes for making ethanol from corn stover 30-fold from $5 per gallon in 2001 to a mere 10¢ to 18¢ in 2005. Genencor also achieved a 30-fold reduction in cost by developing a team of genetically enhanced enzymes that act in synergy to convert cellulose to sugar. DOE has determined that enzymatic processing is no longer an obstacle to commercialization of ethanol from cellulose.

Other Enzyme Breakthroughs

Diversa Corporation, which recently merged with **Celunol Corp.** to become **Verenium,** partnered with a consortium that includes **DuPont**, John Deere & Co., the National Renewable Energy Laboratory and Michigan State University to develop a biorefinery that can produce ethanol and other products from the entire corn plant, integrating traditional grain-based ethanol production with ethanol production from stalks and husks.

In 2005, Diversa successfully developed a suite of enzymes that has enabled the consortium to begin developing a demonstration facility for this Integrated Corn-Based Biorefinery concept. DuPont and **Tate & Lyle** also utilize the enzymes to produce a biobased plastic, bio-PDO (1,3 propanediol), at a facility in Loudon, Tenn., that officially opened in June 2007.

Dyadic International has developed an integrated technology platform from a fungus (C1) that will enable researchers to identify, select and analyze novel enzymes best suited to convert biomass materials into biofuels. Dyadic has also developed enzymes for the textiles, pulp and paper, food and feed, and chemicals industries that greatly reduce waste while offering new and enhanced products to a variety of markets.

The First Commercial Production of Cellulosic Ethanol

In 2004, **Iogen Corporation** of Ottawa, Canada, became the first company to begin commercial production of cellulosic ethanol. Using biotech enzymes and genetically enhanced yeasts -- licensed from Dr. Nancy Ho of Purdue University -- Iogen built the world's first demonstration-scale cellulosic ethanol facility, converting wheat straw to clean-burning cellulose ethanol, with an annual capacity of approximately 660,000 gallons/year. Iogen's cellulosic ethanol is used by the company's fleet of flexible fuel vehicles as well as by some departmental fleets within the Government of Canada. Iogen was recently selected as a U.S. Department of Energy grant recipient of up to $80M to construct a commercial-scale cellulosic biorefinery in Idaho.

In August 2005, **Abengoa Bioenergy** began construction in Salamanca, Spain of the world's first commercial-scale ethanol from cellulose plant. Upon completion in 2007, the plant will process 70 tons of agricultural residues, such as wheat straw, each day, producing nearly 1.3 million gallons (5 million liters) of ethanol annually. Advances in cellulose enzymatic hydrolysis and fermentation were key to enabling Abengoa to take their technology to market. The plant utilizes the steam explosion pretreatment technology developed by **SunOpta**.

Abengoa has also received a $10 million DOE grant to develop a next-generation dry mill corn ethanol plant. This next-generation plant, scheduled for completion in 2007, will produce ethanol from the entire corn kernel – both starch (the only portion currently utilized for ethanol) and the residual fiber (also known as dry distillers grains, or DDGs), which would require processing with cellulase enzymes. The application of cellulosic technology could dramatically increase the ethanol yield of the nation's over 100 existing ethanol facilities. Abengoa is converting an existing ethanol facility in York, Neb. adding 500,000 gallons of ethanol from cellulose to the plant's annual production of 50 million gallons of ethanol from starch.

DuPont, the largest chemicals producer in the United States, worked with John Deere and Verenium (formerly Diversa) to develop an integrated corn-based biorefinery that would produce fuels and chemicals from the entire corn plant. DuPont has also partnered with **BP** to develop bio-butanol, a more energy-rich fuel alcohol, first from starch, and eventually from cellulose.

POET (formerly Broin), the largest dry-mill ethanol producer, is jointly funding with the DOE the conversion of an existing 50 million gallon per year grain ethanol plant in Emmetsburg, Iowa into a commercial cellulosic biorefinery. The facility will utilize POET's proprietary fractionation process and lignocellulosic conversion technologies to produce ethanol from corn fiber and cobs in addition to grain. Once complete, the facility will produce 125 million gallons per year, 25 percent of which will be from cellulosic feedstock. By adding cellulosic production to an existing grain ethanol plant, POET will be able to produce 11 percent more ethanol from a bushel of corn, 27 percent more from an acre of corn, while almost completely eliminating fossil fuel consumption and decreasing water usage by 24 percent.

Mascoma recently received a $14.8 million award from the New York State Department of Agriculture and Markets and the New York State Energy Research and Development Authority to build and operate a biomass-to-ethanol demonstration plant in Rochester, New York. Mascoma is partnering with Genencor and a consortium that includes International Paper Co., Cornell University, Clarkson University and the Natural Resources Defense Council to turn agricultural and/or forest products – including paper sludge, wood chips, switch grass and corn stover – into half a million gallons of ethanol.

Verenium (formerly Celunol/Diversa) broke ground in 2007 on a new demonstration-scale cellulosic ethanol production facility, located at its Jennings, La. site, with a design capacity of 1.4 million gallons per year. The facility will convert locally grown sugarcane bagasse and specially bred energy cane to ethanol. The company's technology has also been incorporated into Bioethanol Japan's 1.4 million liters-per-year cellulosic ethanol plant in Osaka, Japan – the world's first commercial-scale plant to produce cellulosic ethanol from wood construction waste.

Bioethanol Experts

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- Gerson Santos Leon, Corporate Director, Research & Development, Abengoa Bioenergy – (636) 728-0508 (biorefinery production)

- Lee Lynd, associate professor, Thayer School of Engineering, Dartmouth University – (603) 364-2231
- Glenn Nedwin, CSO & EVP, Dyadic -- (530) 792-8961 (enzymes)
- Dr. Mark Stowers, VP R&D, POET (605) 965-6438

Enzyme, Bioethanol and Biobased Products Companies

Abengoa Bioenergy Corporation – www.abengoa.com
BP – www.bp.com
Cargill – http://www.cargill.com/
Codexis, Inc – www.codexis.com
DuPont – http://www.dupont.com/
Dyadic International, Inc – www.dyadic-group.com
Genencor International – www.genencor.com
Iogen Corporation – www.iogen.ca
Lignol – http://www.lignol.ca/
Mascoma Corporation – www.mascoma.com
NatureWorks, LLC – www.natureworksllc.com
Novozymes – http://www.novozymes.com/en
Poet – www.poetenergy.com
SunOpta – http://www.sunopta.com/
Tate & Lyle – www.tate-lyle.co.uk
Verenium – http://www.verenium.com/

What Policies Will Help Ethanol Meet the Challenge of the Advanced Energy Initiative?

Rapid commercialization of ethanol from cellulose will require government support in four key areas: advanced research and development, biorefinery construction, infrastructure to supply feedstocks to biorefineries, and market creation/expansion.

Biorefinery Construction

The single greatest hurdle to commercialization of ethanol from cellulose is construction of the first integrated biorefineries. Potential refiners are unable to secure private financing for construction of the first plants without some government assurance/participation.

The Energy Policy Act of 2005 established a suite of programs to provide the necessary government assurance. These programs must be funded at a level sufficient to mobilize private financing:

- **Biorefinery Loan Guarantee Program:** For the construction of up to four demonstration cellulosic ethanol facilities (Sec. 1511).
- **Biorefinery Grants Program:** Authorizes $750 million in grants over three years for the commercial production of ethanol from cellulose (Sec. 1512).

In February 2007, Energy Secretary Samuel W. Bodman announced the awards of cost-sharing grants – totaling up to $385 million over three years – to help build six new biorefineries in the United States. Companies receiving the awards included Abengoa, Iogen and Poet (Broin).

R&D

The cost of ethanol production from cellulose has declined dramatically over the past five years with breakthroughs in industrial biotechnology. However, production at prices competitive with the mature petroleum industry still depends on advances in new crop varieties, sustainable harvesting, storage, transportation and processing. This will require an intensive, focused R&D and demonstration program.

The Energy Policy Act of 2005 provided for strong programs at both the Department of Energy and USDA. These R&D and demonstration programs should be funded at least at the level authorized in the Act:

- **DOE Bioenergy Program** – $738 million authorized over three years for R&D and integrated biorefinery demonstrations. The program received $91 million in FY06 appropriations. The President has proposed increasing funding to $150 million for FY07. This is still just over half the authorized level.
- **Joint USDA/DOE Biomass R&D Program** – $2 Billion authorized over 10 years for enhanced USDA/DOE Biomass Research Program. The program is funded at just $14 million in FY06. The President has given no indication that this will be increased in FY07.
- **DOE Office of Science** – $196 million over three years for programs, projects and activities through DOE Office of Science for additional integrated bioenergy research and development. This program has not yet been funded.

Infrastructure
Sustainable production and collection of cellulosic agricultural feedstocks will require considerable infrastructure construction and upgrading. Congress should consider implementing the following policy measures in the 2007 Farm Bill:

- Fund research and development and provide incentives for the development of one-pass harvesting equipment and other new harvesting equipment for collection of cellulosic agricultural feedstocks;
- Develop and make available simple-to-use soil carbon computer models to allow individual farmers to compute how much crop residue can be collected without degrading soil quality;
- Provide support to farmers to assist with the transition to no-till cropping for biomass production;
- Provide incentives for the development and expansion of short line and regional rail networks for transport of cellulosic feedstocks;
- Fund regional demonstration projects to streamline the collection, transport and storage of cellulosic feedstocks;
- Develop a system to monetize greenhouse gas credits generated by production of ethanol and other products from agricultural feedstocks; and
- Fund programs to help farmers identify and grow the most suitable crops for both food production and cellulosic biomass production.

The Bush Administration's proposals for the 2007 Farm Bill, released in February 2007, include $1.6 billion in funding for research in alternative energy, specifically targeting ethanol from cellulose and biobased products. Significantly, the proposals include funding for loan guarantees for ethanol from cellulose facilities; for research in renewable energy, biobased products, and sustainable production of biomass; and for incentives to increase market demand for biobased products.

Market Creation/Expansion
Even with ethanol production from cellulose underway, federal policy is needed to ensure penetration into the petroleum-dominated market for transportation fuels.

The Energy Policy Act of 2005 established several programs to create and expand the market for ethanol. These programs should be fully implemented:

- **Renewable Fuels Standard** – Beginning in 2013, a minimum of 250 million gallons a year of cellulosic ethanol must be blended into the nation's fuel supply.
- **Federal Offtake Agreement** – Federal government will award cellulosic ethanol producers a per gallon incentive until U.S. production reaches 1 billion gallons per year. Awards are offered through production auctions, with awards given to the lowest bidders, thereby reducing the per gallon subsidy over time. Program is authorized at $250 million.
- **Federal Procurement** – Requires the use of renewable fuels in all government fleet vehicles capable of their use.
- **Flex Fuel Pumps** – Fueling stations will receive a tax credit for 30 percent of the cost of installing pumps dispensing ethanol blends of at least 85 percent, up to $30,000 per station through 2010.

*Glossary**

Terms used frequently in this and other discussions of bioenergy are defined below. The definitions given here are in some cases specific to the context of energy supply from biomass and may not be complete or fully accurate in other contexts.

Bioenergy – Useful energy (fuel, electricity, heat) produced from biomass.

Biomass – Plant matter of recent (non-geologic) origin or materials derived therefrom.

Biorefinery – A processing facility in which multiple products are produced from biomass feedstocks.

Cellulosic biomass – Biomass composed primarily of inedible plant fibers having cellulose as a prominent component. These fibers may be hydrolyzed to yield a variety of sugars that can subsequently be fermented by microorganisms. Examples of cellulosic biomass include grass, wood, and cellulose-rich residues resulting from agriculture or the forest products industry.

Co-utilized crops – Crops that are used for both energy and non-energy uses, each of which are of comparable importance.

Dedicated energy crops – Crops grown for the primary purpose of energy production. Examples of such dedicated crops include corn used for production of ethanol in a dry mill, as well as switchgrass and short rotation trees used for production of fuels and/or power.

Feedstock – The source of carbon for production of organic fuels and chemicals via industrial processes.

Lignin – A component of cellulosic biomass that is rich in energy but not fermentable.

Residues – Biomass feedstocks available as a result of activities or processes undertaken for some purpose other than energy production. Examples of such residues include corn stalks and other non-edible parts of plants used to produce food, waste sludge produced at paper mills, and animal manure.

Sustainability – Having the capacity to be utilized for an indefinite period of time without degradation. Thus a sustainable resource can be used by present generations without compromising opportunities for use by future generations. Similarly, a sustainable activity can be undertaken by present generations without compromising opportunities for future generations to undertake that activity.

* From Dr. Lee Lynd, Dartmouth University.