

## Bioenergy and Energy Alternatives

### National Program 307 Annual Report

Fiscal Year 2003

#### Introduction

The mission of this national program is to create jobs and economic activity in America, reduce the Nation's dependence on foreign oil, and improve the environment by developing alternate energy sources and increasing the use of agricultural crops as feedstocks for biofuels. During the past year research to achieve this mission addressed several major problems and issues including:

- Cost of ethanol and biodiesel production
- Conversion of cellulosic biomass to ethanol
- Environmentally sustainable processing
- Fundamentals of stress tolerance of microbes for ethanol production
- Fermentation inhibitors that form during hydrolysis of cellulosic biomass
- Enzymes and organisms for converting biomass to fuels and products
- Fuel properties and combustion characteristics of biodiesel
- Genomic technologies for energy crop production and quality
- Germplasm and production strategies for herbaceous energy crops
- Harvesting and storage of cellulosic crop biomass
- Energy for remote rural areas

#### Specific Accomplishments for Fiscal Year 2003

##### Component I: Ethanol

Phytosterol coproducts may improve barley-to-ethanol economics. Hull-less barley can be grown in parts of the Mid-Atlantic region generally unsuitable for growing corn. Only about 60 percent of the barley kernel can be fermented to produce fuel ethanol. ARS scientists in the Crop Conversion Science and Engineering Unit at the Eastern Regional Research Center in Wyndmoor, Pennsylvania, found that oil extracted from nonfermentable portions of the barley kernel is high in phytosterols, which have positive dietary attributes. Profits from this co-product could improve the economics of converting barley to fuel ethanol, and could help to establish hull-less barley as a profitable grain crop in the Mid-Atlantic region.

New process lowers cost of valuable coproduct of corn ethanol. Zein is a unique corn protein that has value because it has good water vapor barrier properties, which makes it ideal for applications such as adhesives and moisture and grease-resistant coatings for pharmaceuticals and for packaging materials. Currently, zein is too expensive for many applications, partially because the centrifugation step used in its production is expensive. ARS scientists in the Crop Conversion Science and Engineering Unit at the Eastern Regional Research Center in Wyndmoor, Pennsylvania, have developed a technology for removing the solids from a zein isolate by passing the ethanol extract over a layer of water and allowing the solids to settle. This technology could significantly lower the cost of zein and allow its more abundant use in biobased adhesives, coatings, and films.

Improved model developed for simulating ethanol production processes and estimating costs. Scientists and engineers working to develop more efficient ways to convert grain to ethanol need simulation tools so they can evaluate new process ideas inexpensively on a computer rather than needing to build an expensive plant. Process and cost engineers in the Crop Conversion Science and Engineering Unit at the Eastern Regional Research Center in Wyndmoor, Pennsylvania, updated and validated, in cooperation with MBI International and Delta T Corporation, a computer model they had previously developed. The previous model was for a 15 million gallon per year dry grind ethanol plant and was the first publicly available process and cost simulation computer model made available to researchers worldwide. The newly updated model, with its enhanced capabilities, will be publicly distributed for use in research and industry and will be used to develop the simulation model for the National Corn-To-Ethanol Research Center at Southern Illinois University at Edwardsville.

Enzymes for biomass ethanol production isolated from an anaerobic fungus. Converting cellulosic biomass to ethanol requires first breaking down the biomass into its constitutive sugars. This is most efficiently done enzymatically; however, commercially available enzymes are expensive, have low activity, meaning too much enzyme is required, and often produce low sugar yields. ARS scientists and engineers in the Fermentation Biotechnology Research Unit at the National Center for Agricultural Utilization Research, Peoria, Illinois, have isolated hydrolyzing enzymes with very high specific activities from a novel source, the anaerobic fungus *Orpinomyces* PC-2. They have also applied recombinant DNA technology to construct hosts capable of producing these enzymes in large quantities. These advances will contribute to development of cost-effective enzymes for sugar production from biomass. This in turn will allow ethanol producers to use feedstocks other than starch containing grains to greatly increase production above current levels.

Yeasts detoxify fermentation inhibitors and improve biomass ethanol production. As interest in alternative energy sources increases, lignocellulosic materials have become attractive resources for biofuel production. However, during acid hydrolysis of lignocellulose numerous compounds are generated that inhibit subsequent fermentation of the sugars produced. Furfural and 5-hydroxymethylfurfural (HMF) are among the most potent inhibitors to fermentation. ARS scientists and engineers in the Fermentation Biotechnology Research Unit at the National Center for Agricultural Utilization

Research, Peoria, Illinois, found that *Saccharomyces cerevisiae* had the highest level of tolerance of the ethanologenic yeasts evaluated. The study provided the first direct evidence that *S. cerevisiae* converts HMF to the much less toxic HMF alcohol. The research team used directed adaptation methods to improve the tolerance level of one strain of *Pichia stipitis* and two strains of *S. cerevisiae* to at least six times that of their parent strains. Findings suggest that HMF tolerant strains have potential to be used to detoxify fermentation inhibitors and improve bioethanol production rates, yield, and economics.

Enzymes from mushrooms may improve ethanol economics. Agriculturally derived fuel ethanol will only displace large quantities of petroleum if abundant biomass sources, such as fibers, grasses, straws, and grain middlings are converted to ethanol via cost-effective isolation of fermentable sugars; which is presently costly. Enzymes with specific high activity and extended stability are crucial to reducing processing costs. ARS scientists in the Bioproduct Chemistry and Engineering Research Unit at the Western Regional Research Center, Albany, California, have identified and evolved various plant-degrading enzymes appropriate for this task. Xylanases and cellulases which were previously unknown were isolated and characterized from cultures of Shiitake mushroom mycelia, and expressed at higher levels by use of directed molecular evolution in yeast. These developments will lead to improved industrial plant cell-wall degrading enzymes, which are critical in reducing the chemical and energy consumption needed for economical cellulosic-biomass to ethanol manufacture.

Enzymatic milling may allow recovery of valuable ethanol coproducts. Dry grind corn processing is used exclusively for ethanol production and as currently practiced is limited to the production of only two low value coproducts, distillers dried grains with solubles and carbon dioxide. ARS scientists in the Crop Conversion Science and Engineering Research Unit at the Eastern Regional Research Center in Wyndmoor, Pennsylvania, are adapting the concept of enzymatic milling to the dry grind ethanol process for the purpose of recovering additional high value coproducts. Bench scale procedures for fractionation have been developed and a patent application related to this technology has been submitted to the United States Patent and Trademark Office. Major enzyme companies have provided large numbers of enzyme samples for testing the process. Enzymatic milling should improve the efficiency of dry-grind ethanol production and by increasing the value of coproducts should improve its economics.

## **Component II: Biodiesel**

Biodiesel process improves economics and eliminates organic solvents. The high cost of the refined vegetable oil, the most common feedstock for biodiesel, prevents this biofuel from competing economically with petroleum-derived diesel fuel. ARS scientists of the Fats, Oils and Animal Coproducts Research Unit at the Eastern Regional Research Center in Wyndmoor, Pennsylvania, developed and optimized a process to produce biodiesel directly in oilseeds. In cooperation with Ohio State University researchers, the scientists tested the suitability of the spent meal resulting from the process as an animal feed, which is crucial to the economic viability of the process. Young chickens accepted the oil-

depleted meal in their diets, gained weight normally, and showed no signs of toxicity. Findings indicate that this process could reduce the cost of producing biodiesel while eliminating the need for EPA-regulated organic solvents that current biodiesel production requires.

#### Model developed to estimate cost and increase efficiency of biodiesel production.

Individuals interested in constructing a biodiesel production facility, contemplating process modifications to such a plant, and developing methods to produce biodiesel from new feedstocks need tools to calculate the cost of the biodiesel product and to estimate the impact of process changes on that cost. ARS scientists of the Fats, Oils and Animal Coproducts Research Unit at the Eastern Regional Research Center in Wyndmoor, Pennsylvania, in collaboration with the Center's cost engineer, developed quantitative computer models to estimate the cost of biodiesel production from refined vegetable oil, from vegetable oil soapstock, and from whole soybeans. The first model, for alkali-catalyzed transesterification of refined vegetable oil, has been completed. The model, which has been made available to the general public, assists in estimating the effects of changes in feedstock and in process technology on the cost of producing biodiesel, should eliminate trial-and-error, and help increase the efficiency of biodiesel production.

New method to measure quality of biodiesel fuel. The fatty acid composition of biodiesel fuel is one of the major factors affecting its quality as a fuel. There is need for low-cost, rapid, easy, and reliable measurement of fuel quality at several points during the production and marketing of biodiesel. ARS scientists with the Food and Industrial Research Unit at the National Center for Agricultural Utilization Research, Peoria, Illinois, developed a spectroscopic method for determining the fatty acid composition of biodiesel fuel. Results with this new method were in good agreement with measurements made with the slower, more expensive chromatographic method normally used. The new method will not only help determine fatty acid composition but will also be useful in assessing the fuel quality of biodiesel that has been subjected to deterioration-promoting conditions.

### **Component III: Energy Alternatives for Rural Practices**

More reliable solar panel. Farmers and ranchers need a reliable, low-cost means of pumping water for cattle and other livestock in remote rangelands. ARS engineers with the Conservation and Production Research Laboratory, Bushland, Texas, found that amorphous-silicon solar panels showed no performance degradation with time when used to power a 1-kW solar-photo-voltaic water pumping system. The performance of a more expensive alternative, a polycrystalline solar panel, tends to degrade at 15 percent per year. These findings show that amorphous-silicon solar panels may be an effective means to provide electricity for remote rural needs.

### **Component IV: Energy Crops**

Corn stover removal reduces corn yields. Production and harvest of plant biomass for production of energy and products must be done in ways that are sustainable, including

ways that maintain soil productivity. A long-term experiment was established in 1998 in eastern Nebraska to quantify carbon sequestration of switchgrass grown and managed as a biomass energy crop in comparison to rain-fed, no-till corn. Because of U.S. Department of Energy interest in corn stover as a biomass energy source, corn plots were split in 2000 and half of the stover was mechanically harvested each year from one-half of each corn plot. ARS scientists at the Wheat, Sorghum and Forage Research Unit, Lincoln, Nebraska, found that corn grain yields were significantly reduced where corn stover was removed. In addition, potential ethanol yields from switchgrass biomass were found to be significantly greater than potential ethanol yields from corn, including that from both grain and stover. These findings show there is a need to more carefully determine the potential consequences of corn stover removal before the practice is recommended as a way to provide feedstock for biorefineries.

Phenotypic variability documented for switchgrass. Prairie remnant populations of switchgrass from plant growth hardiness zones 3, 4, and 5 have not been characterized or evaluated for agronomic potential, limiting our understanding of the potential for using switchgrass as a biofuel crop in these zones. ARS scientists with the U.S. Dairy Forage Research Center in Madison, Wisconsin, evaluated fifty-seven populations for several agronomic and biofuel traits. They found phenotypic variability among prairie remnant populations to be closely associated with both the USDA hardiness zone (defined by minimum cold temperatures) and ecoregion (defined by historic native vegetation) from which a population was collected. This is the first experimental and quantitative data to support the hypothesis that switchgrass populations have spatial structure at the landscape level, information that will be useful in identifying optimal germplasm for breeding, conservation, and production.

Stover harvest method improves efficiency and reduces storage losses. Energy crops and plant residues are being looked upon as feedstocks for fuel ethanol production and for production of other biobased products. However, there is need to develop lower cost and more effective ways of handling, storing, and delivering quality material to the biorefinery. The ARS U.S. Dairy Forage Research Center in Madison, Wisconsin, in a specific cooperative agreement with the University of Wisconsin-Madison showed that harvesting corn stover at higher moisture level (~40 percent moisture) and storing the stover in plastic film under non-ensiling conditions economized field operations, increased the rate and efficiency of harvesting, and reduced storage losses of dry matter to under 5 percent.

Guidelines to aid selection and establishment of warm season grasses. Though perennial-cropping systems could reduce degradation of agricultural soils, provide a source of renewable fuels to lower national dependence on foreign oil, and reduce emissions of greenhouse gases and toxic pollutants, management systems do not exist for establishing warm-season grasses on marginal lands and buffer strips and there is limited information on relative productivity of warm season cultivars. ARS scientists with the Natural Resource Management Research Unit in Mandan, North Dakota, found that optimal pH and temperature conditions for establishment of big bluestem and switchgrass were 20 C, pH =7 and 30 C, pH = 6, respectively. And, though very difficult to establish,

indiangrass was found to produce three to five times more biomass, and biomass of higher quality, than either switchgrass or big bluestem. These findings will aid producers in selection of warm-season grass cultivars and in establishing stands to produce biomass either for forage or for biofuel production.

Pellets from cotton gin trash are valuable fuel. There is need to find higher value uses for cotton gin trash. ARS engineers at the Cotton Production and Processing Research Unit, Cropping Systems Research Laboratory in Lubbock, Texas, conducted studies in cooperation with the USDA-ARS Environmental and Plant Dynamics Research Unit in Phoenix, Arizona, to evaluate performance and emissions of pellet stoves when burning cotton byproduct (COBY) pellets. COBY and COBY/Guayule pellets burned in commercial pellet stoves were found to have acceptable exhaust gas emissions while having heating values equivalent to other pellet stove fuels. Findings show that pelleted fuels made from cotton gin trash would be a use for a waste product, a reliable and economical source of heating fuel, and another income stream for cotton gins.