

Bank on biobutanol with wheat straw

By Jan Suszkiw, Agricultural Research Service – USDA

A modified method of producing biobutanol is in the works to make the fuel more competitive with ethanol as a clean-burning alternative to petrol and other fossil fuels.

Biobutanol offers several advantages over ethanol. It can be transported in existing pipelines, is less corrosive and less prone to water contamination, can be mixed with petrol or used alone in internal combustion engines, and packs more energy per litre than ethanol.

Up until the mid-20th century, biobutanol was produced from fermented sugars, such as glucose in corn or molasses. But low yields, coupled with high recovery costs and the increased availability of petroleum feedstocks after World War II, sidelined fermentation-based systems for biobutanol production.

Today, petrochemicals still reign supreme as the feedstock of choice for making butanol, a four-carbon alcohol that's mainly used as an industrial solvent. But jumps in the price of oil have rekindled in-

terest in tapping butanol as biobased fuel, notes Nasib Qureshi. He's a chemical engineer with the ARS National Center for Agricultural Utilization Research at Peoria, Illinois. Indeed, in June 2006, DuPont, Inc., of Wilmington, Delaware, and the British energy company BP announced joint plans to operate a United Kingdom-based production plant dedicated to producing biobutanol from sugarbeets.

Turning straw into biofuel gold

Up until late 2003, Nasib's own biobutanol studies dealt with new ways of fermenting glucose and other sugars from corn. But then he switched to wheat straw, drawn by its abundance and promise as a lower-cost alternative to glucose-based feedstocks.

"There's only so much you can do with sugar because of the competing uses for it," says Nasib, who's in the Peoria centre's Fermentation Biotechnology Research Unit. Wheat straw, by comparison, ... 12 ▷



Chemical engineer Nasib Qureshi adds enzymes into a bioreactor to simultaneously hydrolyse wheat straw into simple sugars and produce butanol.

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is typically left on crop fields after harvest. The team is also working with barley straw, corn stover, and switchgrass (a native pasture in North America).

Like other biobutanol processes, Nasib's approach calls on species of *Clostridium* bacteria to carry out the critical task of fermentation. But before the bacteria can perform such work, the straw must be pretreated and hydrolysed.

The hydrolysis step uses enzymes to break apart the straw's cellulose and hemicellulose components. This liberates the simple sugars within so that the bacteria can ferment them into three products – acetone, butanol, and ethanol. Butanol is produced in the greatest concentration, but all three are valuable chemicals.

Three steps in one

Normally, four preparatory steps (pretreatment, hydrolysis, fermentation, and recovery) are carried out separately and sequentially. But in his studies, Nasib – together with ARS Peoria colleagues Michael A. Cotta and Badal C. Saha – devised a way to combine three of the four steps.

After the wheat straw has been pretreated with dilute sulphuric acid or other chemicals, the material is fermented in a bioreactor containing three different types of commercial enzymes and a culture of *C. beijerinckii* P260, a strain Nasib obtained

from Professor David Jones of the University of Otago in Dunedin, New Zealand.

Nasib's approach allows the enzymes and bacteria to do their jobs simultaneously. As soon as the enzymes hydrolyse the straw and release its simple sugars, the bacteria set to work fermenting them. Throughout, a procedure known as 'gas stripping' (the fourth step) is used to remove the acetone, biobutanol, and ethanol as they're produced. Gas stripping also protects the bacteria by keeping biobutanol levels from reaching levels harmful to them.

In early trial runs, the method increased biobutanol productivity by twofold above traditional glucose-based fermentation – but the bacteria fermented the sugars faster than the sugars became available. So, to ensure optimum performance, Nasib found it necessary to feed small batches of additional sugar to the bioreactor.

Later studies showed that the adjustment, called 'fed-batch feeding', significantly increased biobutanol production. During a 22-day fed-batch operating period, a culture of *C. beijerinckii* P260 converted nearly 430 grams of sugar (glucose, xylose, arabinose, galactose, and mannose) into 192 combined grams of acetone, biobutanol, and ethanol.

If scaled up further, the process could yield 307 combined kilograms, or 375 litres, of acetone, biobutanol, and ethanol from one tonne of wheat straw. The P260 strain produces a specific ratio of the three

chemicals, but efforts are now under way at Peoria to develop genetically modified bacteria that will make only biobutanol.

Nasib says he is planning to scale up production levels in 2009. "Then, we'll look at the economics of using hydrolysed wheat straw to see how we're doing and move this process forward."

Making microbes do more work

Another USDA-ARS research team is developing a two-stage fermentation process for producing biobutanol from corn fibre – but any plant material containing cellulose and hemicellulose will do.

The first step involves pretreating the fibre with chemicals or hot water. The softened fibres are then fed to mixed cultures of different microbial species. By adjusting pH levels and other environmental factors, the microbes are coaxed to convert the fibre's sugars into butyrate, a butanol precursor.

The enzymes usually used to convert pretreated biomass to fermentable sugars are very expensive. But the mixture of microbes used in this case to make butyrate produce their own enzymes. This means huge potential cost savings.

Unwelcome microbes pose a contaminant problem in traditional fermentation systems, but this mixed microbial culture will be more resistant to contaminants.

Nasib Qureshi is in the Fermentation Biotechnology Research Unit, USDA-ARS National Center for Agricultural Utilization Research, Peoria. Ph: (309) 681 6318.



Technician John Michael Henderson combines dilute sulfuric acid with ground wheat straw as a pretreatment. The wheat straw is then autoclaved and transferred to a bioreactor for butanol production. (PHOTO: Peggy Greb)



Biochemical engineer Bruce Dien (left) loads reactor vessels with corn fibre while microbiologist Loren Iten lowers samples into a sand bath heater to pretreat for fermentation. (PHOTO: Peggy Greb)