

## **BESC** scores a first with isobutanol directly from cellulose

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In the quest for inexpensive biofuels, cellulose proved no match for a bioprocessing strategy and genetically engineered microbe developed by researchers at the Department of Energy's BioEnergy Science Center.

Using consolidated bioprocessing, a team led by James Liao of the University of California at Los Angeles for the first time produced isobutanol directly from cellulose. The team's work, published online in <u>Applied and Environmental Microbiology</u>, represents across-the-board savings in processing costs and time, plus isobutanol is a higher grade of alcohol than <u>ethanol</u>.

"Unlike ethanol, isobutanol can be blended at any ratio with <u>gasoline</u> and should eliminate the need for dedicated infrastructure in tanks or vehicles," said Liao, chancellor's professor and vice chair of Chemical and Biomolecular Engineering at the UCLA Henry Samueli School of Engineering and Applied Science. "Plus, it may be possible to use isobutanol directly in current engines without modification."

Compared to ethanol, higher alcohols such as isobutanol are better candidates for gasoline replacement because they have an <u>energy density</u>, octane value and Reid vapor pressure – a measurement of volatility – that is much closer to gasoline, Liao said.

While cellulosic biomass like corn stover and switchgrass is abundant and cheap, it is much more difficult to utilize than corn and sugar cane. This is due in large part because of recalcitrance, or a plant's natural



defenses to being chemically dismantled.

Adding to the complexity is the fact biofuel production that involves several steps – pretreatment, enzyme treatment and fermentation – is more costly than a method that combines biomass utilization and the fermentation of sugars to biofuel into a single process.

To make the conversion possible, Liao and postdoctoral researcher Wendy Higashide of UCLA and Yongchao Li and Yunfeng Yang of Oak Ridge National Laboratory had to develop a strain of Clostridium celluloyticum, a native cellulose-degrading microbe, that could synthesize isobutanol directly from cellulose.

"This work is based on our earlier work at UCLA in building a synthetic pathway for isobutanol production," Liao said.

While some Clostridium species produce butanol, these organisms typically do not digest cellulose directly. Other Clostridium species digest <u>cellulose</u> but do not produce butanol. None produce isobutanol, an isomer of butanol.

"In nature, no microorganisms have been identified that possess all of the characteristics necessary for the ideal consolidated bioprocessing strain, so we knew we had to genetically engineer a strain for this purpose," Li said.

While there were many possible microbial candidates, the research team ultimately chose Clostridium cellulolyticum, which was originally isolated from decayed grass. The researchers noted that their strategy exploits the host's natural cellulolytic activity and the amino acid biosynthetic pathway and diverts its intermediates to produce higher alcohol than ethanol.



The researchers also noted that Clostridium cellulolyticum has been genetically engineered to improve ethanol production, and this has led to additional more detailed research. Clostridium cellulolyticum has a sequenced genome available via DOE's Joint Genome Institute. This proof of concept research sets the stage for studies that will likely involve genetic manipulation of other consolidated bioprocessing microorganisms.

**More information:** The paper is titled "Metabolic Engineering of Clostridium Cellulolyticum for Isobutanol Production from Cellulose," and is available online at <u>aem.asm.org/</u>

## Provided by Oak Ridge National Laboratory

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