Biofuel breakthrough: scientists use GMO yeast to produce fuel

Scientists have discovered two new ways of using genetically modified yeast to produce biofuel more efficiently than previously possible. The two methods are described in two new studies recently published in *Science*. The studies can make biofuel production much faster and cheaper and reduce the need for fossil fuels, says Professor Jens Nielsen, Chalmers University of Technology in Gothenburg and the Novo Nordisk Foundation Center for Biosustainability at Technical University of Denmark, who is behind one of the studies.

"The combination of the two studies are important and interesting. One can imagine that this combination can lead to major advances in ethanol fuel production," says Nielsen.

Danish scientist: "it's a major breakthrough"

Professor Claus Felby from the University of Copenhagen's Department of Geosciences and Natural Resource Management, studies the use of natural resources -- using ethanol for biofuel production being one of his areas of expertise. He was not involved in the new study, but has read the papers and is very impressed -- especially by the Danish study.

"The first thing I thought was: "Wow!" The Danish study is really a major breakthrough because scientists have succeeded in something that people have struggled with for a great number of years."

Yeast is the most used microorganism in the industry, explains Felby. "If yeast can operate at higher temperatures it can be important for the pharmaceutical industry, chemical industry, and fuel industry. It's a revolution."

New technique increase ethanol production by 80 percent

The scientists behind the second study are American scientists from Massachusetts Institute of Technology (MIT). They also believe that ethanol production will be drastically improved with the two new studies.

In the study the scientists made yeast more tolerant to the ethanol that the yeast cells produce themselves.

This means that the yeast doesn't die when the ethanol reaches a certain concentration.

By increasing the yeasts cells tolerance, the American scientists increases the production of biofuels by 80
The yeast tolerance to alcohol is probably the single biggest problem in the production of ethanol today. The problem can be solved with these study results,” writes Professor of Chemical Engineering at MIT, Gregory Stephanopoulos, in an email to ScienceNordic.

Stephanopoulos is behind the second study published in Science. [9]

How to make ethanol

Ethanol is produced in large tanks. The process generates heat, which means that the tanks must be cooled down, otherwise the yeast dies. Cooling the tanks is expensive and also, the added enzymes actually work better at high temperatures around 45-50 degrees, but the yeast simply stops working around 35 degrees.

Also, the yeast cells do not tolerate the ethanol they produce themselves. When the concentration of alcohol becomes high enough the yeast cells degrade and the ethanol production decreases.

Both problems are solved in the new studies. Here, the Danish/Swedish scientists have modified the yeast cell genomes so that they can tolerate temperatures up to 40 degrees instead of only 35 degrees.

“It’s a big step in the right direction that we can make yeast that can operate at higher temperatures,” says Nielsen.

Heat tolerant yeast

In the Danish/Swedish study, Nielsen and his colleagues used nature's own mechanisms to make the yeast more heat tolerant.

They exposed yeast cells to 40 degrees for three months. What happened was that the yeast itself developed ways to beat the heat.

Nielsen explains that the survival and production rate of the yeast cells was low in the start of the experiment. Slowly the cells began to develop mutations that made them able to cope with higher temperatures.

At the end of the three months the scientists had developed seven yeast strains that had evolved to grow twice as fast at 40 degrees than normal yeast.

"This is great progress. It means we can raise the temperature significantly”, says Nielsen. "It will make the enzyme decomposition of starch into sugar much quicker and therefore the entire production will be faster."

One mutation was sufficient

After the scientists had isolated the seven yeast strains, they sequenced the whole genomes in order to find out which mutations had made the cells more heat tolerant.

They found that the yeast strains each had between 20 and 30 mutations in different parts of the genome. The results also showed that all seven yeast strains had mutations in the same gene.

Actually, they discovered that a single mutation was enough to make the yeast cells heat tolerant. This was a big surprise to the scientists.
"It's quite unique that we found that a single mutation is enough to change something so dramatic. It's probably also the reason why our study is so interesting. It's much easier to transfer a single mutation to other yeast cells than if we had found out that it required a large number of mutations in different parts of the genome to obtain heat tolerance,” says Nielsen.

**Mutation change the cell membrane**

The scientists also found out why the mutation had made the yeast more heat tolerant.

The mutation is in a gene which is involved in production of sterols in the yeast cells. Sterols are an important class of organic molecules. Among others, cholesterol is a sterol.

Normal yeast creates the sterol ergosterol -- with the mutation made the yeast produced fecosterol instead. The sterols become part of the cell membrane. It makes a big difference, which sterols are incorporated into the fat layer in the cell membrane.

Sterols influence how liquid the fatty cell membrane is at different temperatures. Fecosterols create a more firm membrane at higher temperatures than ergosteroler.

“At 40 degrees ergosterolerne makes the cell membrane too liquid, this effects the well-being of the yeast and the ethanol production,” says Nielsen.

**More energetic yeast cells**

In the American study, the scientist were originally trying to manipulate the yeast cells genes. A project they eventually had to give up on.

Instead, they began to study whether it would change anything to alter the chemical environment around the yeast cells.

Here, the scientists found that it made a major difference to the production of ethanol when they added a lot of potassium chloride and also made the yeast cells environment more alkaline by adding potassium hydroxide.

They found that the yeast cells could suddenly produce more ethanol as they were no longer being poisoned by their own self-produced ethanol.

"By changing the concentration of potassium inside and outside the cells, the cells gets more energy to counteract the unfavorable ethanol environment and the production continues," writes Stephanopoulos. "The discovery will have a direct impact on the commercial processes involved in the production of ethanol from high sugar concentrations."

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Read the original article in Danish on Videnskab.dk [10]

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Most biofuel is produced from maize. Scientists have recently discovered how to produce biofuel from maize faster and cheaper. (Photo: <a href="http://www.shutterstock.com/dl2_lim.mhtml?id=164005082&size=small_jpg&src=xWc3NTMhLwjmL_1xz1-4" target="_blank">Shutterstock</a>) [11]
Fact box

Biofuel is traditionally divided into first and second generation.

First-generation biofuel is made from the edible parts of crops such as maize.

Second generation of biofuel is produced from residues such as straw and stover.

Top-class biofuel from the depths of the forest. Turning seaweed to biofuel Barley gene could help feed the world Denmark aims to be rid of fossil fuels by 2050 Researchers develop best method yet for turning CO2 into fuel

Altered sterol composition renders yeast thermotolerant, Science (2014), DOI: 10.1126/science.1258137

Engineering alcohol tolerance in yeast, Science (2014), DOI: 10.1126/science.1247859

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