Scientist shows DNA detective work with paper-eating bacteria that 'glide'

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The eco-friendly fuel ethanol is usually made from grain, but the U.S. Department of Energy (DOE) would like to find other renewable materials that will be cost-effective alternatives, such as paper pulp, sawdust, straw and grain hulls.

A UWM professor recently helped DOE do just that by analyzing the DNA of a bacterium that can break down cellulose, the major structural component of plants that is also found in forestry by-products (including paper) and waste feedstocks.

Mark McBride, a professor of biological sciences, worked with DOE's Joint Genome Institute and scientists at Los Alamos National Laboratory to examine the genes of Cytophaga hutchinsonii that are responsible for the organism's ability to digest cellulose – the first step in the carbohydrate's conversion into ethanol.

Sequencing the genome of C. hutchinsonii provides what McBride calls a "parts list" for the microbe, allowing scientists to explore how bacteria use these parts to build and run their key functions – some of which have potential uses in bioenergy.

The genome has revealed surprises, he says.

"Microorganisms typically require two kinds of enzymes to efficiently break down cellulose," he says. "One type cuts the long carbohydrate molecule through the middle, while another chews small pieces from the ends."

Not so with C. hutchinsonii. Although it efficiently digests cellulose, in DNA analysis it appears to be lacking one of the usual enzymes, suggesting that it may use either a novel strategy or novel enzymes.

The information McBride reports could help DOE devise mixtures of microorganisms or enzymes that will more efficiently convert cellulose into glucose, and finally into ethanol.

McBride's interest in C. hutchinsonii goes beyond its possible value in bioenergy.

What really intrigues him is that it's a "gliding bacterium," able to crawl rapidly over surfaces by an unknown mechanism, which is the main subject of McBride's research with another glider called Flavobacterium johnsoniae. The two microbes are not closely related.

"You are more closely related to a fruit fly than these two organisms are to each other," he says.

However, from analysis of genes from the two bacteria, McBride suspects that they use the same basic machinery to move.

And there may be another connection. F. johnsoniae doesn't eat cellulose, but it is able to digest a similar carbohydrate polymer, chitin. Like cellulose, chitin, which is found in the hard shells of lobsters and insects, is also difficult to break down.

McBride hypothesizes that digestion of cellulose and chitin may also be linked to cell movement, or motility.

"Loss of motility results in loss of ability to digest chitin," he says. "This suggests that motility and digestion of some carbohydrate polymers may be connected in both gliding microbes."

McBride and his students have used F. johnsoniae to study the motility of gliding bacteria for more than a decade. They cloned "mutants" of F. johnsoniae that are unable to move, and then attempted to "repair" them by inserting certain pieces of DNA.

In this way, they have uncovered nearly all the genetic components that propel the cells. It has been a long process. A decade ago, his lab had found one protein involved. He now knows of 24, and he doesn't expect to find many more.

Until recently, McBride was not able to image the bacteria closely enough to see the structures involved in movement. Instead, he bonded latex spheres to the surface of F. johnsoniae cells and observed that they moved in all directions around the cell's perimeter.

"The cell wall appears to have a series of moving conveyer belts," he says.

He also has learned that some of the motility proteins ("parts") act at the surface of the cell, and he thinks some are involved in forming nearly invisible filaments around the perimeter of the cell.

These filaments were recently imaged in collaboration with Sriram Subramaniam and Jun Liu at the National Institutes of Health by cryo-electron tomography.

"The filaments may be the cell's 'tires,' and there are different kinds," McBride says. "They are designed to help the organism move over a variety of surfaces, like an all-terrain vehicle."

Besides providing movement, McBride says the filaments also may be needed to move the cellulose and chitin molecules to certain sites where they can be digested or taken into the cell.

McBride hopes the complete genome for C. hutchinsonii will yield other clues to the interconnections among gliding bacteria. He is now collaborating with DOE to sequence the entire genome of F. johnsoniae, which will allow a full comparison of the genes of the two microorganisms.

Source: University of Wisconsin - Milwaukee

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