Engineering algae to make the 'wonder material' nanocellulose for biofuels and more

April 7 2013

Genes from the family of bacteria that produce vinegar, Kombucha tea and nata de coco have become stars in a project—which scientists today said has reached an advanced stage—that would turn algae into solar-powered factories for producing the "wonder material" nanocellulose. Their report on advances in getting those genes to produce fully functional nanocellulose was part of the 245th National Meeting & Exposition of the American Chemical Society (ACS).

"If we can complete the final steps, we will have accomplished one of the most important potential agricultural transformations ever," said R. Malcolm Brown, Jr., Ph.D. "We will have plants that produce nanocellulose abundantly and inexpensively. It can become the raw material for sustainable production of biofuels and many other products. While producing nanocellulose, the algae will absorb carbon dioxide, the main greenhouse gas linked to global warming."

Brown, who has pioneered research in the field for more than 40 years, spoke at the First International Symposium on Nanocellulose, part of the ACS meeting. Abstracts of the presentations appear below.

Cellulose is the most abundant organic polymer on Earth, a material, like plastics, consisting of molecules linked together into long chains. Cellulose makes up tree trunks and branches, corn stalks and cotton fibers, and it is the main component of paper and cardboard. People eat cellulose in "dietary fiber," the indigestible material in fruits and vegetables. Cows, horses and termites can digest the cellulose in grass, hay and wood.

Most cellulose consists of wood fibers and cell wall remains. Very few living organisms can actually synthesize and secrete cellulose in its native nanostructure form of microfibrils. At this level, nanometer-scale fibrils are very hydrophilic and look like jelly. A nanometer is one-millionth the thickness of a U.S. dime. Nevertheless, cellulose shares the unique properties of other nanometer-sized materials—properties much different from large quantities of the same material. Nanocellulose-based materials can be stronger than steel and stiffer than Kevlar. Great strength, light weight and other advantages has fostered interest in using it in everything from lightweight armor and ballistic glass to wound dressings and scaffolds for growing replacement organs for transplantation.

In the 1800s, French scientist Louis Pasteur first discovered that vinegar-making bacteria make "a sort of moist skin, swollen, gelatinous and slippery"—a "skin" now known as bacterial nanocellulose. Nanocellulose made by bacteria has advantages, including ease of production and high purity that fostered the kind of scientific excitement reflected in the first international symposium on the topic, Brown pointed out.

Brown recalled that in 2001, a discovery by David Nobles, Ph.D., a member of the research team at the University of Texas at Austin, refocused their research on nanocellulose, but with a different microbe. Nobles established that several kinds of blue-green algae, which are mainly photosynthetic bacteria much like the vinegar-making bacteria in basic structure; however, these blue-green algae, or cyanobacteria, as they are called, can produce nanocellulose. One of the largest problems with cyanobacterial nanocellulose is that it is not made in abundant amounts in nature. If it could be scaled up, Brown describes this as "one of the most important discoveries in plant biology."

Since the 1970s, Brown and colleagues began focusing on *Acetobacter xylinum* (*A. xylinum*), a bacterium that secretes nanocellulose directly into the culture medium, and using it as an ideal model for future research. Other members of the *Acetobacter* family find commercial uses in producing vinegar and other products. In the 1980s and 1990s, Brown's team sequenced the first nanocellulose genes from *A. xylinum*. They also pinpointed the genes involved in polymerizing nanocellulose (linking its molecules together into long chains) and in crystallizing (giving nanocellulose the final touches needed for it to remain stable and functional).

But Brown also recognized drawbacks in using *A. xylinum* or other bacteria engineered with those genes to make commercial amounts of nanocellulose. Bacteria, for instance, would need a high-purity broth of food and other nutrients to grow in the huge industrial fermentation tanks that make everything from vinegar and yogurt to insulin and other medicines.

Those drawbacks shifted their focus on engineering the *A. xylinum* nanocellulose genes into Nobles' blue-green algae. Brown explained that algae have multiple advantages for producing nanocellulose. Cyanobacteria, for instance, make their own nutrients from sunlight and water, and remove carbon dioxide from the atmosphere while doing so. Cyanobacteria also have the potential to release nanocellulose into their surroundings, much like *A. xylinum*, making it easier to harvest.

In his report at the ACS meeting, Brown described how his team already has genetically engineered the cyanobacteria to produce one form of nanocellulose, the long-chain, or polymer, form of the material. And they are moving ahead with the next step, engineering the cyanobacteria to synthesize a more complete form of nanocellulose, one that is a polymer with a crystalline architecture. He also said that operations are being scaled up, with research moving from laboratory-sized tests to larger outdoor facilities.

Brown expressly pointed out that one of the major barriers to commercializing nanocellulose fuels involves national policy and politics, rather than science. Biofuels, he said, will face a difficult time for decades into the future in competing with the less-expensive natural gas now available with hydraulic fracturing, or "fracking." In the long run, the United States will need sustainable biofuels, he said, citing the importance of national energy policies that foster parallel development and commercialization of biofuels.

More information: Abstract

What we have learned about cellulose biosynthesis from Acetobacter xylinum (Gluconacetobacter xylinus): A brief history and future prospects
R. Malcolm Brown, Jr. The University of Texas at Austin

This presentation will briefly cover the long history of research on Acetobacter xylinum, with particular attention to the research that has yielded new ideas and data for cellulose biosynthesis in general. Also to be covered is research on cellulose biosynthesis among certain oxygen-evolving photosynthetic bacteria known as cyanobacteria. The Brown lab has achieved a functional transfer of Acetobacter xylinum cellulose synthase genes into cyanobacteria. The major goal is to secure equivalent biosynthetic capacity in Acetobacter xylinum. If this can be achieved, then a new global source of cellulose biosynthesis will be available for commercial exploitation. Revealed for the first time will be a short video of time lapse sequences showing cellulose biosynthesis in Acetobacter. The talk will conclude with a summary of the major existing problems for developing Acetobacter commercially, and what could be done to rectify these long-standing problems. Attendees will be encouraged to find avenues for open communication so that microbial nanocellulose

production can come to fruition as one of the most useful natural products in polymer chemistry.

Provided by American Chemical Society