A corny turn for biofuels from switchgrass

November 18 2011

This image shows the overxpression of the Cg1 gene in switchgrass (left) compared to Wild-type of switchgrass of the same age and grown under the same conditions. Credit: Photo courtesy of USDA/ARS

Many experts believe that advanced biofuels made from cellulosic biomass are the most promising alternative to petroleum-based liquid fuels for a renewable, clean, green, domestic source of transportation energy. Nature, however, does not make it easy. Unlike the starch sugars in grains, the complex polysaccharides in the cellulose of plant cell walls are locked within a tough woody material called lignin. For advanced biofuels to be economically competitive, scientists must find inexpensive ways to release these polysaccharides from their bindings and reduce them to fermentable sugars that can be synthesized into fuels.

An important step towards achieving this goal has been taken by researchers with the U.S. Department of Energy (DOE)'s Joint BioEnergy Institute (JBEI), a DOE Bioenergy Research Center led by the Lawrence Berkeley National Laboratory (Berkeley Lab).

A team of JBEI researchers, working with researchers at the U.S. Department of Agriculture's Agricultural Research Service (ARS), has demonstrated that introducing a maize (corn) gene into switchgrass, a highly touted potential feedstock for advanced biofuels, more than doubles (250 percent) the amount of starch in the plant's cell walls and makes it much easier to extract polysaccharides and convert them into fermentable sugars. The gene, a variant of the maize gene known as Corngrass1 (Cg1), holds the switchgrass in the juvenile phase of development, preventing it from advancing to the adult phase.

"We show that Cg1 switchgrass biomass is easier for enzymes to break down and also releases more glucose during saccharification," says Blake Simmons, a chemical engineer who heads JBEI's Deconstruction Division and was one of the principal investigators for this research. "Cg1 switchgrass contains decreased amounts of lignin and increased levels of glucose and other sugars compared with wild switchgrass, which enhances the plant's potential as a feedstock for advanced biofuels."

The results of this research are described in a paper published in the *Proceedings of the National Academy of Sciences* (*PNAS*) titled "Overexpression of the maize Corngrass1 microRNA prevents flowering, improves digestibility, and increases starch content of switchgrass."

Lignocellulosic biomass is the most abundant organic material on earth. Studies have consistently shown that biofuels derived from lignocellulosic biomass could be produced in the United States in a sustainable fashion and could replace today's gasoline, diesel and jet fuels on a gallon-for-gallon basis. Unlike ethanol made from grains, such fuels could be used in today's engines and infrastructures and would be carbon-neutral, meaning the use of these fuels would not exacerbate global climate change. Among potential crop feedstocks for advanced biofuels, switchgrass offers a number of advantages. As a perennial grass that is both salt- and drought-tolerant, switchgrass can flourish on marginal cropland, does not compete with food crops, and requires little fertilization. A key to its use in biofuels is making it more digestible to fermentation microbes.

"The original Cg1 was isolated in maize about 80 years ago. We cloned the gene in 2007 and engineered it into other plants, including switchgrass, so that these plants would replicate what was found in maize," says George Chuck, lead author of the *PNAS* paper and a plant molecular geneticist who holds joint appointments at the Plant Gene Expression Center with ARS and the University of California (UC) Berkeley. "The natural function of Cg1 is to hold pants in the juvenile phase of development for a short time to induce more branching. Our Cg1 variant is special because it is always turned on, which means the plants always think they are juveniles."

George Chuck and Sarah Hake, plant molecular geneticists at the Plant Gene Expression Center, Albany, California, introduced a variant corngrass gene into switchgrass. Credit: Photo courtesy of USDA/ARS

Chuck and his colleague Sarah Hake, another co-author of the PNAS paper and director of the Plant Gene Expression Center, proposed that since juvenile biomass is less lignified, it should be easier to break down into fermentable sugars. Also, since juvenile plants don't make seed, more starch should be available for making biofuels. To test this hypothesis, they collaborated with Simmons and his colleagues at JBEI to determine the impact of introducing the Cg1 gene into switchgrass.

In addition to reducing the lignin and boosting the amount of starch in the switchgrass, the introduction and overexpression of the maize Cg1 gene also prevented the switchgrass from flowering even after more than two years of growth, an unexpected but advantageous result.

"The lack of flowering limits the risk of the genetically modified switchgrass from spreading genes into the wild population," says Chuck.

The results of this research offer a promising new approach for the improvement of dedicated bioenergy crops, but there are questions to be answered. For example, the Cg1 switchgrass biomass still required a pre-treatment to efficiently liberate fermentable sugars.

"The alteration of the switchgrass does allow us to use less energy in our pre-treatments to achieve high sugar yields as compared to the energy required to convert the wild type plants," Simmons says. "The results of this research set the stage for an expanded suite of pretreatment and saccharification approaches at JBEI and elsewhere that will be used to generate hydrolysates for characterization and fuel production."

Another question to be answered pertains to the mechanism by which Cg1 is able to keep switchgrass and other plants in the juvenile phase.

"We know that Cg1 is controlling an entire family of transcription factor genes," Chuck says, "but we have no idea how these genes function in the context of plant aging. It will probably take a few years to figure this out."

Provided by Lawrence Berkeley National Laboratory