Cellulosic biofuels

Building a collaborative culture for research...

n its 2006 report entitled Breaking the Biological Barriers to Cellulosic Ethanol, the US Department of Energy (DOE) articulated a vision that "A robust fusion of the agricultural, industrial biotechnology, and energy industries can create a new strategic national capability for energy independence", and observed that "Cellulosic biomass is an attractive energy feedstock because it is an abundant, domestic, renewable source that can be converted to liquid transportation fuels". Despite the vicissitudes of financial crisis, leading to unmet US Renewable Fuel Standard schedules, the fundamentals supporting this vision remain:

- The price of cellulosic biomass at \$60/dry ton has the same cost per unit energy (\$4/GJ) as oil at \$23/barrel;
- There is increasing evidence that large-scale biofuel production can be reconciled with feeding humanity and preserving the environment. E.g. In the International Energy Agency's BLUE Map Scenario, for example, biomass provides 23% of global primary energy in 2050 with cellulosic biomass the largest contributor;
- Cellulosic biofuels will likely be a significant part of the energy supply picture for the indefinite future if key obstacles can be overcome, and achieving a sustainable transportation sector is much more likely with biofuels than without them. In the most aggressive scenarios for electrification of light-duty vehicles, liquid fuels still provide more than 50% of total US transportation energy.

The main barrier to realising the benefits

of cellulosic biofuels is the availability of cost-effective technology to overcome the difficulty of converting lignocellulosic feedstock to reactive intermediates, which we have defined as the 'recalcitrance' of cellulosic biomass. Removal of this barrier will enable production of a broad range of biofuels and other commodities of which cellulosic ethanol will be the first to be commercialised.

Lignocellulosic biomass can be converted to fuels by a variety of routes, including: thermochemical processes involving reactive intermediates other than sugars (e.g. synthesis gas, pyrolysis oil), fermentative processes that overcome recalcitrance primarily by non-biological means (e.g. acid hydrolysis, phosphoric acid swelling, ionic liquid pre-treatments), and fermentative processes that overcome recalcitrance with key biotechnology-driven advances. The BioEnergy Science Center (BESC) was formed in 2007 to adopt the third approach – particularly the development of less recalcitrant plants and/or microbes that more effectively convert lignocellulose – as the most promising approach to overcome the recalcitrance barrier. BESC is funded at a level of \$25m a year by the US's Office of Biological and Environmental Research, recently renewed for an additional five years.

Building a collaborative environment

In the formation of BESC, we gathered a team of world-class scientists from 18 different institutions in academia, industry and the DOE national laboratories with the unifying theme of understanding and overcoming recalcitrance. Our management and operational structures and processes ensured that the research teams focused their efforts on the core thematic path by supporting strong, focused integrative science, effectively managing our milestones, and revising our priorities in response to changing demands, needs and developments. The core strength of BESC has been its ability to manage and integrate the distinct sets of expertise and staffing distributed across the institutions to yield a synergy that has informed our understanding of recalcitrance beyond our initial expectations. Indeed, we believe we have created a culture where the primacy of collaborative and integrative approaches, expressed in both the research and the management, is key to the overall goals of the project.

The combination of researchers brings some distinct advantages:

- The BESC academic partners' primary function remains basic research on the fundamental science and applied strategies required to achieve our goals, as well as educating and training the future bioenergy research workforce;
- The DOE national laboratories bring a cross-disciplinary team approach; strong capabilities in technology development and applications, particularly in the physical and computational sciences; unique infrastructure, computing platforms, and other advanced equipment; and excellent management and environmental, safety, health, and quality (ESH&Q) systems;

 The research-based companies, which are key links in the biofuels supply chain, ensure that the BESC programme is well-informed by commercial needs, can validate our scientific findings in a commercial setting, and can accelerate the translation of BESC discoveries into real-world impact. Several of the industrial partners also bring unique research platforms and capabilities to the centre.

Outcomes to date

Since its initiation, BESC has made substantial progress in understanding and reducing plant cell wall recalcitrance and has generated biomass feedstock with improved conversion. Transgenic and selected natural variant Populus and switchgrass lines with modified biomass properties have been identified and studied for wall composition and digestibility using unique recalcitrance phenotypic screens. Over 1,000 overexpression and knock-down constructs representing either Populus and switchgrass recalcitrance target genes have been generated. To date, over 40 reduced-recalcitrance feedstock lines have been identified with increased sugar yields, and three or more have been moved into field trials. BESC has demonstrated the consolidated bioprocessing (CBP) potential of microbes from several genera. A key success has been the development of genetic tools for CBP thermophiles, including *Clostridium* thermocellum and Caldicellulosiruptor spp. These tools are now being used to manipulate metabolic pathways to increase ethanol yield and titre. We have extended the CBP proof of concept by genetically engineering a microbe to make isobutanol from cellulose. Additionally, in yeast, the ability to add cellulolytic capabilities to an ethanologen was demonstrated. We have demonstrated that CBP can be combined with reduced-recalcitrance plants to further increase conversion.

Measures of collaboration

The collaborative culture and value system

that has led to this level of productivity is best reflected in the project's publication statistics (available on the BESC website). Of the 503 articles published in peerreviewed scientific literature since the commencement of the project in 2007, 70% of the articles involve collaboration between scientists within or across the partnership; this includes teams within partner institutions, across institutions and indeed reflects collaborations between BESC scientists and others in the bioenergy science research.

Of those same 503 papers, 171 involve collaborations with international scientists from 24 countries outside the US. While most of these are informal, scientist-to-scientist collaborations, the BESC project also has a number of formal memoranda of understanding (MOUs) with international groups such as Scion, a Crown Research Institute in New Zealand focused on forestry research; the University of British Columbia; and Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP) in Brazil. We have research agreements with personnel at the Weizmann Institute in Tel Aviv, Israel, and at the Stellenbosch University near Cape Town.

Key principles

There are a number of key organisational principles that are designed to support this collaborative culture:

- Use of independent oversight and review via an external science advisory board and a board of directors, designed to provide expert scientific programme management and industrial advice and to approve annual performance goals, projects, and budget plans;
- Development of integrated project management across multiple institutions with designated centre-level practices and operations management, providing clear leadership for ESH&Q and effective business management using proven practices

and tools for performance monitoring of research schedule and budget;

- There is a shared vision and common goals across the centre. Recalcitrance began as an operationally-defined phenotype. As a team, we are redefining recalcitrance as phenomena in terms of pathways and interactions in both cell wall formation and in bioconversion based on chemistry, structure and biochemistry;
- Communication and collaborations are encouraged by regular conferences and an annual retreat. Openness is supported by broad non-disclosure and material transfer agreements to simplify data and sample sharing.

In the first five years of BESC, one of our most important outputs, in addition to a compelling body of published work, has been our scientists. As over 100 young undergraduates and postgraduates have passed through the project to positions in academia and industry, they have been trained with the skills needed to operate in (and indeed lead) interdisciplinary and inter-institutional teams and have been endowed with a highly tuned valuesystem for domestic and international collaborative science. Perhaps this will be our greatest legacy to the scientific community.



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