

Critical National Need Idea Title:

Improving Cellulosic Biomass Crops through Genomics Technology

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Summary

The development of alternatives to fossil fuels as an energy source is an urgent national and global priority. Cellulosic biomass has the potential to contribute to meeting the demand for liquid fuel, but land-use requirements and process inefficiencies represent hurdles for large-scale deployment of biomass-to-biofuel technologies. Genomic information gathered from across the biosphere, including potential energy crops, will be vital for breaking those barriers and improving the prospects of significant cellulosic biofuel production.

A. Maps to Administration Guidance – Breaking the Barriers to Sustainable Bioenergy

In November 2006, the President's Council of Advisors on Science and Technology (PCAST) published a report called "The Energy Imperative" which discussed the potential for advanced energy technology to enhance U.S. energy security while protecting the environment. The report concluded that technology innovation offers the prospect of a cleaner, more efficient, and more economically sustainable future. Since the publication of this report, The Energy Independence and Security Act (or EISA), signed into law by President Bush in December 2007, established a number of provisions that will contribute to cleaner and more efficient energy use. It established a renewable fuel standard (RFS) with mandatory annual production volumes of biofuels, quadrupling the RFS of the Energy Policy Act of 2005. For the first time, EISA establishes separate mandatory targets for cellulosic ethanol, biodiesel, and other advanced biofuels. Today, biofuels production is approaching 10 billion gallons per year (mostly corn ethanol, but also including 0.5 billion gallons per year of biodiesel). The RFS requires the use of 36 billion gallons of biofuels per year by 2022, of which only 15 billion gallons can be corn ethanol. The legislation also requires that biofuels reduce life-cycle GHG emissions by amounts ranging from a 20 percent reduction for corn ethanol to a 60 percent reduction for cellulosic biofuels when compared to gasoline.

As discussed in the 2006 President's Council of Advisors on Science and Technology (PCAST) report, one of Department of Energy (DOE)'s primary goals in biofuels is to make cellulosic ethanol (or other advanced biofuels) practical and cost-competitive by 2012. A more challenging goal will be to enable cost-competitive biofuels at the large volumes specified by the new RFS. Thus, funding from the TIP program to make this goal a real possibility maps well to Administration guidance.

B. Justification for Government Attention - National Energy Security

While the United States consumes about a quarter of the global energy supply – roughly the same as its share of global economic output – the growth of energy demand in developing nations in recent years has far outstripped that of developed nations. The Energy Information Administration (EIA) of the DOE projects that in the period 2005–2030, energy consumption in developed nations (i.e., members of the Organisation for Economic Co-operation and Development, OECD) will grow by 19 percent, while developing nations will increase their energy consumption by 85 percent (EIA-DOE 2008c). Altogether, world energy consumption is expected to grow by 50 percent over that 25 year period. Similarly, carbon emissions are projected to grow by 15 percent in

developed nations, 85 percent in developing nations, and 50 percent overall during that same period.

Projected global trends in petroleum consumption are particularly instructive. The EIA expects that OECD nations will increase consumption of petroleum (and other fossil fuel liquids) from 49.3 million barrels per day (or mbd) in 2005 to 53.3 mbd in 2030, which represents only an 8 percent increase. Non-OECD nations, on the other hand, are projected to increase their petroleum consumption by 73 percent, from 34.3 to 59.3 mbd, over the same period. Because crude oil is traded on a global market, the rapid growth of demand in developing nations could impact gasoline prices as much as if the increased demand was local. This suggests that technology innovation, which can alter consumption patterns on a global scale, is vital to addressing the energy challenges faced by the United States.

The geopolitical challenges associated with crude oil supplies – highlighted this summer by oil prices rising to well over \$100 per barrel – emphasize the need to displace oil with alternative fuels. This is especially important in the transportation sector, which accounts for over two-thirds of overall domestic oil consumption. An EIA paper (Kendell 1998) has outlined measures of energy security that can be used to interpret a degree of U.S. vulnerability as a result of oil imports. These include measures such as oil imports as a proportion of U.S. oil consumption, the net cost of oil imports, calculated U.S. oil intensity (consumption per dollar of gross domestic product), the Persian Gulf share of the world's oil exports, and global excess oil production capacity. For the past two decades, most of these measures have indicated that U.S. vulnerability is increasing. However, in the United States two of these indicators have improved since 2005: the volume of imported oil has actually decreased by approximately 10 percent from 12.5 mbd to 11.3 mbd, and oil intensity has decreased more rapidly than at any time in the past two decades. Advanced energy technologies can enable the United States and the world to meet the energy demands of a growing global economy while making progress toward environmental and other societal goals.

Despite the increasing use of biofuels such as biodiesel and sugar- or starch-based ethanol, evidence suggests that transportation fuels based on lignocellulosic biomass represent the most scalable alternative fuel source. Lignocellulosic biomass in the form of plant materials (for example, dedicated energy crops) offers the possibility of a renewable, geographically distributed and greenhouse-gas-favourable source of sugars that can be converted to ethanol and other liquid fuels. Calculations of the productivity of lignocellulosic feedstocks, in part based on their ability to grow on marginal agricultural land, indicates that they can probably have a large impact on transportation needs without significantly compromising the land needed for food crop production. A transition to advanced biofuels produced from cellulosic feedstocks (i.e., not from grains) could substantially reduce U.S. oil consumption while having little or no impact on the food supply. In addition, biomass can become renewable feedstocks for other industries too. The manufacture of chemicals, plastics, hydraulic fluids, pharmaceuticals, and other industrial products accounts for fully 25 percent of domestic oil consumption (EIA/DOE 2008b). Biomass feedstocks can supply the raw materials for processes and products that replace petroleum. A large market for these products could accelerate private investment in biorefineries and contribute significantly to rural jobs and income.

C. Essentials for TIP Funding

Of all sustainable resources, only biomass can be transformed into organic fuels and chemicals that have inherent convenience, cost, and efficiency advantages and integrate so well into our transportation infrastructure. Lignocellulosic biomass, in particular, provides a vast, inexpensive resource, and development of low cost biomass processes will be invaluable to sustainably meeting such needs. The efficiency of the biological cellulosic ethanol production process depends strongly on feedstock characteristics and composition, the pretreatment applied, enzymes used, and fermentation technologies employed. Key barriers to large-scale biofuels production include insufficient feedstock availability, unfavorable environmental impacts, and the cost and feasibility of technologies at each step in the production process (e.g., collection, pretreatment, hydrolysis, fermentation, and distillation for biochemical processes, and gasification, cleanup, and synthesis for thermal processes). Partially because of the historically low demand for biologically based transportation fuels, each step in this process is in the early stages of optimization for efficiency and throughput. The two major obstacles are first, the crops from which biomass are currently derived have not been domesticated for this particular purpose; and second, the present methods for saccharification and fermentation are inefficient and expensive. Genomics information from feedstock plants can potentially provide breakthroughs in many of the above areas through their improvements.

Energy crops. Until recently, minimal effort has been directed towards optimizing potential energy crops for the generation of transportation fuels. This is in stark contrast to the agronomic development of food crops, which have been domesticated for thousands of years to maximize productivity. Some of the most rapid increases have occurred in the past 40 years, both from advances in agronomic practices and, importantly, from the application of modern genetics and most recently genomics. The optimization of bioenergy crops as feedstocks for transportation fuels is in its infancy, genomic information and resources will be essential for accelerating their domestication. As we can retrospectively view the features that made certain wild plants desirable for domestication thousands of years ago to become today's food crops, we are now prospectively defining criteria to choose plants with potential to serve as dedicated bioenergy crops in the future. These include cell wall composition, growth rate, suitability for growth in different geographical regions, and resource-use efficiencies.

Many of the traits targeted for optimization in potential cellulosic energy crops are those that would improve growth on marginal agricultural lands, to minimize competition with food crops over land use, and to make production of low per-unit value biomass more economical. Perennial crops are essential to bringing marginal lands into sustainable biomass production (Wagoner, 1990; Scheinost, 2001; Cox et al., 2002), maximizing ecosystem productivity (Field, 2001) and minimizing losses of topsoil (Pimentel et al., 1995) (Gantzer et al., 1990), water, and nutrients (Randall and Mulla, 2001). Nonetheless, there has been only limited research into the genetics of perenniality, and in only a few crops (Cox et al., 2002). In particular, the allocation of photosynthate to vegetative versus reproductive organs remains complex and controversial. Better understanding of the genetic basis of perenniality and associated re-allocation of photosynthate would accelerate progress toward deterministic breeding of dedicated bioenergy crops suitable for utilization on marginal lands.

Energy crops and biorefining. Because biomass at ~ \$55/dry ton is competitive with petroleum at ~\$17/barrel on an equivalent energy basis, the challenge is to reduce processing costs: in other words, to overcome the recalcitrance of biomass. In the United States, the Department of Energy (DOE) is the major funding source for the research and development of biofuels. DOE has announced plans to invest up to \$385 million, representing a total \$1.2 billion including private cost-share, for as many as six commercial-scale, cellulosic biorefinery projects over four years. DOE also plans to invest \$114 million in four pilot-scale biorefinery projects, which will be about 10 percent of the size of a commercial-scale facility, to test innovative processes for converting a variety of cellulosic feedstocks – sugarcane residues, wood product residues, other agricultural wastes, and dedicated energy crops – into ethanol. However, many of these projects are focused on bioprocessing rather than on energy crops and the interplay among biomass features and pretreatment and enzymatic hydrolysis has not been systematically studied or capitalized on, even though data has shown significant differences in sugar release with type of feedstock and this knowledge could help overcome a major barrier to low cost biofuels.

Genomics studies of dedicated bioenergy crops can help break the barriers. Tropical grasses are among the most efficient biomass accumulators known, thanks to 'C4' photosynthesis, a complex combination of biochemical and morphological specializations that confer efficient carbon assimilation at high temperatures. One model C4 crop is sorghum. Its small genome (~740 Mb) and low level of gene duplication makes Sorghum an attractive model for Saccharinae functional genomics, and motivated its complete sequencing under the US Department of Energy Joint Genome Institute (JGI) 'Community Sequencing Program' (Paterson et al. 2008). The Saccharinae clade of cereals is distinctive in that it includes three leading candidate lignocellulosic biofuels crops, Sorghum (currently the #2 US biofuels crop), Saccharum (sugarcane and its relatives, currently the worlds leading biofuels crop), and Miscanthus [argued to be among the highest-yielding biomass crops with nearly twice the yield of switchgrass (Heaton et al. 2004)].

Many potential dedicated energy crops, especially Miscanthus, lack genomic tools required for comparative and bioinformatic approaches to enhance fundamental knowledge of genome structure, function, and organization. Miscanthus has had only very limited genetic analysis (Atienza et al. 2002), and based largely on semi-arbitrary (AFLP) markers that do not provide for deductions about the correspondence of its chromosomes to those of sorghum. A clear need in the employment of innovative strategies for breeding of this leading candidate for plant feedstock production is more information about the degree to which existing genomic information translates to Miscanthus, as well as fundamental information about Miscanthus transmission genetics that is important to implementing de novo genomic approaches in its improvement.

Genomics studies can potentially reveal many genes that are potential targets for the improvement of energy crops. These include many genes involved in cellulose and hemicellulose synthesis as well as those believed to influence various morphological growth characteristics such as height, branch number and stem thickness. In addition to homology-based strategies, other genome-enabled strategies for identifying domestication candidate genes are being used. These include quantitative trait analysis of natural variation and genome-wide mutagenesis coupled with phenotypic screens for traits such as recalcitrance to sugar release, acid digestibility and general cell wall composition. The availability of high-throughput transgenesis in several plant systems

will facilitate functional studies to determine the in vivo activities of the large number of domestication candidate genes. Using these strategies, genes affecting features such as plant height, stem elongation and trunk radial growth, drought tolerance, and cell wall stability are but a few of the features that are likely to be identified as targets for domestication in a fraction of the time required to carry out similar studies unaided by the plant genomes and genomic approaches.

Reference:

- Cox TS, Bender M, Picone C, Van Tassel DL, Holland JB, Brummer EC, Zoeller BE, Paterson AH, Jackson W** (2002) Breeding perennial grain crops. *Critical Reviews in Plant Sciences* **21**: 59-91
- Energy Information Administration, United States Department of Energy (EIA-DOE)**. 2008a. Annual energy outlook 2008. Washington, DC: EIA. DOE/EIA-0383(2008). <http://www.eia.doe.gov/oiaf/aeo/>
- _____. 2008b. Annual energy review 2008. Washington, DC: EIA. DOE/EIA-0384(2008). <http://www.eia.doe.gov/emeu/aer/contents.html>
- _____. 2008c. International energy outlook 2008 – highlights. Washington, DC: EIA. DOE/EIA-0484(2008). <http://www.eia.doe.gov/oiaf/ieo/>
- _____. 2008d. Monthly energy review October 2008. Washington, DC: EIA. DOE/EIA-0035(2008/10). <http://www.eia.doe.gov/mer/>
- _____. 2008e. Short-Term Energy Outlook, October 2008. Washington, DC: EIA. DOE/EIA (2008/10). <http://www.eia.doe.gov/steo>
- Field CB** (2001) Sharing the garden. *Science* **294**: 2490-2491
- Farrell, A., et al.** 2006. "Ethanol can contribute to energy and environmental goals." *Science* 311 (5760): 506-8, <http://www.sciencemag.org/cgi/content/short/311/5760/506>.
- Glauber, J.** 2008. Congressional testimony before the Joint Economic Committee of the United States Congress, May 1, 2008. [http://www.house.gov/jec/news/2008/May/Testimony percent20USDA percent20Drpercent20 percent20Glauber-Food percent20Prices.pdf](http://www.house.gov/jec/news/2008/May/Testimony%20USDA%20percent20Drpercent20percent20Glauber-Food%20Prices.pdf)
- Gantzer CJ, Anderson SH, Thompson AL, Brown JR** (1990) Estimating soil erosion after 100 years of cropping on Sanborn Field. . *Journal of Soil and Water Conservation* **45**: 641-644
- Kendell, J.** 1998. "Measures of Oil Dependence," in *Issues in Midterm Analysis and Forecasting 1998*. Washington,
- Pimentel D, Harvey C, Resosudarmo P, Sinclair K, Kurz D, McNair M, Crist S, Shpritz L, Fitton L, Saffouri R, Blair R** (1995) Environmental and economic costs of soil erosion and conservation benefits. *Science* **267**: 1117-1123
- Randall GW, Mulla D** (2001) Nitrate nitrogen in surface waters as influenced by climatic conditions and agricultural practices. *Journal of Environmental Quality* **30**: 337-344
- Scheinost P** (2001) Perennial wheat: a sustainable cropping system for the Pacific Northwest. *American Journal of Alternative Agriculture* **16**: 147-151
- Wagoner P** (1990) Perennial Grain Development - Past Efforts and Potential for the Future. *Critical Reviews in Plant Sciences* **9**: 381-408