Joint BioEnergy Institute

Biological & Environmental Research Advisory Committee

29 November 2007

Washington, DC
JBEI at a glance

• Start-up company approach
  – Highly focused research agenda
  – Single operation and facility

• Four Science and Technology Divisions
  – Feedstocks
  – Deconstruction
  – Fuels Synthesis
  – Cross-cutting Technologies

• Six Partners
  – Three DOE National Laboratories
  – Two Universities
  – One Foundation

• Industry Partnership Program
  – Underpin growth of the biofuels industry
  – Ensure technology transfer to biofuels industry
Lignocellulosic Biomass to Fuels

Plants → Lignocellulose → Enzymes → Monomers → Microbes

JBEI Joint Bioenergy Institute
Some Key Challenges in Converting Lignocellulosic Biomass to Fuels

Challenges

- Cellulose and hemicellulose are occluded by lignin
- Lignin is recalcitrant to depolymerization
- Inhibitors released from biomass
Some Key Challenges in Converting Lignocellulosic Biomass to Fuels

Plants → Enzymes → Microbes

Lignocellulose → Monomers

Lignin is recalcitrant to depolymerization
Some Key Challenges in Converting Lignocellulosic Biomass to Fuels

Challenges

- Lignocellulose is difficult to depolymerize
- Pretreatment methods form inhibitory by-products
Some Key Challenges in Converting Lignocellulosic Biomass to Fuels

Plants → Enzymes → Microbes

Lignocellulose → Monomers

Pretreatment methods form inhibitors
Some Key Challenges in Converting Lignocellulosic Biomass to Fuels

Challenges

- Existing biofuels are not optimal
- Organisms can only utilize a fraction of the monomers
- Inhibitors released from biomass limit fuels production
Some Key Challenges in Converting Lignocellulosic Biomass to Fuels

Plants → Enzymes → Microbes

Lignocellulose → Monomers

Existing biofuels are not optimal
JBEI: an interlocking approach with three scientific divisions
JBEI: an interlocking approach supported by a Technologies Division
JBEI: an interlocking approach underpinned by Genomics:GTL
Feedstocks: Developing Bioenergy Crops

Challenges

- Cellulose and hemicellulose are occluded by lignin, making deconstruction difficult.
- Functional groups on hemicellulose can inhibit fermentation & are not efficiently converted to fuels.
- Lignin is recalcitrant to depolymerization.

Somerville et al. 2004, Science
Feedstocks: Developing Bioenergy Crops

**Approach**

- Understand & modify polysaccharide biosynthesis
  - Focus on hemicellulose
- Reduce feruloylation by engineering alternative pathway
- Modify lignin to aid deconstruction.
  - Introduction of cleavable linkages
- Switchgrass, rice and Arabidopsis as model plants
  - Switchgrass sequencing
Example: reduce the complexity of cell wall building blocks or change their composition

- Xylans contain acetate esters and grass xylans contain additional ferulic acid esters
- Acetate esters are problematic for deconstruction and subsequent fuels fermentations
- Ferulate & diferulate esters are crosslinked with lignin. This results in grass cell walls being difficult to enzymatically digest

Arabinoxylan, a type of xylan found in grasses

Objective: change composition & crosslinking
Example: change the monomers in lignin

- Systematic analysis of (lignin) cell wall oxidases
- Develop replacement strategies for lignin
- Apply advanced imaging technology to determine structure of plant cell walls

Objective: change the monomer composition of lignin
Deliverables

• Improved understanding of all cell wall synthesizing and modifying enzymes in rice and Arabidopsis

• Transgenic plants with optimized cell wall composition for deconstruction

• Translate genetic developments from model plant systems to proposed bioenergy crops
Deconstruction: providing a source of fermentable sugars

Challenges

• Lignocellulose is difficult to process due to:
  – low accessibility of crystalline cellulose fibers
  – presence of lignin “seal” & hemicellulose cross-links
  – small pore sizes in lignocellulose

• Acid pretreatment methods result in the formation of by-products that are inhibitory to subsequent biofuels fermentation and result in a loss of sugars
Deconstruction: providing a source of fermentable sugars

Approach

- Understand the chemical and structural changes resulting from current and new biomass pretreatment approaches
- Understand the fundamental interactions that govern lignocellulolytic enzymes
- Explore new microbial environments and employ directed evolution to produce more active and stable lignocellulolytic enzymes

Rain forest floor
Example: Improving enzyme performance

- Enzyme-substrate, enzyme complexes, and enzyme-product interactions are key components of enzyme performance.

- We will delineate the enzymatic mechanisms involved in lignocellulose depolymerization.

- Utilize directed evolution to optimize enzymes to improve performance characteristics and lower cost.

Objective: optimize enzyme structure and function.
Deconstruction: providing a source of fermentable sugars

Deliverables

• Optimal pretreatment methods for target biomass feedstocks
• Improved lignocellulolytic enzymes with enhanced activity and stability
• Understanding of how microbial communities degrade lignocellulose
• Cost-effective pretreatment & enzymatic depolymerization methods with minimal by-products and inhibitor formation
Recent Results: Ionic Liquid Pretreatment Studies of Switchgrass

- Raw switchgrass samples were processed to isolate different parts of the plant
- Intact bulk samples were then exposed to 1-ethyl-3-methylimidazolium (acetate salts) at 120°C
- Biomass deconstruction tracked as a function of temperature and time
- Confocal images taken with dual wavelength excitation (405 and 543 nm)

Initial

T = 120°C, t = 20 mins

T = 120°C, t = 50 mins
Biofuels Synthesis: ethanol and next generation biofuels

Challenges

• Existing biofuels
  – do not have the full fuel value of gasoline
  – require energy-intensive purification processes
  – are toxic at high concentrations
  – cannot be transported using traditional means

• Microorganisms convert only a limited number of precursors to fuels.

• Inhibitors resulting from the pretreatment process prevent growth and biofuel production
Biofuels Synthesis: ethanol and next generation biofuels

Approach

- Develop pathways for production of future biofuels
- Understand mechanisms of fuel toxicity and stress response
- Engineer organisms to produce & withstand high concentrations of biofuels
- Engineer organisms for consolidated bioprocessing (cellulase production with simultaneous fermentation of sugars to biofuels)
Example: Production of next-generation biofuels

- Large number of potential fuel molecules can be produced from central metabolic intermediates.
  - Alkanes
  - Alcohols
  - Esters
- Need to construct precursor biosynthetic pathways.
- Understand their impact on cell physiology.

Objective: Engineer pathways for fuels synthesis
Biofuels Synthesis: ethanol and next generation biofuels

Deliverables

• Organisms engineered to produce and withstand high concentrations of biofuels
• Organisms resistant to by-products formed during deconstruction
• Sequence and regulatory information for metabolic pathways producing biofuels
• Models of metabolic pathways for fuel synthesis and their mode of regulation
Technology: new tools for biofuels research

Challenges

• Few tools available for bioenergy/biomass research
• New high throughput biochemical and `omics approaches needed for all aspects of bioenergy research
• Advanced imaging techniques can be leveraged to characterize biomass and biomass deconstruction processes
Technology: new tools for biofuels research

Approach

• Provide technologies for scientific discovery

• Implement high-throughput off-the-shelf systems

• Automate, parallelize and miniaturize throughput-limiting procedures

• Develop new technologies for enzyme characterization
Technology: new tools for biofuels research
Example: Automation of Limiting Processes

- Cloning, protein expression and enzyme assays will be rate limiting without high-throughput technologies.
- JBEI will implement HT cloning and expression technologies and develop new microfluidic tools for HT enzyme assays.
Deliverables

- High-throughput microfluidics platforms for large scale analysis of plant and microbial enzyme activities
- Ligno- and glyco-arrays for rapid screening of enzymatic function
- ‘Omics pipelines for systems biology
- Integrated data capture, analysis and dissemination
- Parts, devices, chassis for synthetic biology
Interdependent research

- Cross-cutting technologies will aid the research in multiple divisions
- Research will be interdependent with discoveries in one area influencing the research in the other areas.
A single JBEI facility will foster research interactions.

Integrated operation ensures effectiveness, cost-efficiency, and unity.
JBEI Facility: EmeryStation East

61,000 rentable square feet on-floor
~43,000 assignable square feet
  • Environmentally friendly building
  • Access to adjacent 80 seat conference center
  • Shuttle services and 90 parking spaces
Accelerated start-up: Research at Partner Facilities

FY07 research starts at partner institutions

- LBNL, UCB
  - Berkeley West Biocenter
    - dedicated 12,000 sf lab, office space
- Sandia National Laboratories, California
- Lawrence Livermore National Lab
- Carnegie Institute of Science
- UC Davis
A single location
JBEI leverages key capabilities of partner institutions

Systems & Synthetic Biology

Plant Research
- UC Davis Genome Center & Plant Genomic Program
- Carnegie Institute of Stanford

Energy Research
- Helios
- National Combustion Research Facility

Imaging
- UC Berkeley Imaging
- National Center for Electron Microscopy

Nanoscience
- Molecular Foundry
- Center for Integrated Nanoscience

Computation
- Synthetic Biology Engin. Res. Center (SynBERC)
- DOE: Genomics VIMSS ESPP
- Joint Genome Institute

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JBEI Organization

Research organization designed to be decisive and nimble, modeled on technology “start–up”

- Bold
- Flexible
- Adaptable

Dedicated management for science, technical integration, and scalability
JBEI leverages the Bay Area biotech and high tech industry

Vibrant industries grow around intellectual centers
- Silicon Valley and Biotech Industry around UCB, UCSF, Stanford
- Bay Area and CA becoming centers for renewable energy

Benefits of JBEI location in Bay Area and CA
- Intellectual environment
- Recruiting
- Commercialization
Interactions with industry

- Equipment makers/suppliers
- Bioinformatics companies

Cross-cutting Technologies
- High Throughput Microsystems
- Omics
- Multi-scale Imaging
- Integrated Informatics

- Sunlight
- Plants
- Lignocellulose
- Microbes & Enzymes
- Monomers (sugars & aromatics)
- Synthetic Biology
- Fuels Synthesis

- Feedstocks
- Deconstruction

- Crop genetics companies
- Biofuels companies
- Automobile companies

- Biomass suppliers
Commercializing JBEI’s Products

JBEI’s Technology Transfer Program will:

- Efficiently commercialize innovative biofuels technologies
- Promote dialogue among researchers, industry, and VCs
- Provide opportunities for industry to collaborate with JBEI
- Complement and further JBEI’s biofuels research

Mechanisms:

- Industry Advisory Committee
  - Companies from key sectors: feedstocks, enzymes, fuels production, biotechnology, genetics, chemistry
  - Provide feedback on JBEI research from an industry perspective
- Central management of IP and industry interface
- Industry Partnership Program
- Central data repository of all JBEI IP and IP agreements
- Industrial scientist sabbaticals at JBEI
Impacts

• Elucidate & modify plant cell wall structure and synthesis
• Efficient, cost-effective routes for deconstruction of lignocellulose
• Engineered organisms for scalable production of ethanol and next generation biofuels
• Enabling and integrating technologies for bioenergy research
• Integrated science & technology to transform the U.S. biofuels industry
Accelerated start-up: research and operations personnel

Supplemental Funding vs. Original Plan for FY'08

Q1 Q2 Q3 Q4